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Chilling Injury, Respiration, and Sugar Changes in Sweet Potatoes Stored at Low Temperature

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Abstract. **External chilling injury symptoms, primarily surface pitting followed by secondary fungal decay, developed in six sweet potato [*Ipomoea batatas* (L.) Lam] cultivars after an exposure to 7°C for 2 weeks or more followed by storage at 15.6°. Internal chilling injury symptoms, primarily darkening of the cambium and vascular bundles, were observed in noncured 'Whitestar' and 'Rojo Blanco' roots after exposure to 7° for 3 weeks and in 'Centennial' after exposure to 7° for 4 weeks followed by storage at 15.6°. 'Jewel' was the cultivar most tolerant to low temperature. Chilling injury and respiration rate were greater with increasing lengths of exposure to 7° and were greater in noncured than cured roots. Enhanced sucrose and total soluble sugar content occurred at 7° compared to 15.6°. The primary sugar responsible for low-temperature sweetening was sucrose, but there was considerable variation among cultivars in the extent of low-temperature sweetening and specific sugar changes.**

Sweet potatoes are susceptible to chilling injury when stored at temperatures <12°C (8, 19). Exposure to low temperatures may occur in storage warehouses during the fall and winter months, in transit, and at retail outlets. More than 42% of 2525 households surveyed said they put sweet potatoes in the refrigerator after purchase (7).

Reported symptoms of chilling injury include root shriveling, surface pitting, and fungal decay (5, 6, 8) and internal tissue browning (8, 9, 16). Storage of roots at chilling injury-inducing temperatures adversely affects the culinary quality of the baked product by development of hardcore (2, 3).

The severity of chilling injury depends on the temperature and length of exposure below 12°C (10). The effects of holding sweet potatoes at chilling temperatures are often unnoticed until a return to higher temperatures. Cultivar differences in susceptibility to chilling injury were reported (3, 8, 16), and noncured roots were found to be more susceptible than cured roots (3, 10).

Little information exists on physiological changes induced by low temperature in currently grown cultivars. The degree of susceptibility to chilling injury among these cultivars and between cured and noncured roots has not been established. Therefore, the objective of this study was to determine the effect of length of low temperature exposure (7°C) on chilling injury symptom development, respiration rate, and soluble sugar content in cured and noncured roots of currently used sweet potato cultivars.

Materials and Methods

Two high dry matter, white-flesh sweet potato cultivars ('Whitestar' and 'Rojo Blanco') and four orange-flesh cultivars with medium to low dry matter ('Centennial', 'Jewel', 'Jasper', and 'Travis') were grown on a silt loam soil at the LSU Hill Farm in Baton Rouge, La. following cultural practices recommended for commercial production (12). All roots were hand-harvested in mid-October with the aid of a turn-plow, prior to any adverse cool or wet weather.

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Disease-free U.S. #1 grade roots were washed immediately following harvest and randomly assigned to the following treatments: cured (10 days at 32°C; 90% RH), plus 1, 2, 3, or 4 weeks at 7°; noncured plus 1, 2, 3, or 4 weeks at 7°; cured or noncured plus 1 week at 7° followed by 4 weeks at 15.6°, 2 weeks at 7° followed by 3 weeks at 15.6°, 3 weeks at 7° followed by 2 weeks at 15.6°, 4 weeks at 7° followed by 1 week at 15.6°. Additional cured and noncured roots were stored for 4 and 5 weeks at 15.6°. The noncured roots were put directly into 7° or 15.6° storage after harvest, resulting in a 10-day age difference upon analysis compared to the cured roots. Relative humidity was 90% in the 7° and 15.6° storage rooms.

One hundred roots from each treatment were rated for amount of surface pitting (the primary external chilling injury symptom) and fungal decay following exposure to the prior treatments. A root classified as having external chilling injury had ≈10% or greater pitting and/or fungal decay over the root surface. No distinction was made between degrees of symptom severity >10%. Roots having 10% or more surface decay and/or pitting would not be marketable. Most of the external chilling symptoms necessary to result in an unmarketable root developed during the first week of 15.6° storage following removal from 7°.

After visual examination, each root was cut longitudinally in half, held at 21°C for 3 min, and then observed for the presence of internal darkening.

Respiration rates of eight random disease-free roots per cultivar were determined at 15.6°C for 22 days following removal from 7°. Respiration rates also were determined at 15.6° for cured and noncured roots previously stored at 15.6° for 4 weeks. Two roots per cultivar, replicated four times, were placed in 4-liter sealed glass jars, and CO_2 in the head space was measured after 1 hr by gas chromatography. Displaced air inside the jar due to root volume was taken into account in all respiration calculations. Results were expressed on a root fresh weight basis.

Six random disease-free 'Whitestar', 'Centennial', 'Jewel', and 'Travis' roots from each of four replications were taken from each treatment and analyzed for soluble sugar content and alcohol-insoluble solids (AIS). Unpeeled roots were cut in half longitudinally and uniformly grated over the entire surface to a depth of about 3 mm. Ten grams of flesh tissue from the combined six roots per replication was extracted in 80% ethanol and analyzed for sugar content by HPLC as previously described

Table 1. Amount of external chilling injury in sweet potato roots after different lengths of exposure to 7°C followed by 15.6° storage.

	Percentage of roots with external chilling injury[z]					
	Cultivar					
Treatment	Jewel	Whitestar	Rojo Blanco	Travis	Centennial	Jasper
		Cured[y]				
1 week 7° + 4 weeks 15.6°	0[x]	0	0	0	0	0
2 weeks 7° + 3 weeks 15.6°	0	6	5	6	11	11
3 weeks 7° + 2 weeks 15.6°	6	14	15	23	25	25
4 weeks 7° + 1 week 15.6°	7	25	27	38	37	36
		Noncured[w]				
1 week 7° + 4 weeks 15.6°	0	0	0	0	0	0
2 weeks 7° + 3 weeks 15.6°	11	12	11	11	15	12
3 weeks 7° + 2 weeks 15.6°	48	52	44	66	72	72
4 weeks 7° + 1 week 15.6°	85	100	81	100	100	100

[z]A root classified as having external chilling injury contained 10% or greater pitting and decay of the entire root surface area.
[y]Cured = 10 days at 32°C; 90% RH.
[x]Each treatment consisted of 100 total roots.
[w]Noncured = roots stored at 7° on the day of harvest.

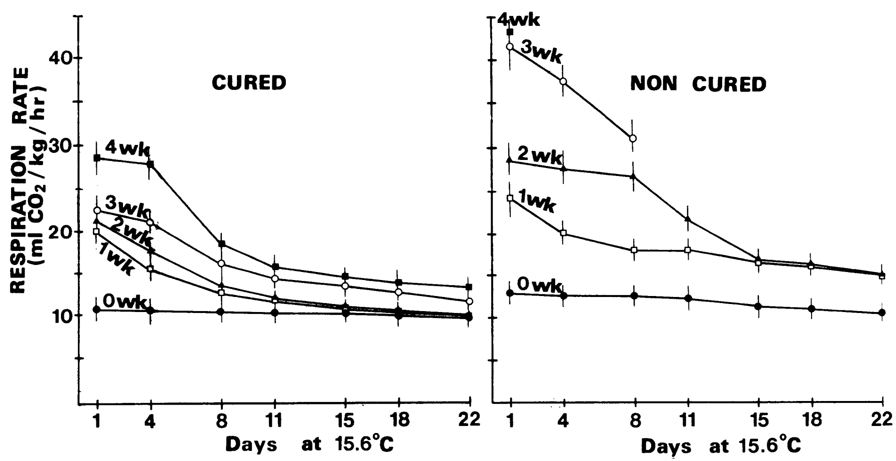

Fig. 1. Respiration rate of cured and noncured 'Centennial' sweet potato roots at 15.6°C following previous exposure to 7° for 0, 1, 2, 3, or 4 weeks. Only disease-free roots were analyzed. Due to external fungal development, measurements were not made after 1 day for noncured roots exposed to 7° for 4 weeks; or after 8 days for noncured roots exposed to 7° for 3 weeks. Vertical bars represent SE of the mean.

(14). AIS content was determined as previously described (15) by weighing the ethanol insoluble residue after 24 hr of vacuum drying at 35°C.

Standard error of the mean was calculated for each time interval in the respiration study. Significant differences ($P < 0.05$) between storage treatments in the amount of each sugar were determined by analysis of variance and means were separated according to Fisher's least significant difference test (18).

Results and Discussion

External chilling injury. No pitting or decay was observed in cured or noncured roots exposed to 7°C for 1 week followed by 4 weeks at 15.6° (Table 1). Increasing the length of 7° exposure to 2 weeks followed by 3 weeks at 15.6° resulted in external chilling injury in 11% to 15% of the noncured roots, and lesser amounts in cured roots. No chilling injury symptoms developed in cured 'Jewel' roots after 2 weeks at 7° followed by 3 weeks at 15.6°. After 3 weeks at 7° followed by 2 weeks at 15.6°, 44% to 72% of the noncured roots developed external chilling injury symptoms, while only about one-third the amount of chilling injury developed in cured roots under equivalent conditions. Severe pitting, which coalesced to sunken water-soaked lesions followed by secondary fungal decay, developed on almost all noncured roots exposed to 7° for 4 weeks followed by 1 week at 15.6°. Equivalent storage of cured roots of all cultivars, except 'Jewel', resulted in the development of external chilling injury symptoms in 25% to 38% of the total roots. Only 7% of cured 'Jewel' roots had external chilling symptoms after 4 weeks at 7° followed by 1 week at 15.6°.

All sweet potato cultivars were susceptible to pitting and eventual fungal decay, but differences existed among cultivars. Among cured treatments, 'Jewel' roots displayed the fewest visible symptoms of chilling. 'Travis', 'Centennial', and 'Jasper' roots responded in a similar manner and displayed the most external chilling symptoms. Cured roots were less susceptible than noncured roots, presumably due to the beneficial effects of suberization and wound periderm formation during the curing process (1). Lutz reported cured 'Porto Rico' roots decayed less than noncured roots when exposed to 4.5°C (10).

Commercially unacceptable levels of chilling injury developed in noncured roots after only 2 weeks at 7°, which underscores the susceptibility of sweet potatoes to chilling injury and

Table 2. Sugar content of cured and noncured 'Centennial' sweet potato roots after different lengths of exposure to 7°C and after return to 15.6°.

Treatment	Sugar content (g/100 g fresh wt)			
	Sucrose	Glucose	Fructose	Total soluble sugar
	Cured roots			
After curing[z]	3.68	0.11	0.10	3.89
1 week 7°	4.09	0.17	0.15	4.41
1 week 7° + 4 weeks 15.6°	4.16	0.30	0.29	4.75
2 weeks 7°	4.86	0.13	0.14	5.13
2 weeks 7° + 3 weeks 15.6°	4.75	0.30	0.29	5.34
3 weeks 7°	5.63	0.18	0.17	5.98
3 weeks 7° + 2 weeks 15.6°	5.24	0.23	0.20	5.67
4 weeks 7°	5.81	0.29	0.26	6.36
4 weeks 7° + 1 week 15.6°	7.20	0.22	0.19	7.61
4 weeks 15.6°	3.85	0.24	0.24	4.33
	Noncured roots			
After harvest	1.92	0.02	0.02	1.96
1 week 7°	3.06	0.07	0.06	3.22
1 week 7° + 4 weeks 15.6°	3.92	0.32	0.29	4.53
2 weeks 7°	4.18	0.16	0.14	4.48
2 weeks 7° + 3 weeks 15.6°	4.85	0.40	0.35	5.60
3 weeks 7°	5.36	0.25	0.23	5.84
3 weeks 7° + 2 weeks 15.6°	6.27	0.45	0.40	7.12
4 weeks 7°	6.31	0.32	0.29	6.92
4 weeks 7° + 1 week 15.6°	6.84	0.39	0.37	7.60
Overall LSD (0.05)	0.37	0.10	0.08	0.43

[z]Curing = 10 days at 32°C; 90% RH.

the importance of curing followed by storage at a nonchilling temperature. No pattern of pitting or decay was observed over the root surfaces that developed external chilling symptoms. Symptoms occurred randomly over the entire surface. Pitting and decay, however, were more severe in the areas of the root that had skin abrasions caused during harvesting and transit from the field. The amount of pitting intensified, and most of the secondary fungal decay developed during the first week at 15.6° following removal from 7°. A number of different fungi can cause decay of chilling injured sweet potatoes (6).

Internal chilling injury. Internal darkening, most pronounced near the cambium and vascular bundles, was the primary internal chilling injury symptom. Darkening of the central pith tissue was observed in severely injured roots. The typical brown to black discoloration was consistent with past observations (6, 16). Histochemical tests indicated phenolics are located in the periderm, cambium, latex of laticifers, and vascular bundles, but not in parenchymatous cells containing many starch granules (17).

No internal darkening was observed in any cultivar after 5 weeks at 15.6°C or after 1 or 2 weeks of exposure to 7° followed by 15.6° storage (data not shown). Some internal darkening was observed in noncured 'Rojo Blanco' and 'Whitestar' roots exposed to 7° for 3 weeks followed by 2 weeks at 15.6°. No other cultivar exhibited internal darkening after 3 weeks at 7°. Severe internal darkening occurred in noncured 'Rojo Blanco' and 'Whitestar' roots exposed to 7° for 4 weeks followed by 1 week at 15.6°. Noncured 'Centennial' roots also exhibited moderate, but objectionable, amounts of internal darkening under the same conditions. Noncured roots from the other three orange-flesh cultivars did not discolor. A previous study also established differences in internal darkening among cultivars held at 5°, with 'Jewel' being the least susceptible to darkening (16). No internal darkening occurred in cured roots of any of the four orange-flesh cultivars exposed to 7° for up to 4 weeks. A slight amoung of darkening appeared in cured 'Rojo Blanco' and 'Whitestar' roots after 4 weeks at 7° followed by 1 week at 15.6°.

The lack of internal darkening in certain cultivars and treatments may have been due to a decline in chlorogenic acid upon transfer to 15.6°C (9) or to the lack of increase in chlorogenic acid during low-temperature storage (11). The mechanism of the beneficial effect of curing in preventing or reducing internal discoloration is not known but may be related to reduced disruption of membrane integrity or to decreased phenolic acid synthesis.

Respiration rate. The influence of storage time at 7°C on the subsequent pattern of respiration rate at 15.6° was similar for all cultivars. Therefore, only the results for 'Centennial' are shown (Fig. 1).

Respiration rate was the highest during the first 24 hr after removal from 7°C, followed by a decline during the next 22 days in all treatments. The rate of decline in respiration was greater during the first 7 days compared to the last 7 days of 15.6° storage. Noncured roots had a higher respiration rate than cured roots from the same length of 7° exposure, and respiration rate increased with increasing lengths of 7° exposure. After 15 days of 15.6° storage, no difference in respiration existed between roots exposed to 7° for 1 or 2 weeks, but the rates for noncured, chilled roots were higher than the noncured, nonchilled controls. Severe fungal decay developed shortly after return to 15.6° on noncured roots exposed to 7° for 3 or 4 weeks. Cured roots exposed to 7° for 4 weeks had a higher respiration rate than those exposed to 7° for 3 weeks, and the respiration rates from both treatments were higher than in nonchilled, cured controls over the entire 22-day period at 15.6°. Cured roots

Table 3. Sugar content of cured and noncured 'Jewel' sweet potato roots after different lengths of exposure to 7°C and after return to 15.6°.

Treatment	Sugar content (g/100 g fresh weight)			
	Sucrose	Glucose	Fructose	Total soluble sugar
	Cured roots			
After curing	3.22	0.70	0.44	4.36
1 week 7°	3.73	0.81	0.54	5.08
1 week 7° + 4 weeks 15.6°	3.77	1.08	0.80	5.65
2 weeks 7°	4.97	0.76	0.69	6.42
2 weeks 7° + 3 weeks 15.6°	4.78	0.85	0.68	6.31
3 weeks 7°	5.59	0.59	0.55	6.73
3 weeks 7° + 2 weeks 15.6°	6.22	0.76	0.64	7.62
4 weeks 7°	6.45	0.70	0.57	7.72
4 weeks 7° + 1 week 15.6°	6.73	0.72	0.55	8.00
4 weeks 15.6°	3.09	1.08	0.66	4.83
	Noncured roots			
After harvest	1.98	0.12	0.11	2.21
1 week 7°	2.95	0.39	0.47	3.81
1 week 7° + 4 weeks 15.6°	3.20	1.09	1.15	5.44
2 weeks 7°	4.71	0.37	0.46	5.54
2 weeks 7° + 3 weeks 15.6°	5.09	0.97	1.00	7.06
3 weeks 7°	6.20	0.46	0.53	7.19
3 weeks 7° + 2 weeks 15.6°	6.68	0.90	1.00	8.58
4 weeks 7°	7.27	0.47	0.53	8.27
4 weeks 7° + 1 week 15.6°	7.20	0.87	0.98	9.05
Overall LSD (0.05)	0.40	0.10	0.10	0.51

[z]Curing = 10 days at 32°C; 90% RH.

Table 4. Sugar content of cured and noncured 'Travis' sweet potato roots after different lengths of exposure to 7°C and after return to 15.6°.

Treatment	Sugar content (g/100 g fresh weight)			
	Sucrose	Glucose	Fructose	Total soluble sugar
	Cured roots			
After curing	1.61	1.70	1.11	4.42
1 week 7°	2.09	1.43	1.02	4.54
1 week 7° + 4 weeks 15.6°	2.62	1.61	1.06	5.29
2 weeks 7°	3.05	1.29	0.86	5.20
2 weeks 7° + 3 weeks 15.6°	3.21	1.51	1.07	5.79
3 weeks 7°	3.51	1.22	0.90	5.63
3 weeks 7° + 2 weeks 15.6°	3.52	1.55	1.12	6.19
4 weeks 7°	4.30	1.22	0.86	6.38
4 weeks 7° + 1 week 15.6°	4.68	1.50	1.06	7.24
4 weeks 15.6°	2.34	1.75	1.12	5.22
	Noncured roots			
After harvest	1.15	0.86	0.85	2.86
1 week 7°	1.26	1.29	1.44	3.99
1 week 7° + 4 weeks 15.6°	2.15	1.68	1.72	5.55
2 weeks 7°	2.26	1.48	1.53	5.27
2 weeks 7° + 3 weeks 15.6°	2.84	1.74	1.77	6.35
3 weeks 7°	3.23	1.51	1.60	6.34
3 weeks 7° + 2 weeks 15.6°	3.84	1.70	1.74	7.28
4 weeks 7°	3.55	1.53	1.59	6.67
4 weeks 7° + 1 week 15.6°	4.11	1.81	1.94	7.86
Overall LSD (0.05)	0.43	0.10	0.11	0.51

[z]Curing = 10 days at 32°C; 90% RH.

exposed to 7° for 4 weeks had a lower respiration rate than noncured roots exposed to 7° for 2 weeks.

Increased respiration rate in sweet potatoes is a common result of chilling injury (5, 8). Accelerated respiration occurs before visible symptoms. A simple method for indexing the severity of chilling injury could be based on respiration rate, because of

Table 5. Sugar content of cured and noncured 'Whitestar' sweet potato roots after different lengths of exposure to 7°C and after return to 15.6°.

Treatment	Sugar content (g/100 g fresh weight) Sucrose	Glucose	Fructose	Total soluble sugar
	Cured roots			
After curing	3.89	0.22	0.21	4.32
1 week 7°	4.12	0.36	0.27	4.75
1 week 7° + 4 weeks 15.6°	3.09	0.54	0.48	4.11
2 weeks 7°	5.27	0.34	0.33	5.94
2 weeks 7° + 3 weeks 15.6°	4.30	0.33	0.34	4.97
3 weeks 7°	5.82	0.37	0.36	6.55
3 weeks 7° + 2 weeks 15.6°	5.62	0.30	0.30	6.22
4 weeks 7°	7.28	0.52	0.54	8.34
4 weeks 7° + 1 week 15.6°	7.26	0.47	0.51	8.24
4 weeks 15.6°	2.69	0.43	0.35	3.47
	Noncured roots			
After harvest	1.36	0.05	0.04	1.45
1 week 7°	2.79	0.22	0.27	3.28
1 week 7° + 4 weeks 15.6°	3.19	0.36	0.42	3.97
2 weeks 7°	4.66	0.36	0.46	5.48
2 weeks 7° + 3 weeks 15.6°	4.82	0.30	0.37	5.49
3 weeks 7°	6.41	0.38	0.40	7.19
3 weeks 7° + 2 weeks 15.6°	6.42	0.35	0.41	7.18
4 weeks 7°	7.72	0.43	0.52	8.67
4 weeks 7° + 1 week 15.6°	7.68	0.52	0.66	8.86
Overall LSD (0.05)	0.38	0.07	0.09	0.56

[z]Curing = 10 days at 32°C; 90% RH.

the strong association between respiration and eventual expression of chilling injury. Further studies are needed to identify the intracellular factors responsible for more chilling injury resistance in cured than noncured roots, as indexed by lower respiration rates.

Sugar content. Sucrose, the major soluble sugar in raw 'Centennial' (Table 2) and 'Jewel' (Table 3) roots, increased during the first 7 days at 7°C or 10 days at 32° (curing). No further significant increase in sucrose content occurred in cured roots held at 15.6° for 4 weeks, but storage at 7° enhanced sucrose synthesis in both cultivars. Cured 'Jewel' roots contained twice the amount of sucrose after 4 weeks at 7° than 15.6°. Sucrose levels were maintained or increased during subsequent storage at 15.6° following transfer from 7°. Thus, sucrose-synthesizing enzymes were more active at 7° and were not adversely affected by return to 15.6°. Glucose and fructose content in 'Centennial' and 'Jewel' increased during the first 7 days at 7°, during curing, and after 4 weeks at 15.6°. Glucose and fructose content generally did not increase in 'Jewel' roots stored at 7° for longer than 1 week, but did significantly increase in noncured 'Centennial' roots with increasing length of 7° storage. Glucose and fructose levels in both cultivars generally increased during subsequent storage at 15.6° following transfer from 7°, which was especially evident in noncured roots. Total soluble sugar content in 'Jewel' increased with increasing length of 7° exposure, primarily due to the increase in sucrose content. Total soluble sugar content in noncured 'Jewel' roots increased during the 15.6° period following removal from 7°, primarily due to the increase in glucose and fructose content. The pattern and change in total soluble sugar content in 'Centennial' with temperature and storage duration paralleled the results for sucrose, since total soluble sugars (sum of glucose, fructose, and sucrose) were usually comprised of more than 90% sucrose.

Sucrose content in 'Travis' roots did not change during the first week at 7°C (Table 4) but did increase during curing. Sucrose increased during 4 weeks of storage, but to a lesser degree at 15.6° than 7°. Sucrose content continued to increase at 15.6° following removal from 7°, but generally only in noncured roots. Glucose and fructose content increased during the first week at 7° and after curing, but no further change occurred during 4 weeks at 15.6°. Storage for longer than 1 week at 7° resulted in a loss of glucose and fructose in cured 'Travis' roots, but a slight increase in noncured roots. Return of 7° stored roots to 15.6° resulted in an increase in glucose and fructose content. Total soluble sugar content increased in 'Travis' roots during the first week at 7°, during curing, and subsequent 4-week storage at 15.6°. The increase in total soluble sugar content during storage at 7° was primarily due to increased sucrose. The increase in total soluble sugar content which occurred at 15.6° following 7° storage was due to increased sucrose, glucose, and fructose.

Sucrose content in 'Whitestar' roots increased after 7 days at 7°C and during curing, but significantly decreased in cured roots during storage at 15.6° for 4 weeks (Table 5). Sucrose content increased during storage at 7° and was more than 2.5 times greater after 4 weeks at 7° than at 15.6°. No change in sucrose content occurred during the 15.6°-storage period after removal from 3 and 4 weeks at 7°, but shorter exposures to 7° resulted in a decrease in sucrose in cured roots but an increase in noncured roots. Glucose and fructose content increased during 7 days at 7°, during curing, and in cured roots during 4 weeks at 15.6°. Glucose and fructose contents generally increased with increasing length of 7° exposure. No further increase in these monosaccharides occurred at 15.6° following removal from 7°, except after 1 week of 7° exposure and in noncured roots after 4 weeks of 7° exposure. The pattern and change in total soluble sugar content with temperature and storage duration paralleled the results for sucrose, the major soluble sugar. The decrease in sucrose in cured roots during the 4-week period at 15.6° was attributed to a shift in carbohydrate metabolism in favor of starch synthesis. AIS content [primarily starch in sweet potato (4)] increased significantly during this period (data not shown), and sucrose was presumed to be the substrate for the increased starch synthesis since monosaccharides did not decrease during 4 weeks at 15.6°.

Enhanced sucrose and total soluble sugar content at 7°C compared to 15.6° was common to all four cultivars and also was reported in 'Porto Rico' (5). The increased sugar levels with increasing lengths of exposure to 7° may have contributed to the elevated respiration rates, since sugars are recognized as the primary substrates for respiration. There was considerable variation among cultivars in the extent of low-temperature sweetening, indicating that both genotype and postharvest storage temperature were involved in the regulation of carbohydrate metabolism. It was apparent that the primary sugar involved in low-temperature sweetening was sucrose. AIS content decreased in amounts similar to the increase in total soluble sugar, strongly suggesting that starch was the major source of carbon for sucrose synthesis. Low-temperature sweetening among monosaccharides favored glucose synthesis in cured 'Jewel' and 'Travis' roots, while fructose synthesis was favored in noncured roots. No change or an increase in sucrose, glucose, and fructose content occurred during the 15.6° period following 7° storage in all cultivars, except for a decrease in sucrose in cured 'Whitestar' roots following 1 or 2 weeks at 7°.

Increases in total soluble sugar content with increasing lengths of exposure to 7°C presumably would result in enhanced sweetness of the baked product. However, this potentially improved quality change may be offset by internal darkening in 'Centennial' and 'Whitestar', alterations in flavor volatiles, and increased hardcore (hard tissue in baked roots) in susceptible cultivars like 'Centennial' and 'Jasper' (2, 3). The extent of nutritional changes are not known, although there was no loss in total carotenoids or vitamin A in cured roots of any orange-flesh cultivar after 4 weeks at 7° followed by 1 week at 15.6° (13).

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