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Seasonal Sugar Concentration in Two Peach Cultivars Differing in Cold Hardiness[1]

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Abstract. **Levels of soluble sugars in bark, leaves, leaf buds and flower buds of 2 cultivars of peach (*Prunus persica* (L.) Batsch) differing in cold hardiness were compared throughout the year. Thirteen sugars – galactose, glucose, fructose, xylose, stachyose, sucrose, raffinose, rhamnose, maltose, trehalose, arabinose, ribose and mannose – were present in measurable and variable concentrations. In general, oligosaccharides accumulated, particularly in the bark, during fall and winter, whereas monosaccharides accumulated during periods of active growth. These data do not show significant differences between the 2 cultivars regarding the accumulation of these sugars and cold hardiness.**

Earlier reports by the authors (7, 8) indicate that some correlation exists between the levels of soluble sugars in peach cultivars and winter hardiness. More recent literature also indicates such correlations in apple (18); stone fruits including peach (1, 9); citrus (10, 14); wheat (4) and pine (17). Some correlations between certain sugars, either individually or in groups, and winter hardiness have also been reported (1, 4, 10, 18). Sugars and sugar alcohols were reported by Sakai (13) to be most effective among the 60 compounds used in preventing freezing injury of cabbage cells frozen in suspending medium and subjected to slow cooling and rewarming. In peach cultivars, however, little is known except that fructose, glucose, and sucrose commonly occur throughout the season in peach tissue; and raffinose and stachyose may appear during the winter (12).

Materials and Methods

Two peach cultivars, 'Lizzie' (cold hardy) and 'Loring' (cold tender), were selected for this study according to criteria described earlier (8). Trees were planted in a greenhouse in holes 0.6 m diam × 1.2 m deep lined with black plastic and filled with a 2 soil:1 perlite:1 peat mixture (v/v). To each m^3 of this soil mix were added 15 kg superphosphate, 3 kg 5N-4.3P-8.3K fertilizer, and 3 kg dolomite. To expose trees to prevailing outside temp, the greenhouse was provided with movable side panels which were left open except when killing winter temp were predicted. Each cultivar was represented by 12 trees in a randomized complete block design with 3 replications, each consisting of 4 trees. Starting when trees were 3 years old, samples were collected on the 1st and 15th day of each month, with unbranched shoots collected from Jan. 1 through Dec. 15, mature leaves (mid-shoot) from March 1 through Nov. 1, leaf buds from Nov. 15 through Dec. 15 and flower buds from Nov. 1 through April 1. All samples were collected between 8 and 10 AM and immediately frozen. Leaf and flower buds were dissected from the frozen shoots. The bark was then dissected from the wood of the partly thawed shoots. Bark, leaf and bud samples were lyophilized, ground and stored in desiccators at 5°C. Sugars were extracted from 250 mg samples by shaking in 25 ml 0.1 M H_3BO_3 at pH 8 (regeneration buffer in the analytical procedure). Interfering phenolic compounds were precipitated with lead acetate and potassium oxalate (16). A standard stock solution of sucrose, raffinose, stachyose, maltose, rhamnose, mannose, fructose, galactose, glucose, trehalose, cellobiose, arabinose, sorbose and lactose (internal standard) was prepared by dissolving 125 mg of each sugar in 250 ml 0.1 M H_3BO_3 at pH 8 and stored in a freezer. From the stock solution a working standard was prepared to contain 50 μg/ml of each sugar. A separate internal standard was similarly prepared to contain 50 μg/ml of lactose. Individual sugars were determined on an Auto-Analyzer system in which sugar-borate complexes were formed and chromatographed on a Dowex I, borate-form, anion exchange column; the eluted bands were measured colorimetrically as sugar-orcinol complexes (6, 15). A 1 ml aliquot of each extract and 0.2 ml lactose standard were placed on the column and eluted with a NaCl-H_3BO_3 gradient starting with 0.1 M H_3BO_3 at pH 8 and ending with 0.2 M H_3BO_3 + 0.2 M NaCl at pH 9.5. The eluate and 0.1% orcinol reagent in 70% (v/v) H_2SO_4 were mixed, heated to 95°C and the color measured automatically at 420 nm.

Results and Discussion

Thirteen soluble sugars, in measurable but variable concn,

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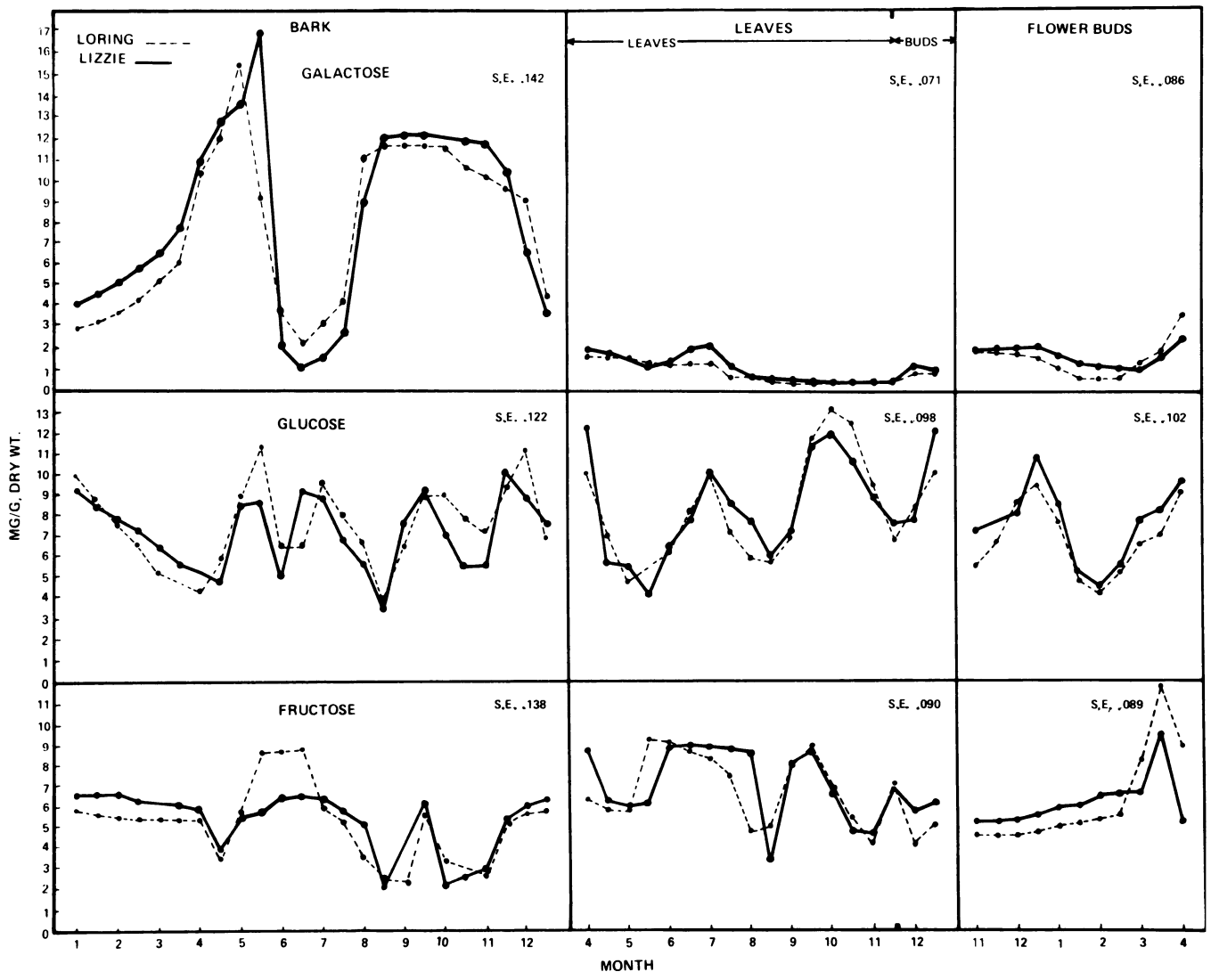

Fig. 1. Seasonal variation of galactose, glucose and fructose in bark, leaves and flower buds of 'Loring' and 'Lizzie' peaches.

were present in bark, leaves or leaf buds and flower buds of both cultivars. The sugars (listed in descending order as they appear in Fig. 1, 2) were galactose, glucose, fructose, xylose, stachyose, sucrose, raffinose, rhamnose, maltose, trehalose, arabinose, ribose and mannose.

In general, oligosaccharides appeared to accumulate during the fall and winter more than monosaccharides (Fig. 2). The latter accumulated in higher concn during periods of active growth in spring and summer. This agrees with reports on other species (4, 10). This accumulation was more pronounced in the bark than in leaves or flower buds. Similar findings were reported for other species (1, 18). However, no significant differences were observed between the 2 cultivars in oligosaccharide content of bark, leaves, leaf buds or flower buds during fall or winter despite their reputed difference in cold hardiness (7, 8). Glucose, fructose, xylose, stachyose and sucrose were the main sugars in both cultivars (Fig. 1-2). Galactose (Fig. 1) showed the highest concn, but only in the bark. Raffinose (Fig. 2) also showed a relatively high concn in the bark. Since no significant differences in sugar concn were observed between the 2 cultivars, the seasonal variations of monosaccharides and oligosaccharides are discussed below with no reference to cultivars.

Seasonal variations of monosaccharides

Galactose. In the bark, galactose generally appeared to accumulate during spring and fall as a storage carbohydrate (Fig. 1). Evidently, its consumption or translocation (possibly for synthesis of galactose-containing oligosaccharides such as raffinose and stachyose) began and continued gradually during fall. This was accelerated during winter when galactose reached its fall and winter minimum in mid-Dec. and early Jan. However, in Jan. it increased rapidly and continued to increase through spring. The concn then sharply decreased until it reached its lowest levels in mid-June for both cultivars, probably coinciding with fruit growth and possible translocation from bark to the developing fruit at that time.

In leaves, galactose was generally low in comparison with that in bark, which may indicate its transient existence in leaves. There was a slight increase in concn around June, reaching its highest level in early July, coinciding with the lowest level of galactose in the bark at that time.

In flower buds, there was no accumulation of galactose during the winter; in fact, there was a steady decrease throughout the period from mid-Nov. to early March when the concn began to increase through March and April.

Glucose. Glucose concn in the bark (Fig. 1) was relatively high in early Jan., but steadily decreased throughout the period from Jan. to March. It then began to accumulate throughout the growing season with the exception of two prominent decreases in June and mid-August, possibly due to translocation

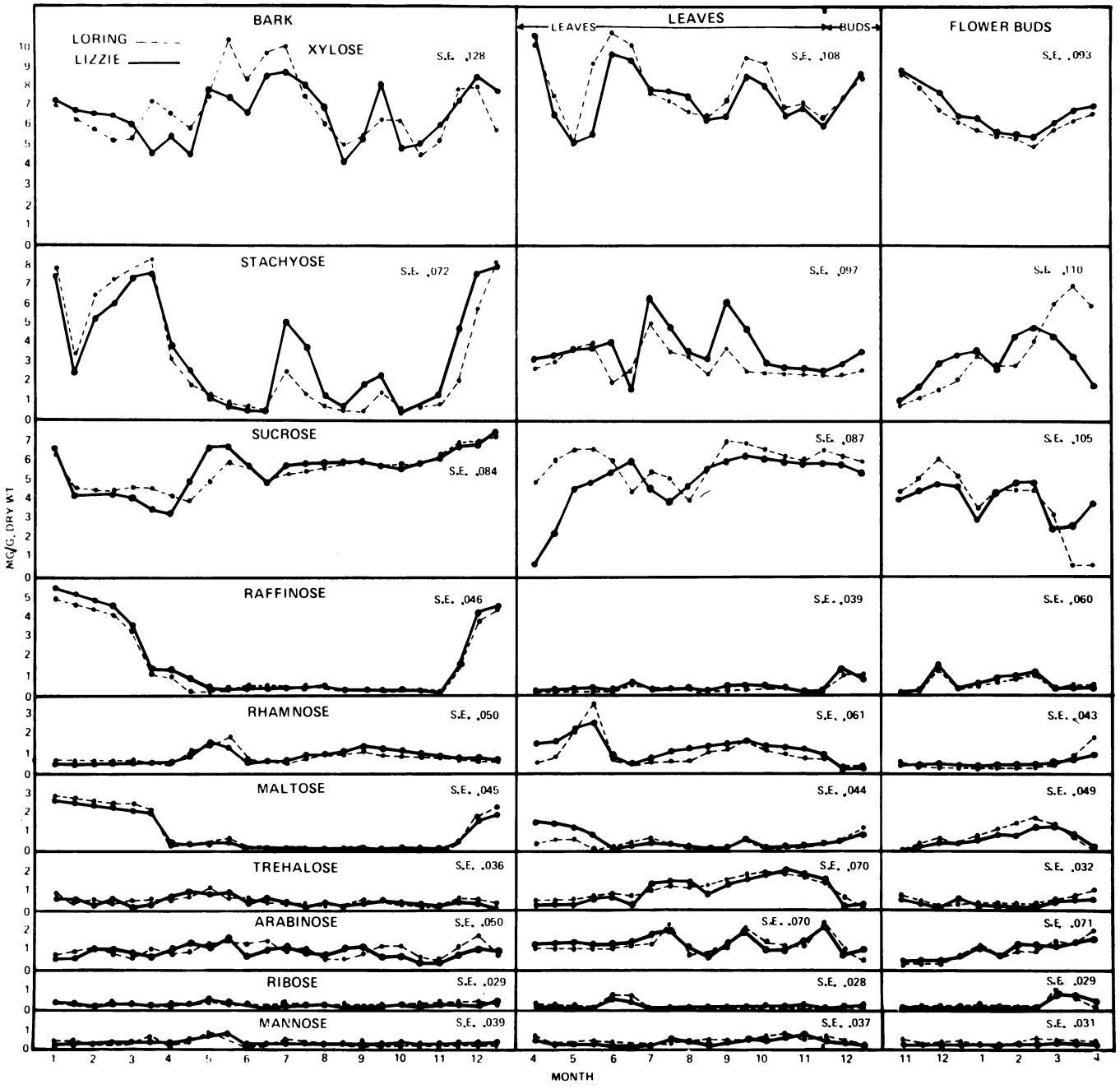

Fig. 2. Seasonal variation of xylose, stachyose, sucrose, raffinose, rhamnose, maltose, trehalose, arabinose, ribose and mannose in bark, leaves, leaf buds, and flower buds of 'Loring' and 'Lizzie' peaches.

to the developing and maturing fruit, respectively. A third decrease during Oct. and early Nov. may be due to glucose being used in starch synthesis which accumulates in shoots during the fall (2). In April, during a period of active photosynthesis of young and rapidly expanding leaves, glucose concn in bark was high but decreased rapidly in early May to mid-May which may be the result of its translocation, probably as sucrose, to the developing fruit (5, 11, 14). This was followed by a steady increase to a peak in early July followed by a transient decrease until mid-August, possibly coinciding with fruit maturation at that time. After fruit harvest, glucose concn steadily increased to a high level in early Oct. This was followed by a sharp and progressive decrease throughout Oct. and early Nov., possibly due to active translocation coupled with decreased photosynthesis in senescing leaves. A steady increase in glucose concn then took place (in leaf buds) from mid-Nov. to mid-Dec.

Earlier work (8) provides evidence that peach flower buds were devoid of starch during winter and suggests that soluble sugars were translocated to those buds from other parts of the tree. Fig. 1 shows that this appears to be the case. In early Nov., an increase in glucose concn took place in flower buds and reached a peak in early Dec. Glucose concn was also high in shoot bark at that time, which may indicate transit from leaves and possibly other parts through the bark. This increase in glucose concn despite the low temp prevailing in Nov. and early Dec. may coincide with the end of rest (3). Throughout Dec. and Jan. there was a transient decrease in glucose concn, possibly due to glucose transformation to sucrose. This was followed by a steady increase throughout the spring.

Fructose. The seasonal variations of fructose in bark and leaves (Fig. 1) appeared somewhat similar to that of glucose. In early Nov., fructose concn in flower buds was moderately

high and continued to increase gradually throughout the winter, but there was no pronounced increase in Nov. and early Dec. similar to those of both sucrose and glucose at that time. In mid-Feb. and early March its level increased sharply to reach a peak in mid-March, then decreased, also sharply, during the rest of March. This may suggest an earlier utilization of fructose rather than glucose and sucrose in bloom and subsequent fruit set.

Xylose. This pentose is the principal constituent of the structural polysaccharide cell wall materials, xylans. As such, its concn may decrease during periods of active growth. In the bark its concn decreased, presumably through translocation to other parts of the tree where active growth was taking place. Fig. 2 shows that this seems to be the case in early spring up to mid-May and during fruit ripening in August and Sept.

In leaves seasonal variations of xylose appeared to follow, somewhat, a similar pattern as that for bark.

In flower buds (Fig. 2) xylose was also high in fall and gradually decreased in winter reaching a minimum in mid-Feb. which was followed by a gradual increase in early spring.

Seasonal variations of oligosaccharides

Stachyose. In the bark, stachyose accumulated in the fall and winter, although during Jan. a definite decrease took place. In spring and summer, its concn generally decreased, but an increase took place from mid-August to early Oct.

Stachyose was generally low most of the year in the leaves, however, 2 increases in its concn, similar to those described above in bark, took place, also at the same time periods.

Stachyose was low in early Nov. in flower buds, but steadily increased until mid-Dec. and decreased slightly during Jan. It then increased reaching its highest level in mid-Feb. to mid-March and was followed by a steady decrease.

Sucrose. Sucrose appears (Fig. 2) to be an important storage carbohydrate in the bark (2, 18); it was generallly high throughout the year. However, starting in early Jan., its level in bark began to decrease until it reached its lowest level in early April. This decrease in Jan. was similar to that observed for stachyose which may be an indication of active translocation to dormant fruit buds (3). Again, the decrease in early spring may be an indication of active translocation to the developing fruit buds. Starting in early April to mid-April, sucrose increased until mid-May after which it gradually decreased until mid-June. This decrease may also be due to translocation of sucrose to the developing fruit. A transient increase then took place and continued throughout the rest of the year.

Seasonal variations in leaves in early spring and summer (Fig. 2) appeared to be similar to the pattern in the bark at that time.

In flower buds, sucrose, like glucose, was also high in fall, and may similarly coincide with the end of rest. Its concn gradually decreased from early Dec. until Feb. when a sharp decrease occurred. This may be the result of increased sugar metabolism, probably involving sucrose transformation to glucose and fructose, by the developing flower buds at a time when reserve sugar (or starch) in the bark was low. It is interesting to note that similar decreases were observed (Fig. 2) in flower buds for stachyose, raffinose and maltose.

Raffinose. This trisaccharide has been reported (4, 10) to play an important role in winter hardiness. The present work, however, suggests that this may be true only in the bark. Fig. 2 shows that raffinose accumulated in considerable concn in late fall and continued to accumulate throughout the winter up to early Jan. At that time it began to decrease gradually until early spring when very low levels were present and continued so throughout the summer and early fall.

In leaves seasonal variations of raffinose were similar to those in bark. They ere low throughout the growing season with a minor increase in Nov. and Dec.

In flower buds, raffinose was generally low with only a moderate increase in fall and winter (Fig. 2). A decrease, similar to that reported above for other oligosaccharides, took place in mid-Dec.

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