

Fig. 3. Several hundred "pathogen-free" strawberry meristem plantlets were stored at 4°C air temperature in individual test tubes held in 22 x 13 cm racks.

Fragaria X ananassa Duch. cvs., Aiko, Aliso, Benton, Cruz, Donner, Fresno, Hood, Lassen, Marshall, Northwest, Puget Beauty, Salinas, Olympus. Sequoia, Shasta, Sierra, Solana, Tioga, Toro, Torrey, Tufts, and 30 experimental selections; F. vesca L. cvs. UC 1, 3, 4, 5 and 6; and F. virginiana Duch. cvs.

UC 10, 11 and 12.

The maximum duration and optimal conditions of storage have not been determined except that, whereas plantlets stored better at 1°C than at 4°. they were killed by slow freezing. Cryogenic storage was not attempted.

This relatively inexpensive storage

method has reduced costly greenhouse maintenance of strawberry nuclear foundation stock in California. In the future, this long storage of "pathogenfree" meristem plantlets may facilitate germplasm preservation of other plants, such as fruit trees and potatoes (1, 4), which can be cultured from meristemtips (3, 6) or from tuber discs (5). This system may also be adaptable for preservation of many other plants, currently maintained as budsticks, scions, or cuttings, when suitable culturing systems have been developed.

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## Field Performance of Cold-stored Plants of Strawberry Cultivars and Selections in the Pacific Northwest<sup>1</sup>

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Abstract. Survival and vigor, after -1°C storage, of Totem strawberry (Fragaria x ananassa Duch.) plants dug at 8 different dates, from November 15, 1973 to April 1, 1974, was affected adversely only when dug April 1. Predigging treatment with Methyl 1-(butylcarbamoyl)-2benzimidazolecarbamate(benomyl) did not affect survival or vigor from any of the dates. In 1975, digging date (March 1 vs April 1) did not affect the survival of plants of 4 cultivars or 6 selections. However, plants of 'Totem' and 'Northwest' and 2 selections dug April 1 showed reduced vigor compared to those dug March 1.

Dormant strawberry plants, previously treated with fungicide, are commonly dug and stored until spring planting (2, 3, 4). Such plants will remain in

tended periods (3). However, in the Pacific Northwest non-dormant plants are often dug and stored at -1°C because more favourable conditions for digging can exist in the spring, after growth has commenced, than during the winter. Moreover, Freeman and Pepin (1) found that non-dormant plants of 'Northwest' did not grow as well as those of 'British Sovereign'

excellent condition at -1°C for exor 'Siletz' after -10 storage.

In recent years 'Totem' has become an important cultivar in the region. Under commercial conditions springdug (non-dormant) plants of the cultivar have appeared less vigorous and, in some instances, have had lower survival rates than winter-dug (dormant) plants after -1°C storage. In the present study, vigor and survival were measured after -10 storage of 'Totem' plants which have been sprayed with the systemic fungicide benomyl compared to those which had not been sprayed from several digging dates through the winter and early spring of 1973-1974. In addition the growth responses of 'Totem' plants and those of 3 other cultivars grown commercially in the Pacific Northwest and of 6 advanced selections from the British Columbia strawberry breeding program were compared subsequent to digging at 2 different dates in 1975 and storage at -1°. The first date was chosen to ensure dormant plants and the second to ensure nondormant plants.

1973-1974 treatments. Sixty-four field plots, each consisting of 12 'Totem' plants, were established at Abbotsford, British Columbia in May, 1973. Individual plots were designated with proposed 2 week digging dates beginning Nov. 15 and ending April 1 (except

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for the period Dec. 15 to Feb. 1). For each digging date plants were either sprayed with benomyl at the rate of 2.24 kg/ha (2 lb./acre of benomyl as 50% WP) in mid-Oct. or not sprayed. The experimental design was a split plot with digging dates as the main plots and benomyl treatments as subplots. At each digging date between 43 and 112 plants were dug per plot and stored in perforated plastic bags at -1°C until May 2. At that time the percentage of plants from each plot with fungi was determined. Twelve plants from each bag were planted in the field on May 2 in the same design from which they were obtained. Leaf spread, as an indicator of vigor, of each plant was determined on June 7. Yield data were taken from the planting in 1975. The remainder of the plants from each plot were placed in an adjacent area, also in the same design, to determine plant mortality after 5 weeks.

Pre-storage benomyl spray applied to the 'Totem' plants in 1973 did not adversely affect survival or plant vigor. Therefore, data were combined from sprayed and non-sprayed plots for each digging date. Post-storage mortality was very low from each digging date except April 1 when 8% of the plants died (Table 1). The most vigorous plants were from digging dates between Nov. 15 and Feb. 1; the least vigorous plants were those dug on April 1 (Table 1). In other years this time period could vary considerably depending on weather conditions through the winter months. The most important consideration would be the assurance of dormant plants prior to storage.

About 90% of the fungi identified on plants after storage was Botrytis cinerea Pers. ex Fr. Benomyl application resulted in significantly fewer plants with fungi than those which had not been sprayed for each digging date (Table 2). Among the sprayed plants those dug December 15 or earlier showed the most fungi and those dug March 15 and April 1 the least. More unsprayed plants dug November 15 showed fungi than those dug at any other date. The presence of fungi did not affect plant survival or vigor. This is in contrast to the results of Maas and Scott (4) and of Guttridge and Montgomerie (2) who found that poststorage survival rates were adversely affected by the presence of fungi. Under less favourable growing conditions the presence of fungi could have adversely affected survival and vigor of plants from storage. It is possible that fungi on stored 'Totem' plants do not adversely affect post-storage performance. Similarly Maas (3) found that benomylsprayed plants of 4 out of 19 cultivars had no advantage over non-sprayed plants. Furthermore the length of storage time may have influenced the

effects of fungi on plant survival and vigor. The maximum length of storage time in the present study was 5½ months whereas in other studies it has varied between 7 and 9½ months (2, 3, 4).

Table 1. Percent mortality and vigor 5 weeks after planting of 'Totem' strawberry plants dug on 8 different dates in 1973 and 1974 and stored at -1 OC prior to planting.

Digging date	Mortality <sup>2</sup> (%)	Leaf spread <sup>y</sup> (cm)	
1973			
Nov. 15	2	18.70 a <sup>x</sup>	
Dec. 1	3	18.86 a	
Dec. 15	< 1	18.81 a	
1974			
Feb. 1	< 1	18.27 ab	
Feb. 15	< 1	16.23 d	
March 1	< 1	17.10 c	
March 15	3	17.69 bc	
April 1	8	14.89 e	

2% mortality was based on the examination of between 75 and 161 plants from each of 4 replications per digging date.

yEach measurement is the mean from 192 plants.

XMean separation by Duncan's multiple range test, 5% level.

Table 2. Effects of a preharvest benomyl spray and digging date on fungi on 'Totem' strawberry plants after storage at -1°C.

	% plants with fungi <sup>zy</sup>		
Digging date	Benomyl	Control	
1973	<del></del>	······································	
Nov. 15	50.3 bcd <sup>x</sup>	94.3 f	
Dec. 1	43.1 bc	83.5 ef	
Dec. 15	44.7 bc	70.7 de	
1974			
Feb. 1	3.2 a	53.7 bcd	
Feb. 15	4.8 a	62.5 cde	
March 1	2.8 a	71.3 de	
March 15	1.2 a	30.8 b	
April 1	1.1 a	48.6 bcd	

 $^{\rm Z}$ Based on mean of 4 replications. Between 43 and 112 plants were examined for each treatment.

<sup>y</sup>Approx 90% Botrytis cinerea. Other fungi were species of Trichoderma, Rhizopus, Phytophthora, Pythium, Penicillium, Alternaria.

XMean separation by Duncan's multiple range test 5% level

There were no significant differences in yield the following year from 'Totem' plants originating from different digging dates or from benomyl treatments. Freeman and Pepin (1) also found no relationship between digging date and crop yield in subsequent years.

1975 treatments. Ten plants of 4 cultivars and 6 selections were dug on each of 2 dates, March 1 and April 1, from plots in each of 4 replications of an established yield trial. The plants were assumed to be dormant on March 1 since there was no evidence of growth. The plants were stored in perforated plastic bags until May 1 when they were planted in the field in a split plot design with cultivar or selection as main plot and digging date as sub-plot. On May 28 leaf spread of each plant was determined and on June 17 leaf weight of each determined. Plant mortality per plot was determined after 6 weeks.

Mortality of plants after -1°C storage was very low and showed no relationship to cultivar or selection or to digging date. Post-storage vigor was greater for plants dug March 1 compared to April 1 for all cultivars and selections except 'BC 70-20R-15' (Table 3). Leaf spread on May 28 was significantly greater for the earlier-dug compared to the later-dug plants of 'Totem', 'Northwest', 'BC 67-18-76' and 'BC 70-17-44'. There were no significant reductions in leaf wt of plants from the April 1 compared to the March 1 digging date.

Post-storage performance of 'Totem' plants dug March 1, compared to April 1, agreed with the 1974 observations. Earlier-dug or dormant plants of 'Northwest', 'BC 67-18-76' and 'BC 70-17-44' also were more vigorous than later-dug or non-dormant plants. The observations on 'Northwest' agreed with those of Freeman and Pepin (1). Also, the observations on 'Totem' and 'Shuksan' agreed with post-storage observations made on later-dug plants of the 2 cultivars grown under commercial conditions. Later-dug plants of 'Shuksan' have had more initial vigor after storage than later-dug 'Totem' plants.

Table 3. Vigor, expressed as leaf spread and leaf wt of plants, of strawberry cultivars and selections dug on 2 dates in 1975 and subsequently stored at -1°C prior to planting.

Cultivar or selection	Leaf spread (cm) on May 28 <sup>z</sup>		Leaf wt (g) on June 17 <sup>2</sup>	
	March 1	April 1	March 1	April 1
Totem	14.56	11.78 a <sup>y</sup>	8.21	6.10
Shuksan	13.84	12.29	8.86	7.15
Northwest	11.85	9.88 a	5.66	4.72
Rainier	14.66	13.28	7.13	6.15
BC 67-18-76	14.21	11.44 a	7.82	6.35
BC 69-5-34	16.99	15.84	9.92	8.05
BC 69-30-3	16.24	15.54	9.83	8.14
BC 70-17-44	16.89	13.79 a	8.71	6.24
BC 70-20R-15	15.68	16.24	10.12	9.97
BC 70-22-72	16.61	15.11	7.79	7.15

<sup>&</sup>lt;sup>z</sup>Each measurement is the mean from 40 plants.

y Indicates significantly less leaf spread, at the 5% level, on plants from the April 1 compared to the March 1 digging for the respective cultivar or selection.

Since winter weather conditions in the Pacific Northwest can often be unfavorable for digging dormant strawberry plants, a potentially valuable attribute of any cultivar for the region would be the ability of non-dormant plants to show maximum post-storage vigor. This could be another selection criterion for strawberry breeding programs in the region. Leaf spread 4 or 5

weeks after planting would be a useful measurement for determining this poststorage vigor.

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## Relation of Pruning Time and Inoculation with *Pseudomonas syringae* van Hall to Short Life of Peach Trees Growing on Old Peach Land<sup>1</sup>

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Abstract. Three-year-old peach trees [Prunus persica (L) Batsch. cv. Elberta] growing on old peach land where a high incidence of bacterial canker was suspected in previous plantings, were pruned or pruned and inoculated with Pseudomonas syringae van Hall in October, December, February, or April. All trees pruned and inoculated in October or December were either dead or dying by May. P. syringae was recovered from all of the October-inoculated and from 86% and 71% of the December- and February-inoculated trees, respectively. Also, 43% mortality occurred in February-pruned and inoculated trees. Uninoculated but early-pruned trees showed severe short life or decline symptoms with 43% mortality following October and December pruning. On the other hand, April-pruned trees, whether inoculated or not, showed less short life or decline symptoms than early-pruned trees and no deaths occurred in April-pruned and inoculated trees.

In the southeastern USA, peach tree short life or decline is severe and there have been numerous reports (1, 3, 4, 6, 13, 14) of factors involved in the premature death of trees. We have shown that on land where peach trees have been grown previously, the time of pruning is one factor which affects the longevity of trees (1, 3, 13). Bacterial canker, caused by Pseudomonas syringae, has also been shown to be one of the factors affecting survival of peach trees on both new (6) and replanted sites (4, 14). In the test reported here, the relationship between pruning time and inoculation with P. syringae to the short life or decline of peach trees growing on old peach land was investigated.

On land in central Georgia where bacterial canker was suspected as a major factor in the history of severe decline, a block of 56 'Elberta' peach trees, 3 years old, was divided into 8 treatments with 7 replications. In Oct., Dec., Feb. and April, 14 trees were pruned on each date and immediately inoculated with *P. syringae* or sterile deionized water. The bacterial culture used, designated as FV-3, had been

isolated by us from a tree in the same orchard. This isolate was identified by its production of green fluorescent pigment on King's Medium B (8) and by its negative reaction to the oxidase test (9) followed by a comparison by us (4) to a known virulent strain of P. syringae, B-3 from California. In greenhouse tests isolate FV-3 was as virulent on peach seedlings as isolate B-3. Bacteria from a 48-hr culture were suspended in water, the concn adjusted to 107 cells/ml and the suspension used as inoculum. On each treatment date, cut ends of 6 branches randomly selected on the periphery of each pruned tree were inoculated using a small hand sprayer. Each inoculated site was then covered with moist absorbent cotton and masking tape to delay drying.

Measurements of resulting cankers and re-isolations of bacteria were conducted 60 days after inoculations. No re-isolations were attempted in June, since reports by other workers (2, 7) and our experience indicated that little or no recovery of *P. syringae* could be expected when daily maximum temp averaged 30°C. Re-isolations were made from living tissue approximately 25 mm below the inoculated site or necrotic tissue. In preparation for re-isolation, a strip of bark approx 6 x 12 mm was removed with a sterile scalpel.

A rotary hand drill fitted with a sterile 3 mm bit was used to remove chips of wood which were caught in a tube containing King's Medium B. Resulting colonies of *P. syringae* were identified by the production of green fluorescent pigment and the oxidase test.

On May 26 and Sept. 8 tree deaths were recorded and all surviving trees were visually rated for decline symptoms from 1 (severe decline) to 8 (no apparent symptoms). Symptoms used in ratings were defoliation, yellowing and dead branches.

Although canker length is not the only measure of severity of the disease, inoculation with P. syringae in Oct. or Dec. resulted in cankers significantly longer than those from later inoculations (Table 1). After 60 days, Oct.and Dec.-pruned and inoculated trees had canker lengths of 45 mm and 31 mm, respectively. Little or no canker development occurred on trees in any of the other treatments. We observed gumming from most of the cankers on the Oct.- and Dec.-inoculated trees. Gumming and cankers are symptoms which we have regularly observed in declining peach trees in Georgia. Trees inoculated with water had no cankers on twigs, but most exhibited other decline symptoms including chlorotic foliage, canker on trunk or scaffold limbs, and symptoms of cold injury (5). In a recent paper (4) we suggested that there are frequent periods in central Georgia during the winter which are favorable for the rapid expansion of bacterial canker.

P. syringae was re-isolated from all the Oct.-inoculated, from 86% of the Dec.-inoculated, and from 71% of the Feb.-inoculated trees (Table 1). However, natural infection apparently occurred in the block during the test, as indicated by isolation of P. syringae from 28 to 43% of the uninoculated trees. We expected some natural infection since bacterial canker is prevalent in the area. Similar results were reported by Crosse and Garrett (2) in field inoculations of cherry, although most of the infection by wild strains in their test occurred through leaf scars. We did not attempt to distinguish our FV-3 isolate from other strains of P. syringae in the orchard as FV-3 had been isolated from a diseased peach tree in this orchard. P. syringae was present in some trees with little or

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