A long white sheath lining the inner part of the soft seedless fruit (Fig. 5A) enclosed an elongated cavity throughout its length and the remains of the degenerate ovule which are located at its distal end. This sheath is the endocarp, consisting mainly of large sclerenchymatous cells in 1 or 2, and occasionally 3 layers. The sclerenchyma is surrounded by a narrow strand of deformed, small cells. The sheath origin became evident in an early stage of fruit development. Cells of the inner epidermis of the pericarp gradually enlarged, becoming vacuolate, and part of them divided into 2 layers. The walls thickened and lignified (Fig. 5B). The neighboring 1 or 2 layers of very small tangentially elongated parenchymatous cells become compressed and deformed. They remained always adherent to the sclerenchymatous cells.

Development of the endocarp was similar in seeded fruit, but the lignified cells were smaller and consisted of only 1 layer. One or 2 layers of small parenchymatous cells were adherent to the sclerenchyma. The endocarp becomes part of the seed coat and almost inseparable from it (Fig. 5C). Valmayor (1967) described this for other cultivars of avocado.

Discussion

Results of this study indicate that the typical seedless fruit in 'Fuerte' and 'Ettinger' avocado is the outcome of seed degeneration (stenospermocarpy) and not automatic parthenocarpy (6). This conclusion is based on the fact that no fruit were found without an embryo or endosperm in the more than 200 seedless fruits examined. Furthermore, all fruitlets examined at the beginning of seed tissue degeneration, had an embryo or endosperm or both. The set of seedless and seeded fruit was also prevented when pollination was prevented (10).

The facts that 1 tree was found with flowers lacking ovules which produced parthenocarpic seedless fruit and parthenocarpic seedless fruit can be produced by growth hormones (gibberellins) indicate that automatic parthenocarpic fruit will develop under special conditions. Profuse flowering of avocado trees and profuse fruit set at the first fruit development stages lead to competitive conditions which probably eliminate the possibility that automatic parthenocarpic fruitlets will survive. It is known that the avocado seed coat contains a high level of growth hormones (1, 2, 4) that may exert a strong sink effect for photosynthetates. The fact that part of the seed coat in seedless fruit remains viable and even continues to grow might not be accidental.

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A Sensitive Method for Measuring Changes in Calcium Concentration in 'McIntosh' Apples Demonstrated in Determining Effects of Foliar Calcium Sprays[1]

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Additional index words. Malus domestica, postharvest physiology

Abstract. **Three different methods of sampling flesh of apple (*Malus domestica* Borkh.) for Ca analysis produced different results due to uneven Ca distribution in the fruit. A sensitive, reproducible method of sampling and analyzing the outer cortex of the calyx half of fruits indicated that Ca concentration in different parts of the fruit changed significantly after harvest, decreasing in the core and increasing in the outer cortical tissues. Massive applications of $CaCl_2$ to trees shortly before harvest increased flesh Ca concentrations and red coloration of the fruit, decreased flesh softening during storage and senescent breakdown after storage, but caused significant injury to the fruit.**

Fruit calcium deficiency is associated with occurrence of some of the most serious apple disorders worldwide (9). Fruit are often too low in Ca to maintain high quality over long storage periods if they have been produced on trees that are quite young, growing on dwarfing rootstocks, or with excessively vigorous vegetative growth, or under moisture stress, or if they are large fruit from trees with a light crop. Soil management practices such as maintaining pH at 6.2 to 6.5, injecting $Ca(OH)_2$ slurry into the soil, and annual applications of gypsum or $Ca(NO_3)_2$ may help maintain adequate levels of Ca in apples under non-stress conditions, but they usually are ineffective in correcting Ca deficiency. For this, the treatment most widely used is the application of 4 to 8 foliar sprays of $CaCl_2$ or $Ca(NO_3)_2$ at intervals throughout the growing season.

Lewis and Martin (4) showed that Ca is not uniformly distributed in apple flesh but ranged from 150 ppm in the stem

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end to less than 80 ppm in the calyx end. Further, Perring and Jackson (7) found that Ca concentration declined as the mass of the apple increased. We have noted that cork spot, bitter pit, and senescent breakdown usually commence in the cortical tissue directly beneath the peel at the calyx end of 'McIntosh' apples, and that these disorders are more prevalent in large than in small fruit. Thus, in trying to accurately assess effects of treatments on fruit Ca concentration and on incidence of disorders, it would appear that tissue for analysis should be taken from clearly identified sources (3). This paper describes a sampling technique that sensitively detects the effects of foliar Ca sprays in increasing fruit Ca, and that measures changes in flesh Ca concentrations during maturation and storage.

Materials and Methods

Sampling techniques. Three sampling techniques were compared for estimating Ca concentration in apple tissue. Apples 7 cm (2¾ inch) in diameter were selected on August 20, 1977, from limbs 1.5m above the ground on five 15-year-old 'McIntosh' apple trees on Malling (M) 7 rootstock, growing in medium drained Gloucester-Scituate sandy loam soil at pH 6.5. Sixty fruit were taken from each tree, divided into lots of 20 apples each, and the flesh was sampled by 1 of the 3 methods.

In Method 1, peel from 20 apples was removed from the calyx half of the apples with a mechanical peeler (White Mountain Apple Peeler, White Mountain Freezer, Inc., Winchendon, Mass.) and the peel was discarded since it contains 2 to 3 times the Ca concentration of subtending flesh. A thin layer of flesh directly beneath the peel then was taken with the peeler from around the circumference of the apple; these samples from the 20 apples were composited and designated "outer cortex." From each fruit another thin circumference layer was taken 1 cm beneath the surface and designated "1 cm deep flesh." A third sample was taken from the core area of the calyx half of each apple; a 13 mm cork borer was used to remove a cylinder of tissue parallel to the core and a 0.5 cm thick disc was taken from the cylinder. This "core" sample included vascular and carpel tissue, but not seeds.

In Method 2, 20 apples were peeled to remove 1 cm of flesh, and 2 wedges 2 to 3 cm wide by 2 cm deep were removed from opposite sides of each apple (Dr. George M. Greene, Biglerville, Pa, personal communication). The 40 wedges per tree were then composited. Method 3 was that of Perring and Wilkinson (8). Twenty apples were peeled and from each fruit 2 thin longitudinal wedges of tissue, extending from calyx to stem and from surface to core, were taken from opposite sides of the apple. Seeds were excluded and the 40 wedges per tree were composited.

All samples were frozen at −30°C for 7 days. Then, 35 g of frozen tissue was thinly sliced onto wax paper and dried in a forced draft oven at 48° for 24 hr; after 3 to 4 hr, the tissue was turned onto new wax paper to reduce adhesion. Temperature was then increased to 65° for 10 days. After drying, the tissue was chilled 5 min at −30°, and ground in a micro-Wiley mill kept in a freezer chest at −30°; this procedure maintained dryness of the samples and produced thorough grinding and mixing.

One g of ground cortical tissue, or 0.5 g of core tissue, was digested with 3 ml of 70% perchloric acid and 2 to 4 ml of 70% nitric acid in a 100 ml Kjeldahl flask, and Ca was analyzed with an atomic absorption spectrophotometer.

Measuring changes in apple Ca concentration. Previous results in our laboratory indicated that Ca concentrations in different parts of the apple change during maturation and storage. To examine this question further, apples were selected from each of the 5 'McIntosh' trees described earlier. On August 10, 20, and 30, 20 apples 7 cm in diameter were selected and "outer cortex," "flesh 1 cm deep," and "core" samples were taken by Method 1. On September 10, sixty 7 cm diameter apples were selected from each tree and were divided into 3 lots of 20. The first lot was sampled September 10; the second lot was placed in 0°C air storage and sampled January 10; the third lot was placed in CA storage (3% O_2, 5% CO_2, 3°) and sampled April 10. Samples were dried and analyzed for Ca as in Method 1, above.

Measuring the effect of $CaCl_2$ sprays on apple Ca concentration. Method 1 was also used in part to assess the effect of a foliar $CaCl_2$ treatment on the Ca concentration in apples, in conjunction with assessment of treatment effects on storage life of the fruit. In 1977, 5 'McIntosh' trees in the orchard block described above received one of the following treatments: (A) Control; (B) 100 kg/ha (90 lb./acre) of $CaCl_2$ applied 14 days before harvest; (C) 100 kg/ha applied 2 days before harvest; (D) 100 kg/ha applied 14 days and again 2 days before harvest. One hundred apples from each of the 20 trees were placed in 0°C air storage until January, or in CA storage until June, when they were examined for incidence of storage disorders. In January "outer cortex" Ca concentration was determined on 20 fruit 7 cm in diameter from each treatment. This test was repeated in 1978 except that a combination of sprays both 14 and 2 days before harvest was not included, and that tissue samples for Ca analysis were taken at harvest rather than after storage in January.

Results and Discussion

Sampling technique. Estimates of Ca concentration in apples varied greatly when different parts of the fruit were analyzed (Table 1). Method 1 showed that Ca concentration in the calyx half of the fruit increased as depth of sampling increased, with the core area containing 4 times the concentration of the outer cortex. Results of similar analyses in 2 other years (Table 1), show the consistency of this relationship of Ca concentration and tissue location among seasons. Method 1 was chosen for further studies because it sensitively detected small Ca differences.

Changes in apple Ca concentration. We reported previously (2) that Ca concentration changed during maturation and storage of 'McIntosh', 'Delicious', and 'Cortland' apples in 1976-77. These data were obtained using Method 1. 'McIntosh' apples were sampled again in 1977-78 to confirm these changes, and Table 2 presents some of our previous data (1976) for comparison with the current data (1977). These results confirm that Ca concentration declines in all 3 portions of the apple during maturation, whereas after harvest it increases in the outer tissues while continuing to decline in the core region. Initial concentrations and changes with time were remarkably consistent between seasons.

Table 1. Concentration of Ca in 'McIntosh' apple flesh (± SE) sampled at harvest, by 3 different sampling methods.

	Flesh Ca concn (ppm dry wt basis)				
	Method 1[z]				
Year	"Outer cortex"	"1 cm deep"	"Core"	Method 2[y]	Method 3[x]
1977	102 ± 3	116 ± 2	422 ± 7	130 ± 2	220 ± 5
1976	91 ± 2	105 ± 2	422 ± 10	---	---
1978	101 ± 1	126 ± 2	548 ± 9	---	---

[z]Samples taken at 3 different depths on calyx half of fruit.

[y]Two-cm deep wedge of tissue, starting 1 cm below the peel, sampled from 2 opposite sides on calyx half of fruit.

[x]Thin longitudinal slice, extending from stem to calyx and from surface to core, taken from 2 opposite sides of fruit.

Table 2. Changes in Ca concentration in 'McIntosh' apple flesh, at 3 depths in the calyx half of the fruit, during maturation and storage.

Depth	Year	Flesh Ca concn (ppm dry wt basis)					
		Aug. 10	Aug. 20	Aug. 30	Sept. 10[z]	Jan. 10[y]	April 10[x]
"Outer cortex"	1976-7	92 b[w]	91 b	76 c	75 c	97 b	108 a
	1977-8	---	101 b	82 c	83 c	131 a	134 a
"1 cm deep"	1976-7	126 a	105 b	103 b	93 c	104 b	110 b
	1977-8	---	116 b	93 c	98 c	151 a	145 a
"Core"	1976-7	506 a	422 b	373 c	333 d	224 e	226 e
	1977-8	---	422 a	334 b	315 b	258 c	228 c

[z]Harvest date.

[y]Stored in 0°C air for 4 months.

[x]Stored at 3° in controlled-atmosphere storage for 7 months.

[w]Mean separation within rows by Duncan's multiple range test, 5% level.

Table 3. Effect of $CaCl_2$ sprays applied at a rate of 101 kg/ha before harvest on Ca concentration and fruit quality of 'McIntosh' after storage.

$CaCl_2$ treatment	Ca in "outer cortex" (ppm)	Senescent breakdown (%)		Firmness (kg) after CA storage
		Air storage[z]	CA storage[y]	
	1977-78			
Control[x]	142 ± 7[w]	13 ± 3	8 ± 2	3.38 ± .16
2 weeks preharvest	171 ± 3	3 ± 2	4 ± 2	3.35 ± .04
2 days preharvest	194 ± 4	2 ± 1	1 ± 0	3.69 ± .07
2 weeks and 2 days preharvest	221 ± 9	5 ± 3	1 ± 0	3.42 ± 0.6
	1978-79			
Control	73 ± 3	12 ± 6	10 ± 2	3.49 ± .07
2 weeks preharvest	191 ± 6	0 ± 0	0 ± 0	3.90 ± .04
2 days preharvest	123 ± 6	4 ± 0	1 ± 0	3.84 ± .09

[z]Stored in 0°C air for 4 months.

[y]Stored at 3° in controlled atmosphere storage for 7 months.

[x]Control samples contained 92 ± 8 ppm Ca at harvest.

[w]± SE.

We believe that the decline in Ca during maturation is a dilution effect from rapid fruit growth, but it could also be influenced by moisture stress during this period, for Ca can be removed from the fruit if water is withdrawn from fruit to leaves (11). The changes after harvest must be due to redistribution of Ca in the fruit, although the way this occurs is not known. We have shown previously that these changes after harvest are independent of the Ca concentration in the outer flesh at harvest (2).

Effect of $CaCl_2$ sprays on apple Ca concentration. Massive application of $CaCl_2$ shortly before harvest markedly increased the Ca concentration in the outer flesh, decreased fruit softening during storage, and decreased senescent breakdown following storage (Table 3). In 1978, treatment 2 weeks before harvest was more effective than treatment 2 days before harvest, but in 1977 the opposite results were obtained. This may be due to a heavy rainfall (2.8 cm) between the 2 treatments in 1977, which would have removed much of the residual $CaCl_2$ from the first spray and therefore reduced the potential for continued uptake during storage.

The Ca concentrations in 1978-79 appear to be much lower than in the previous year. This is because samples were taken in January in 1977-78, rather than at harvest, by which time significant increases in Ca had occurred in this tissue (Table 2). These results illustrate the importance of sampling at the same time of the season if comparisons are to be made.

The massive $CaCl_2$ sprays caused severe marginal necrosis and cupping of the leaves, which in turn increased light penetration into the tree canopy and greatly increased red coloration of the fruits. In 1978, 95% of the fruit from trees sprayed 2 weeks before harvest were graded "Extra Fancy," while only 23% of the controls graded "Extra Fancy" due to lack of red color. The sprays did not increase preharvest drop in these tests. In 1978 many of the fruit sprayed with massive $CaCl_2$ concentrations developed small necrotic spots on the skin which were not detected at harvest but which were clearly evident after storage due to desiccation of flesh around the spots. Previous work did not report any effect of similar treatment on color or fruit injury to 'Spartan' apples (5).

Data in Table 1 clearly show that analyses of Ca concentrations in apples cannot be directly compared unless the same portion of the fruit is sampled in each investigation. We prefer to sample the tissue just beneath the peel in the calyx half of the apple. This appears to be the portion of the fruit with lowest Ca concentration and therefore the least "background" Ca against which to judge treatment effects. Furthermore, this is the area where Ca-related disorders most often originate, and hence may be the area where adequate Ca is most critical. For direct comparison of analytical results, the fruit must have been sampled at the same stage of development, since

Ca concentration changes with time (Table 2). For most purposes, harvest time may be the most appropriate time of sampling, but to assess late-season or postharvest treatments, samples must be taken after storage, since Ca continues to be absorbed and translocated from residues after harvest (1, 6).

We believe that our method of sampling only the outer cortex flesh of the calyx half of the fruit provides a sensitive, reproducible assessment of the Ca nutrition of apples. Its utility in measuring both natural changes during fruit ontogeny and the effects of treatments applied to the fruit tree are shown here. However, for a rapid survey of the nutritional status of different lots of apples, another technique may be preferable. In England, such surveys using whole-fruit samples and wet digestion have been effective in predicting storage life of apples by mineral composition (10).

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Inheritance of Wood Hardiness among Hybrids of Commercial and Wild Asian Peach Genotypes[1]

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Additional index words. Prunus persica, cold tolerance, winter injury, freezing injury, fruit breeding

Abstract. **Seven clones of peach (*Prunus persica* (L.) Batsch) varying in hardiness, and 5 progenies derived from them were used to study the inheritance of cambium, xylem, and vegetative bud hardiness. Mean progeny cambium and xylem hardiness could be predicted from average parental performance, but vegetative bud hardiness could not. Injury within dates and tissues was highly heritable and environmental variation was very low. Correlations among injury ratings for different tissues, temperatures, and dates were low when based on individual trees indicating poor repeatability of individual genotypes over time. Correlations based on family means were higher. The low correlations based on individual trees indicate that selection based on individual phenotypic performance in a single test might be ineffective.**

The need for improved wood hardiness in peaches is well documented (2, 6, 9). Although artificial freezing has long been used to assess hardiness (6), most cultivar improvement is the result of identifying hardy genotypes after test winters. Studies of hardiness in other crops indicate that it is a genetically complex trait (7, 8, 11, 22). Field observations and logic suggest that the same is true for peach. Mowry (17) concluded that inheritance of flower bud hardiness in peach was quantitative in nature. Hardy parents tended to transmit greater hardiness to their offspring than did tender parents. Some evidence of specific combining ability was also indicated.

Recently, very cold-hardy peach germplasm has become available from northern China (19, 22). Its fruit quality is poor, and hybridization with commercially desirable fruiting types is necessary to combine improved hardiness and commercially acceptable fruit quality. This process may require 20 or more years. Therefore, maximizing breeding efficiency is essential.

This study was undertaken to examine inheritance of wood hardiness in crosses between commercial cultivars and recently introduced cold-hardy peaches.

Materials and Methods

Plant material. Seven parents were used. 'Harken' and 'Canadian Harmony' are scion cultivars developed at the Harrow station. 'Garnet Beauty' is an early ripening mutation of 'Redhaven'. All 3 are reasonably well adapted to northern growing conditions.

'Siberian C' and 'Harrow Blood' peach seedling rootstocks are very hardy under northern conditions (14, 15, 16). 'Siberian C' was selected at the Harrow Research Station from seeds originating in China. 'Harrow Blood' is a chance seedling found near Harrow in 1938. 'Harrow 6116-256' and 'Harrow 6116-292' (henceforth designated 'Harrow 256' and 'Harrow 292'

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