

The spring  $k_s$  from both shade- and sun-exposed leaves were generally lower than the  $k_s$  from the more recently expanded leaves in the previous fall. Part of this difference was due to the natural "hardening off" of leaves during stomatal and cuticle development (1). The mid-day decrease in  $k_s$  of all trees in the fall was also apparent in the sun-exposed leaves during the next spring. Though more frequent hourly data would be illustrative, it appears that the lower  $\Psi_L$  was associated with the decreased  $k_s$  of blight affected and senescent leaves. It is possible that the restricted water movement through the plant may hasten natural leaf senescence. The decreased water movement and consequent decreased  $\Psi_L$  and  $k_s$  apparently result in canopy thinning but not in changes in specific leaf weight, leaf osmotic potentials, or in the critical leaf water deficits at which leaves wilt. Whether these responses are specific to the disorder known as blight or young tree decline or typical of citrus responses to water stress in general, remains a question.

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## Sampling Strategies for Estimates of Cluster Weight, Soluble Solids and Acidity of 'Concord' Grapes<sup>1</sup>

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**Abstract.** Variability of cluster weight, soluble solids, and titratable acidity of 'Concord' grapes (*Vitis labruscana* Bailey) was quantified with respect to several fixed and random factors. All 3 measurements were affected by cluster position while only cluster weight and soluble solids were affected by sunlight exposure. Estimates of variance components indicated that the greatest percentage variability was among vines. Variance components were used to examine efficient sampling plans that would detect a specified difference at a selected level of Type I error. Graphs illustrate various schemes of sample allocation which would achieve the desired level of precision.

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Variation from a number of sources is known to exist within grape vineyards. Differences in vine size, crop load, cluster position and exposure to sunlight can affect the sugar content of berries (8, 13) and the number and size of clusters (7). This variation is important for several reasons. Yield and ripeness estimates are frequently made in order to determine whether cultural options are economically feasible and whether the crop has ripened to the degree required by a particular processor. Research efforts are hindered when variation masks the difference between 2 applied treatments. Without a knowledge

of the magnitude and location of variation, a researcher makes mistakes in 2 ways (12): firstly, sample size may be too small to declare a difference statistically significant at the desired level of probability when, with a sample of proper size, significance would have been found; secondly, sample size may be larger than required, resulting in statistical precision greater than can be economically justified. Because significance tests involve error variation, reduction in error or random variation will make treatment differences easier to detect. Such error reduction can be achieved by adequate replication and appropriately chosen sampling plans (3).

Both researchers and individuals assessing field productivity or fruit quality require that samples be representative of the population from which they were taken; i.e., that they are accurate and repeatable. In order to efficiently obtain an unbiased sample, estimates of variation within the vineyard must be known. Statistical procedures which permit variation sources to be quantified have been used for sweet cherry characters (4), apple and peach characters (12), apple fruit maturity measurements (9) and peach wood hardness (2).

Each of these studies used some form of the analysis of variance to estimate variance components for the relevant sources of random variability and in some sense devised an optimum sampling plan. We have used similar procedures to examine sampling strategies for cluster weight, soluble solids and acidity of 'Concord' grapes.

### Materials and Methods

A 'Concord' vineyard in Lawton, Michigan trained to single-curtain, bilateral cordon on the top wire (Hudson River Umbrella) was utilized in 1977 and 1978. The vines were planted in 1905, have since undergone intermittent renewal. All vines had been balanced-pruned (30+10) for at least 3 years and some for as many as 8 years.

Several factors were investigated as sources of variability. Three vine size categories in 1977 and 4 in 1978 were chosen as measured by kg of cane prunings and within each category 6 vines in 1977 and 5 in 1978 were selected so that various locations within the 1 acre (0.4 ha) vineyard were represented. Average kg of cane prunings, numbers of buds yield, and number of clusters of all experimental vines are given in Table 1.

On each vine 8 shoots were selected, 4 from exterior positions judged to be well-exposed to sunlight and 4 from interior, shaded positions. In 1977 all sampled shoots carried at least 2 clusters, which were designated as base and second; third clusters, if present, were disregarded. In 1978 all sampled shoots had 3 clusters, the smallest cluster having no less than 10 berries.

Clusters were collected, stored at  $-20^{\circ}\text{C}$  and analyzed at a later date. The fruit of each cluster was thawed, weighed, homogenized in a Waring blender and centrifuged at 5000 rpm for 10 min. In 1978 clusters were weighed before being frozen. From the clarified juice, soluble solids was measured on a Bausche & Lomb Abbe-3L refractometer and titratable

acidity was measured and expressed as g of tartaric acid per 100 ml of juice (1).

An appropriate statistical model separated the variance components of interest and in this model an individual observation ( $Y_{ijklm}$ ) was described as follows:

$$Y_{ijklm} = \mu + P_i + V(P)_{(ij)} + E_k + (PE)_{jk} + EV(P)_{(ijk)} + S_{(ijk)l} + C_m + (CP)_{lm} + (CE)_{km} + (CPE)_{hkm} + \epsilon_{ijklm}$$

where:

- $\mu$  = overall mean
- $P_i$  = effect of  $i$ th vine size,  $i = 1, 2, \dots, p$
- $V(P)_{(ij)}$  = effect of  $j$ th vine within the  $i$ th size,  $j = 1, 2, \dots, v$
- $E_k$  = effect of  $k$ th exposure,  $k = 1, 2, \dots, e$
- $(PE)_{jk}$  = interaction of vine size and exposure
- $EV(P)_{(ijk)}$  = interaction of exposure and vine within vine size
- $S_{(ijk)l}$  = effect of  $l$ th shoot within exposure, vine and vine size,  $l = 1, 2, \dots, s$
- $C_m$  = effect of  $m$ th cluster,  $m = 1, 2, \dots, c$
- $(CP)_{lm}$ ,  $(CE)_{km}$  and  $(CPE)_{hkm}$  are interactions with cluster
- $\epsilon_{ijklm}$  = residual variability

and:

Vines, shoots and residuals were considered random, with all other effects fixed.

From the expected mean square generated for the allowable lines in the analysis of variance (11), variance components can be estimated (5, 6, 10). The analysis of variance table (Table 2) gives expected mean squares which can be related to observed mean squares (Table 3) for calculation of individual components of variance (Table 4).

### Results

Vine size had no significant effect on cluster weight, soluble solids, or acidity for either year (Table 5). In 1977 base clusters were heavier, higher in sugar, and lower in acidity than second clusters. In 1978 base and second clusters weighed the same but were both much heavier than third clusters. Base clusters were higher in sugar than third clusters while second clusters were intermediate but not significantly different from the other two. Second clusters were lowest in acidity while third clusters were highest. In both exterior clusters had greater soluble solids and in 1978 they were significantly heavier and lower in acid.

Estimates of individual variance components (Table 4) indicated that variation among vines contributed much more to the total variability of the 3 measurements with the exception of cluster weight in 1977. In 1977 the shoot component contributed more to the total variation of cluster wt than did the component  $EV(P)$  while in 1978 the reverse was true. The results for the 2 years have been presented separately, however, we consider the difference in variance component estimates to be well within the range expected due to random variability.

Estimates of individual variance components (Table 4) can also be used to maximize the efficiency of sampling. Our goal was to allocate the number of samples within the four levels of variability so that for a reasonable number of samples the desired accuracy could be attained. The method used here is similar to that used by Schultz and Schneider (12), and involves the equation:

$$d = t_{\alpha} s_d$$

where:

- $d$  = difference to be detected with power 0.5
- $t$  = Student's  $t$ , at  $\alpha$  level of probability

Table 1. Average per vine weight of prunings, number of buds retained, yield and number clusters of 'Concord', 1977 and 1978.

Vine size class	Wt of prunings (kg)		No. of buds		Yield (kg)		No. of clusters	
	1977	1978	1977	1978	1977	1978	1977	1978
1	0.5	0.5	31	31	7.7	10.5	59	95
2	1.1	1.0	42	41	12.5	11.3	102	116
3	1.5	1.4	50	50	12.5	14.4	98	134
4	—	1.9	—	63	—	17.3	—	163

Table 2. Analysis of variance showing expected mean squares used in calculating F values and variance components.

Source	df	Expected mean square
P	p-1	$\sigma^2 + c\sigma^2_S + c\sigma^2_{EV(P)} + c\sigma^2_{V(P)} + c\sigma^2_{evk^2_P}$
V(P)	p(v-1)	$\sigma^2 + c\sigma^2_S + c\sigma^2_{EV(P)} + c\sigma^2_{V(P)}$
E	e-1	$\sigma^2 + c\sigma^2_S + c\sigma^2_{EV(P)} + scv\sigma^2_{PE}$
PE	(e-1)(p-1)	$\sigma^2 + c\sigma^2_S + c\sigma^2_{EV(P)} + scv\sigma^2_{PE}$
EV(P)	p(e-1)(v-1)	$\sigma^2 + c\sigma^2_S + c\sigma^2_{EV(P)}$
S(PEV)	pev(s-1)	$\sigma^2 + c\sigma^2_S$
C	c-1	$\sigma^2 + pves\sigma^2_C$
CP	(c-1)(p-1)	$\sigma^2 + ves\sigma^2_{CP}$
CE	(c-1)(e-1)	$\sigma^2 + pvs\sigma^2_{CE}$
CEP	(c-1)(e-1)(p-1)	$\sigma^2 + vs\sigma^2_{CEP}$
Residual	pe(vs-1)(c-1)	$\sigma^2$

Table 3. Analysis of variance showing observed mean squares (MS) and significance probabilities ( $\alpha$ ) of all effects of interest for cluster weight, soluble solids, and acidity of 'Concord' grapes, 1977 and 1978.

Source	df		Cluster wt				Soluble solids				Acidity			
			1977		1978		1977		1978		1977		1978	
	1977	1978	MS	$\alpha$	MS	$\alpha$	MS	$\alpha$	MS	$\alpha$	MS	$\alpha$	MS	$\alpha$
P	2	3	7595	.36	4356	.84	25.6	.15	33.5	.21	.0059	.88	.1123	.25
V(P)	15	16	6852	—	15533	—	11.9	—	20.0	—	.0462	—	.0736	—
E	1	1	2404	.53	22468	.05	33.1	<.0005	313.2	<.0005	.0012	.82	.0482	.069
EP	2	3	1130	.83	2992	.63	0.6	.68	3.3	.78	.0408	.17	.0163	.314
EV(P)	15	16	5902	—	5004	—	1.6	—	8.8	—	.0202	—	.0127	—
S(PVE)	108	120	3244	—	3036	—	1.4	—	2.5	—	.0122	—	.0080	—
C	1	2	20608	<.0005	106080	<.0005	1.8	.006	3.0	<.0005	.0317	.004	.1270	<.0005
CP	2	6	1245	.38	5172	.001	0.0	.827	0.1	.88	.0029	.45	.0040	.51
CE	1	2	244	.67	728	.563	0.5	.161	0.6	.16	.0003	.79	.0086	.15
CPE	2	6	2686	.12	1045	.550	0.0	.979	0.2	.77	.0138	.024	.0048	.38
Residual	138	304	1259	—	1264	—	.2	—	0.3	—	.0036	—	.0045	—
Total	287	479	—	—	—	—	—	—	—	—	—	—	—	—

Table 4. Estimates of variance components of cluster weight, soluble solids, and acidity of 'Concord' grapes for 2 experiments, 1977 and 1978, and their percentage (in parenthesis) contribution to the total variation.

Variance component	df		Variance component measurements							
			Cluster wt (g)		Soluble solids (%)		Acidity (g/100 ml)			
	1977	1978	1977	1978	1977	1978	1977	1978	1977	1978
Vines	6	5	13.1 (18)	87.7 (68)	0.1078 (87)	0.0930 (56)	0.000271 (56)	0.000508 (83)		
EV(P)	12	10	20.7 (29)	16.4 (13)	0.0015 (1)	0.0529 (32)	0.000083 (17)	0.000039 (6)		
Shoots	48	40	27.7 (39)	14.8 (11)	0.0123 (10)	0.0179 (11)	0.000090 (19)	0.000028 (5)		
Residual	138	120	9.9 (14)	10.5 (8)	0.0024 (2)	0.0027 (2)	0.000038 (8)	0.000038 (6)		

and:

$$s_d = [2((\sigma^2_V/v) + (\sigma^2_{EV(P)/ev}) + (\sigma^2_{S/ev}) + (\sigma^2_{csev}))]^{1/2} \text{ is the standard error of a vine mean difference}$$

Clearly, a number of sampling arrangements can be used to give the same standard error ( $s_d$ ). We have examined the sampling options available to meet the requirements for selected sensitivity demands (d). Fig. 1, 2 and 3 show the minimum vine number — shoot number combinations that will meet the sensitivity requirements. In all cases we assumed that one

cluster would be randomly selected from exposure type and cluster position.

The desired differences in cluster wt to be detected (Fig. 1), 15 g represents a yield difference of  $1 \times 10^3$  kg/ha calculated on the basis of 110 clusters per vine and 570 vines per acre. The soluble solids difference of 0.5% (Fig. 2) is of economic significance (13) and would probably represent the maximum desired accuracy. Titratable acidity is not used as a quality criterion for 'Concord' grapes but is included here for completeness and the difference was arbitrarily selected.

Table 5. Effect of vine size, sunlight exposure and cluster position on cluster weight, soluble solids, and acidity of Concord grapes in Lawton, Michigan, 1977 and 1978.<sup>z</sup>

Variable	Cluster wt (g)		Soluble solids (%)		Acidity (g/100 ml)	
	1977	1978	1977	1978	1977	1978
<i>Vine size and class</i>						
1	140a	123a	16.6a	16.6a	.78a	.75a
2	156a	126a	15.7a	15.8a	.77a	.76a
3	155a	116a	16.7a	15.5a	.76a	.82a
4	--	113a	--	15.4a	--	.76a
<i>Exposure</i>						
Exterior	153a	127a	16.7a	16.6a	.77a	.76a
Interior	148a	113b	16.0b	15.0b	.77a	.78a
<i>Cluster position</i>						
Base	160a	133a	16.4a	16.0a	.76a	.77b
2nd	141b	136a	16.2b	15.8ab	.78b	.75c
3rd	--	90b	--	15.7b	--	80a

<sup>z</sup>Within each main effect, means followed by the same letter are not significantly different ( $P = 5\%$ ) separated by Duncan's multiple range test.

### Discussion

The fixed effects of exposure and cluster position on cluster weight, soluble solids, and acidity are in general agreement with those found by Partridge (8) and Shaulis (13) where data are available for comparison. Although vine size has no significant effect on soluble solids the trend of lower sugar for larger vines is also similar to that found by Partridge (8) and Shaulis (13).

Differences between years for the variance components (Table 4) could be due, in part, to the use of different vines in the 2 experiments. The second-year vines within a size class were more uniform. Also, the second year, base clusters were not different from second clusters, as they were the first year. While there is no data to indicate why this occurred, a rainy period which occurred during the first days of bloom of base clusters in 1978 may have reduced pollination (14) and subsequent fruit set.

In these experiments, whole clusters from vines were sampled in order to gain information about cluster weight variation. This permitted collection of sample clusters and from their

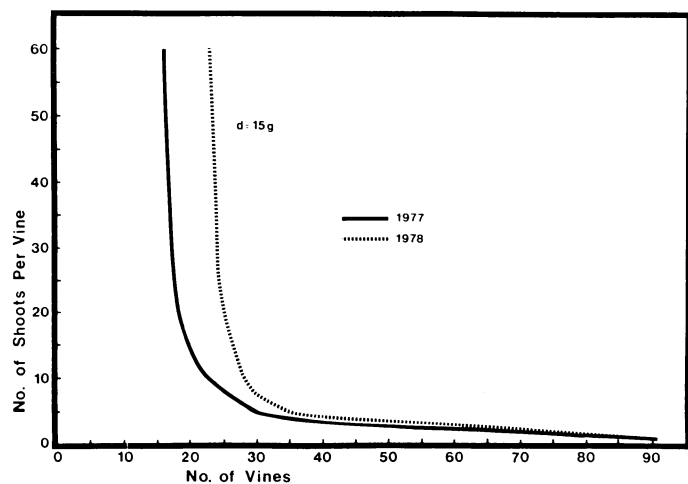


Fig. 1. Combination of vines and samples per vine necessary to declare a cluster wt difference (d) statistically significant as a specified level of probability ( $\alpha$ ) with power of 0.5.

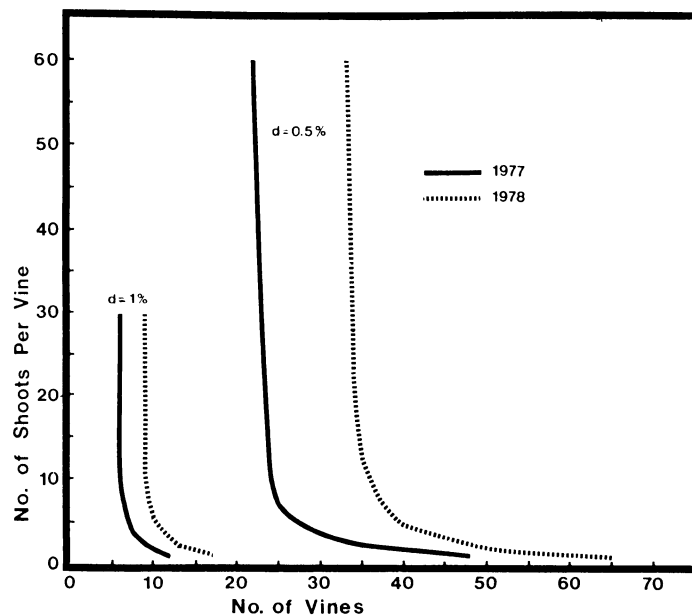


Fig. 2. Combinations of vines and samples per vine necessary to declare a soluble solids difference (d) statistically significant at a specified level of probability ( $\alpha$ ) with power of 0.5.

weight, prediction of the yield of vines of known cluster numbers. If one wishes information about only soluble solids and acidity, the apical 4 berries accurately predict the soluble solids of clusters while greatly reducing the amount of material in the sample (13).

For the sampling combinations in Fig. 1, 2, and 3, exposure and cluster numbers were held at 1 and considered to be randomly selected. Random selection of material is difficult but important for unbiased estimates. Selecting shoots equally from each of the 2 exposure categories would assume that in the population there are equal numbers of exposed and shaded shoots. The same is true for selection of equal numbers of base, second and third clusters. *A priori* knowledge of the numbers of

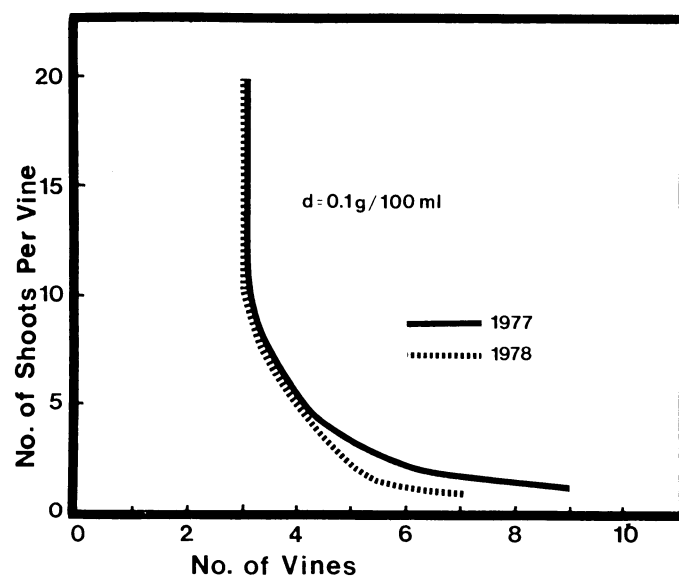


Fig. 3. Combinations of vines and samples per vine necessary to declare an acidity difference (d) statistically significant at a specified level of probability ( $\alpha$ ) with power of 0.5.

exposed and shaded shoots and base, second and third clusters is necessary in order to sample them in proportion to their presence in the population. Since this is not known we opted for random selection of exposure and cluster categories.

Cluster weight, soluble solids, and acidity, respectively, can be estimated by growers by randomly selecting shoots and vines in numerical combinations indicated by the curves of Fig. 1, 2, and 3. Because of large variability among vines there is a minimum number of vines which must be sampled to gain an accurate estimate of any measurement. Sampling smaller numbers of vines would not provide an estimate of desired accuracy even if every cluster were analyzed (12). In situations where growers have not balanced-pruned, more vines must be sampled to offset higher variability among vines.

The sampling combinations for all measurements which result in the fewest total samples is that employing 1 shoot on each of the appropriate numbers of vines. This assumes that all shoots and vines are randomly selected from the vineyard of interest or that vines are selected so that areas of the vineyard which are known to vary in slope, aspect, vine vigor, soil type, etc. are represented. It also assumes that the cost per unit samples is the same regardless of the combinations of vines and shoots (10).

Sampling for researchers is more complex. In grape research experiments, vines are balanced-pruned and harvested individually so that the earlier assumption of equal cost per unit of sampling does not apply. Here fewer vines and more shoots sampled per vine reduces the overall work in the experiment. Because vines are harvested individually (i.e., completely sampled for cluster variation) and because accurate acidity measurements require so few samples, soluble solids measurement becomes the limiting factor. Based on the data in Fig. 2, our recommendation is to sample between 24 and 36 vines per treatment combination (based on 1977 and 1978 data respectively) and 10 shoots per vine. This would provide the desired accuracy of 0.5% soluble solids while keeping the experiment small enough to be manageable. These vines would most effectively be arranged in 4-6 replicates of 6 vines each to avoid difficulties of missing data in case single vines are lost.

The data presented here provide strategies for estimating cluster weight, soluble solids, and acidity of 'Concord' grapes useful to industry and researchers. While they provide a background for sampling other grape cultivars, more research is needed to verify their applicability.

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## Calcium Deficiency of *Anthurium andreanum* Lind. Spathes<sup>1</sup>

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**Abstract.** Color breakdown of spathe tissue of anthurium, *Anthurium andreanum* Lind., exhibited typical calcium deficiency symptoms. Anatomical studies revealed cell separation and collapse in affected tissue. Cell separation may have resulted from instability of the middle lamella due to calcium deficiency. Calcium application in the field, either as nitrate or silicate, significantly reduced the incidence of the disorder.

Anthurium is one of Hawaii's principle ornamental exports.

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The long vase life and ease of handling and packaging make it a durable product for shipping throughout the world. The flower consists of a large colorful spathe and a spadix protruding from the base of the spathe.

One of the serious problems facing growers is the appearance of water soaked lesions which eventually discolor the lobes of the spathe. These lesions turn dark brown and eventually dry out, rendering the flowers unsuitable for sale. Preharvest losses of 50% have been reported, while losses after shipments to the overseas markets have been as high as 20%. Visual symptoms may not be evident during grading and packing, but appear after