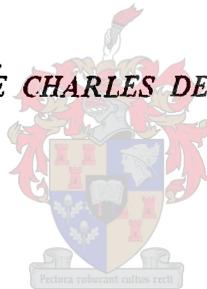


The Effect of Summer Pruning on Growth and Grape Composition of
Vitis vinifera L. cv. Cape Riesling.

ANDRÉ CHARLES DE LA HARPE



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Promoter: Prof. Dr. J.H. Visser

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INTRODUCTION

Topping is a summer pruning viticultural practice and is commonly used in South Africa as a practical manipulation to enable the producer to enter rows of vigorously growing vines. In some cases, however, for example Muscat d'Alexandrie, topping is used to improve berry set. Topping is also done to minimize wind damage and to create more uniform vines (Theron, 1944).

This practice comprises the removal of the apical 25 cm or more of the shoot apex. The rationale behind topping is a displacement of translocation sinks. The sinks for photosynthetate are normally considered to be the apical meristem, bunches, stem and roots. Competition between bunches and other sinks could easily result in altered quality of the grapes.

Thus topping is a manipulation to effect the translocation of photosynthetate in the vine and is of importance in practical viticulture due to the fact that the translocation of photosynthetate plays an important role in the quantitative and qualitative composition of the bunch.

The import and export of photosynthetate in the lateral shoots of vines are affected by climatic conditions (Koblet, 1977). Results obtained from the northern hemisphere showed that juvenile leaves of lateral shoots are sinks and remain sinks until at least two leaves reach maturity (Hale & Weaver, 1962; Koblet, 1977). The lateral shoots will then start exporting photosynthetate. According to Koblet & Perret (1971; 1972) only basipetal translocation of photosynthetate occurs and lateral shoots export their products to surrounding bunches. However, it is important to know how the translocation pattern of photosynthetate is affected by South African conditions before any recommendation can be made regarding the advantages or disadvantages of topping.

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CHAPTER 2

The Determination of the Homogeneity of a Vitis vinifera L. cv. Cape Riesling
Vineyard

ABSTRACT

The value of Principal Component and Stepwise Discriminant analyses in selecting uniform plants for experimental purposes is discussed. Twenty-seven variables were taken into account to establish the homogeneity (uniform plants) of 297 Vitis vinifera L. cv. Cape Riesling vines. A detailed study of the relationship and interrelationship of these variables resulted in 208 vines being selected as an uniform population. This selection provides the possibility for the researcher in viticulture to use single vines as experimental units, but it must be pointed out that Principal Component and Stepwise Discriminant analyses can only be used as an aid to normal statistical evaluation of experimental results and not as substitute for statistical experimental design.

INTRODUCTION

The main statistical tools in compensating for variability are replication, randomisation and blocking (Hammer, 1981). Replication normally involves multiple experimental units and together with randomisation it results in valid estimates of the experimental error (variance). Biological variation can be decreased by selecting more uniform plants at the pretreatment stage and then using replication and randomisation for treatment applications (Hammer, 1981). According to Hammer (1981) this will allow the scientist to detect differences between treatments with fewer replications. The complexity of biological material, with intercorrelating variables, has as result that single variables cannot be treated as independent components of a factor (Broschat, 1979).

The problem of identification of uniform plants before treatments are applied could therefore be solved by measuring the appropriate variables and subsequently performing a Principal Component analysis (PCA) decreasing the dimensionality of the data.

PCA has been successfully used in psychology (Hotelling, 1936) and in the biological and horticultural sciences for a number of years (Orlocki, 1967; Sneath & Sotol, 1973; Glendon & Staby, 1976; Oliver, Siddiqi & Goward, 1978; Leegwater & Leegwater, 1981 ; Van Rooyen & Tromp, 1982).

The purpose of this study was to select relatively uniform vines in a Vitis vinifera L. cv.Cape Riesling vineyard by means of different growth and quality parameters with the aid of Stepwise Discriminant analysis (SDA) and PCA in order to decrease the large number of vines per treatment needed for physiological studies on this specific vineyard. The relatively small number of

plants in the vineyard made the normal randomised block design with a large number of experimental units per replication impossible.

MATERIAL AND METHODS

Experimental vineyard: A 15 year old vineyard consisting of 297 vines of *V. vinifera* cv. Cape Riesling grafted onto 99 Richter, planted in a vineyard consisting of four soil types namely a Southwold, Avalon, Glencoe and Kanonkop series (soil series as described by Macvicar, C.N. & Soil Survey Staff, 1977) on the experimental farm, Nietvoorbij, Stellenbosch, South Africa was used in this study. The vines were trellised on a Perold system (Zaeman, 1981) and spur pruned to 16 buds per kilogram shoot mass. Rainfall was supplemented by two 200 mm irrigations during November 1981 and January 1982, respectively.

Variables: The investigation was executed in two phases. In phase I, the 22 growth variables depicted in Table 1 were measured on two shoots per cordon and the mean of these measurements were used as data points. The leaf area of a vine was determined by measuring the area of individual leaves with a model LI-3000 Li-Cor Portable Area Meter and summated. Leaf dry mass was determined after drying to constant mass at 80°C. The vines were visually evaluated by five judges into three categories: sick and poorly developed vines taken as 100; normally developed vines as 500 and well developed vines as 900. All measurements were carried out at harvesting time.

TABLE 1

Variables measured in a *Vitis vinifera* L. cv. Cape Riesling vineyard

<u>Variable Number</u>	<u>Variables</u>	<u>Unit</u>
<u>Phase I</u>		
1.	Shoot length cordon 1	cm shoot. cordon 1 ⁻¹
2.	Shoot length cordon 2	cm shoot. cordon 2 ⁻¹
3.	Total shoot length of both cordons	cm shoot. cordons ⁻¹
4.	Spurs cordon 1	
5.	Spurs cordon 2	
6.	Spurs per vine	
7.	Number of leaves per shoot of cordon 1	*
8.	Number of leaves per shoot of cordon 2	**
9.	Total number of leaves of the shoot of variables 7 and 8	
10.	Total leaf area per shoot of cordon 1	*cm ² . shoot ⁻¹
11.	Total leaf area per shoot of cordon 2	**cm ² . shoot ⁻¹
12.	Total leaf area of both shoots	cm ² . shoots ⁻¹
13.	Mean area per leaf of the shoots of cordon 1	*cm ² . leaf ⁻¹
14.	Mean area per leaf of the shoots of cordon 2	**cm ² . leaf ⁻¹
15.	Total mean area per leaf of both shoots	cm ² . leaf ⁻¹
16.	Total leaf dry mass per shoot of cordon 1	*g. total leaf number ⁻¹
17.	Total leaf dry mass per shoot of cordon 2	**g. total leaf number ⁻¹
18.	Total leaf dry mass of both shoots	g. total leaf number ⁻¹
19.	Mean dry mass per leaf of the shoots of cordon 1	*g. leaf ⁻¹
20.	Mean dry mass per leaf of the shoots of cordon 2	g. leaf ⁻¹
21.	Total mean dry mass per leaf of the shoots of both cordons	g. leaf ⁻¹
22.	Evaluation of the vines	
<u>Phase II</u>		
Phase I plus the following 5 variables		
23.	Total soluble solids of must	°B
24.	Total titratable acids of must	g.dm ⁻³
25.	pH	log (H ⁺)
26.	Yield per vine	Kg
27.	Number of bunches per vine	

* Mean of the two shoots of the 2nd spur on cordon 1

** Mean of the two shoots of the 2nd spur on cordon 2

In phase II five quality variables were measured additionally, namely total soluble must solids in ^oBalling (°B), pH, total titratable must acidity (TTA) (g.dm⁻³), as well as the total number of bunches per vine and yield per vine, as practised in the laboratories of the VORI (Anon, 1981).

Data processing: The data was processed using a BMD-07M SDA programme (Health Sciences Computing Facility, UCLA) and a PCA programme forming part of the pattern recognition system "Arthur" (Harper, Duerer & Kowalski, 1977). The subroutines used in the "Arthur" programme are listed in Table 2.

TABLE 2

Programme of Arthur as performed on the data set

<u>Programme</u>	<u>Programme function</u>
<u>Phase I</u>	
Input	Creates a data matrix as output to a binary file that is compatible with all other routines in Arthur.
Utilit	Provides a line printer listing of the data matrix and/or the distance matrix.
Scale	Scales the data to some proportions. The scaling factors are derived from the u data vectors of the training set and applied to all the data.
Correl	Calculates all feature - feature and feature - property covariances and correlations.
Kaprin	The extraction of the eigenvalues and eigenvectors of the data dispersion matrix as performed.
Katran	Creates a new data matrix from the first K factors of the data.
Varvar	Produces line printer plots of a data matrix.
Kaprin-Kavari-Katran-Varvar	Perform a principal component analysis plus rotation of eigenvalues with plotting.
<u>Phase II</u>	
Input	Same as phase I
Scale	Same as phase I
Kaprin	Same as phase I
Katran	Same as phase I
Varvar	Same as phase I
Kavari	Executes a Varimax rotation on the eigenvectors.
Katran	As phase I but with Kavari results.
Varvar	Same as phase I

The BMD-07M programme was executed on a Burroughs 7800 computer of the Department of Agriculture and the "Arthur" programme on a Univac 1100 computer of the University of Stellenbosch.

Prior to PCA all the data were scaled to a standard deviation (S_i) of one and zero mean. The normalised standard deviation is defined as

$$S_i = \frac{\sigma_i}{\bar{x}_i}$$

where σ = standard deviation

\bar{x}_i = weighted mean

$$\text{and } \bar{x} = \frac{\sum_{j=1}^n \left[\frac{x_{i,j}}{u_{i,j}^2} \right]}{\sum_{j=1}^n \left[\frac{1}{u_{i,j}^2} \right]}$$

where $u_{i,j}$ is the uncertainty associated with the feature $x_{i,j}$

$$x_{i,j} = \frac{[x_{i,j} - \bar{x}_{i,j}]}{\left[\sum_{j=1}^n [x_{i,j} - \bar{x}_{i,j}]^2 \right]^{1/2}}$$

and where n = total number of data vectors in the training data set, and x is the i th feature associated with the j th data vector.

RESULTS AND DISCUSSION

Phase I: Table 3 represents the scaled data with the mean, standard deviation, normalised standard deviation as previously defined as well as minimum and maximum values.

Three of the PCA factors have eigenvalues (the sum of the variances) greater than one and are retained for discussion (Table 4). They account for 65% of the variance in the original variables with the remaining 35% caused by random variation.

TABLE 3

The scaled data of phase I with the mean, standard deviation, normalised standard deviation and minimum and maximum values

Variable Number	Variables	Mean	Standard Deviation	Normalised Std. Deviation	Minimum	Maximum
<u>Phase I</u>						
1.	Shoot length cordon 1	140,80	96,31	0,68	0,00	514,00
2.	Shoot length cordon 2	128,50	92,07	0,72	0,00	609,00
3.	Total shoot length of both cordons	266,40	146,30	0,55	0,00	768,00
4.	Spurs per cordon 1	2,70	1,07	0,39	0,00	6,00
5.	Spurs per cordon 2	2,73	1,03	0,39	1,00	5,00
6.	Spurs per vine	5,43	1,59	0,29	0,00	10,00
7.	Number of leaves per shoot of cordon 1	24,76	15,16	0,61	0,00	64,00
8.	Number of leaves per shoot of cordon 2	19,51	14,51	0,74	0,00	59,00
9.	Total number of leaves of the shoots of variables 7 and 8	44,42	21,54	0,48	0,00	102,00
10.	Total leaf area per shoot of cordon 1	132,00	952,60	0,68	0,00	6176,00
11.	Total leaf area per shoot of cordon 2	114,00	967,60	0,76	0,00	3933,00
12.	Total leaf area of both shoots	2508,00	1309,00	0,52	0,00	2966,00
13.	Mean area per leaf of the shoots of cordon 1	48,99	23,95	0,49	0,00	204,50
14.	Mean area per leaf of the shoots of cordon 2	46,13	27,35	0,59	0,00	171,50
15.	Total mean area per leaf of both shoots	57,79	18,16	0,34	0,00	146,80
16.	Total leaf dry mass per shoot of cordon 1	7,05	4,89	0,69	0,00	32,41
17.	Total leaf dry mass per shoot of cordon 2	5,82	4,52	0,78	0,00	20,67
18.	Total leaf dry mass of both shoots	12,05	6,08	0,53	0,00	37,81
19.	Mean dry mass per leaf of the shoots of cordon 1	0,78	0,50	0,22	0,00	2000,00
20.	Mean dry mass per leaf of the shoots of cordon 2	0,24	0,13	0,57	0,00	0,49
21.	Total average dry mass per leaf of the shoots of both cordons	0,27	0,09	0,32	0,00	0,53
22.	Evaluation of the vines	2537,00	655,20	0,26	1000,00	3000,00

* Discrepancies in the data set are attributable to computer rounding off.

TABLE 4

Factor loadings for the first 3 Eigenvalues for 22 variables

(Programmes used: Input, Utilit, Scale, Correl, Kaprin, Katran, Varvar)

<u>Variable Number</u>	<u>Variables</u>	<u>Factor 1</u>	<u>Factor 2</u>	<u>Factor 3</u>
<u>Phase I</u>				
1.	Shoot length cordon 1	-0,1319	-0,0187	+0,0198
2.	Shoot length cordon 2	-0,1658	-0,0462	-0,0915
3.	Total shoot length of both cordons	-0,2239	-0,0428	-0,0689
4.	Spurs per cordon 1	-0,1167	+0,0043	-0,3900
5.	Spurs per cordon 2	-0,1364	-0,0837	-0,3501
6.	Spurs per vine	-0,1672	-0,0516	-0,4901
7.	Number of leaves per shoot of cordon 1	-0,1972	+0,3836	+0,1089
8.	Number of leaves per shoot of cordon 2	-0,2567	-0,2877	+0,2254
9.	Total number of leaves of the shoots of variables 7 and 8	-0,2976	+0,0677	+0,2146
10.	Total leaf area per shoot of cordon 1	-0,2061	+0,4049	+0,0395
11.	Total leaf area per shoot of cordon 2	-0,2527	+0,2991	+0,2031
12.	Total leaf area of both shoots	-0,3138	+0,0975	+0,1621
13.	Mean area per leaf of the shoots of cordon 1	-0,2010	+0,2236	-0,1987
14.	Mean area per leaf of the shoots of cordon 2	-0,2151	-0,2868	-0,0802
15.	Total mean area per leaf of both shoots	-0,2190	+0,0714	-0,2584
16.	Total leaf dry mass per shoot of cordon 1	-0,2050	+0,3928	+0,1050
17.	Total leaf dry mass per shoot of cordon 2	-0,2480	+0,3004	+0,2098
18.	Total leaf dry mass of both shoots	-0,3039	+0,0819	+0,2119
19.	Mean dry mass per leaf of the shoots of cordon 1	-0,0046	-0,0614	+0,1666
20.	Mean dry mass per leaf of the shoots of cordon 2	-0,2204	-0,3061	-0,0625
21.	Total mean dry mass per leaf of the shoots of both cordons	-0,2349	+0,0494	-0,1882
22.	Evaluation of the vines	-0,0647	-0,0104	-0,1187
<u>Eigenvalues</u>		8,7	3,5	2,1
Factor percentage responsible for variance		39,6	15,8	9,5
Cumulative percentage of variance		39,6	55,4	64,9

The first PCA factor with an eigenvalue of 8,7 account for 39,6% of the variance of the original variables. This factor has relatively high factor loadings on the total number of leaves, leaf area and leaf dry mass of all measured shoots indicating that leaf canopy variables dominate this factor. Factor 2 has an eigenvalue of 3,5 explaining 15,8% of the total variance. The variables with the highest factor loadings are the number of leaves per shoot of cordon 1, the total leaf area and the total leaf dry mass of cordon 1 which is also leaf canopy variables respectively.

Figure 1 represents a plot of factor 1 (representing mainly total leaf canopy) against factor 2 (representing mainly total leaf area). From this plot it can be deduced that the vineyard consists of two groups of vines, separated mainly by factor 2.

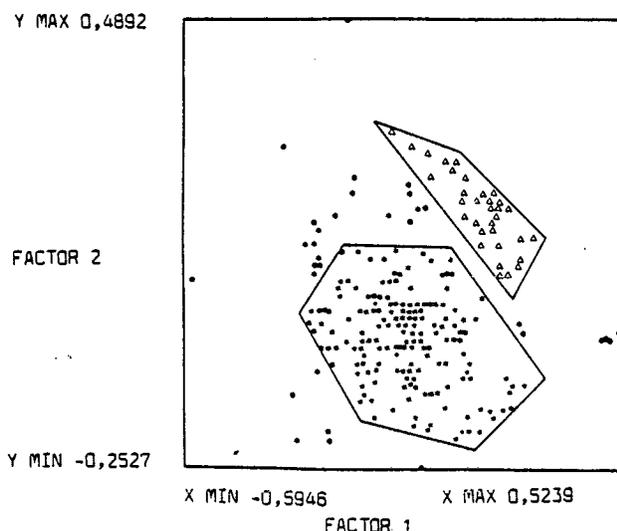


Fig. 1. PCA of 297 vines with 22 variables of a *Vitis vinifera* L. cv. Cape Riesling vineyard. Factor loadings for growth components for PCA I and II (* vines considered homogeneous; • and Δ vines considered to be heterogeneous to the homogeneous group).

Factor 3 has an eigenvalue of 2,1 representing 9,5% of the variance in the original variables. The highest loadings in this factor are the spur variables (Table 4). This may be interpreted as being a general growth factor or component.

In Fig. 2 factor 1 (X-axis) and factor 3 (Y-axis) are plotted. It is evident that the leaf canopy factor (factor 1) correlates with the growth factor (factor 3) and that the grouping of the vines is well defined. In Fig. 3 the leaf

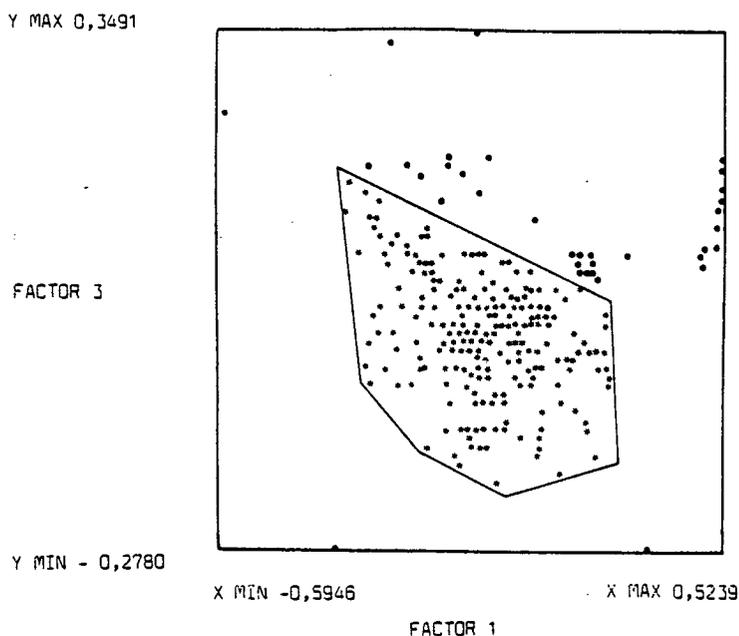


Fig. 2. PCA of 297 vines with 22 variables of a *Vitis vinifera* L. cv. Cape Riesling vineyard. Factor loadings for growth and leaf canopy components for PCA I and III (* homogeneous and • heterogeneous).

canopy of cordon 1 (factor 2, X-axis) is plotted against the growth factor (factor 3, Y-axis). Once again the vines seem to be well grouped into clusters indicating uniform vines as far as the leaf covering and other growth parameters are concerned. A further indication of the grouping is given in the totals on the Y-axis showing the total of plotted vines on the 2-dimensional plane.

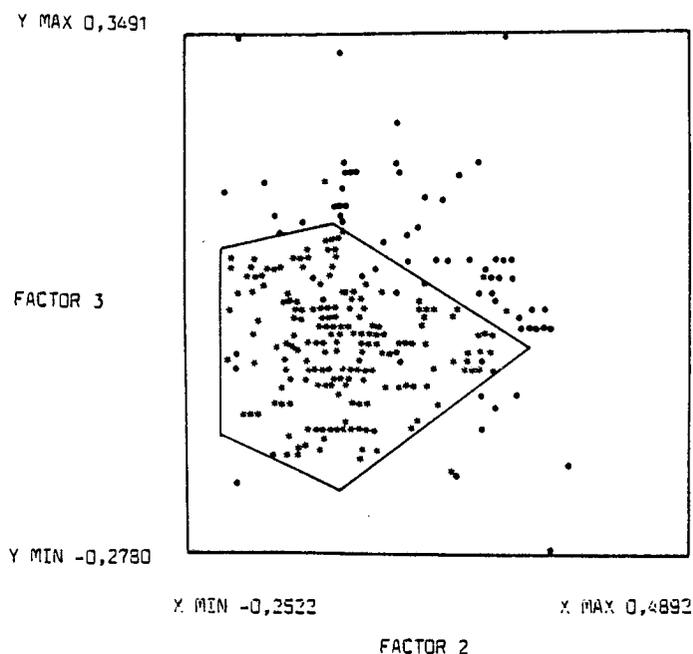


Fig. 3. PCA of 297 vines with 22 variables of a *Vitis vinifera* L. cv. Cape Riesling vineyard. Factor loadings for leaf canopy and growth components for PCA II and III (+ homogeneous and • heterogeneous).

Outlying vines, which are not considered part of the clusters, were eliminated from further experimentation. These are the vines where the relative distance between any two vines is too large in relation to the average distance of the other vines to one another. The assessment of the distances is a subjective choice of the authors and this may lead to criticism as far as the objectivity is concerned. However, it must be kept in mind, that this grouping was done to get an indication of the homogeneity of the data set and it gives the researcher sufficient scope to use his own initiative as far as the choice of experimental units goes. Emphasis must be placed on the fact that this is not a statistical analysis for each variable alone but an analysis for the complete set of variables.

After the vines were classed into a homogeneous group A (the 245 vines considered in the cluster) and a heterogeneous group B (the 52 vines not considered part of the cluster) a SDA was performed on the data set of groups A and B. The

means and standard deviation of the variables for the two groups are given in Table 5.

The results of the SDA showed that 208 of the original 245 vines considered to be homogeneous (85%) could be retained as category A vines while 34 of the original 52 vines considered to be heterogeneous vines (65%) were retained in category B (Fig. 4). Although the percentage grouping for category B is low, the vines excluded from this group had not been taken into consideration for category A because of the relatively large distances between these vines and those vines of category A. This low percentage may be because of some unexplained variance in the data set. After establishing the homogeneous group of vines (A) another SDA was performed on the data, this time classing the vines according to the four soil types. Table 6 gives the means and standard deviation of the 22 growth parameters.

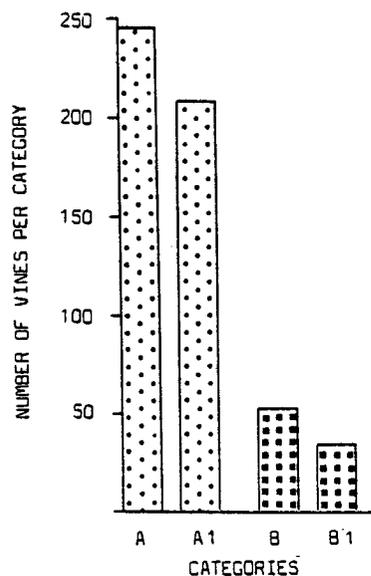


Fig. 4. Vines grouped into homogeneous and heterogeneous categories by PCA and SDA techniques. (A = vines selected by PCA as homogeneous; A1 = vines selected by SDA as homogeneous; B = heterogeneous vines; B1 = heterogeneous vines grouped by SDA).

From the Southwold series 58 vines (73%), the Avalon series 102 vines (79%), the Glencoe series 44 vines (80%) and the Karonkop series 41 vines (75%) were

TABLE 5

The means and standard deviation of the two categories as classified by stepwise discriminant analysis

Variable Number	Variables	Means		Grand Means over Categories	Standard Deviation		
		Category A	Category B		Category A	Category B	
Phase I							
1.	Shoot length cordon 1	140,04	165,56	145,09	36,30	124,30	
2.	Shoot length cordon 2	128,50	169,07	135,47	92,06	142,20	
3.	Total shoot length of both cordons	266,41	338,56	278,80	146,31	219,99	
4.	Spurs per cordon 1	2,70	2,47	2,66	1,06	1,27	
5.	Spurs per cordon 2	2,73	2,49	2,69	1,03	1,36	
6.	Spurs per vine	5,43	4,96	5,35	1,58	2,11	
7.	Number of leaves per shoot of cordon 1	24,75	28,45	25,39	15,16	23,79	
8.	Number of leaves per shoot of cordon 2	19,50	25,49	20,53	14,51	24,00	
9.	Total number of leaves of the shoots of variables 7 and 8	44,41	53,94	46,05	21,53	31,57	
10.	Total leaf area per shoot of cordon 1	1 392,20	1 851,02	1 470,99	992,56	1 711,11	
11.	Total leaf area per shoot of cordon 2	1 124,31	1 777,97	1 244,84	867,59	1 665,91	
12.	Total leaf area of both shoots	2 507,85	3 585,67	2 692,93	1 309,05	2 267,65	
13.	Mean area per leaf of the shoots of cordon 1	18,98	47,79	48,78	23,94	30,97	
14.	Mean area per leaf of the shoots of cordon 2	16,13	56,28	47,87	27,34	71,16	
15.	Total mean area per leaf of both shoots	23,78	62,11	55,21	18,15	30,77	
16.	Total leaf dry mass per shoot of cordon 1	7,04	9,70	7,43	4,88	8,25	
17.	Total leaf dry mass per shoot of cordon 2	5,82	8,61	6,30	4,51	8,67	
18.	Total leaf dry mass of both shoots	12,84	17,92	13,72	6,00	11,24	
19.	Mean dry mass per leaf of the shoots of cordon 1	8,37	6,24	6,98	127,52	0,14	
20.	Mean dry mass per leaf of the shoots of cordon 2	6,23	6,22	6,23	6,13	0,17	
21.	Total mean dry mass per leaf of the shoots of both cordons	9,27	6,30	6,27	0,08	0,10	
22.	Evaluation of the vines	2 526,58	2 392,15	2 511,78	665,19	723,28	

* Discrepancies in the data set are attributable to computer rounding off.

** Category A homogeneous group

*** Category B heterogeneous group

TABLE 6
 The means and standard deviations of the five categories of vines as defined by the four cell types and heterogeneous group as pointed out by PCA in which they are growing as classified by stepwise discriminant analysis.

Variable Number	Variable	Mean					Grand Mean of Category	Standard Deviation				
		1	2	3	4	5		A	B	C	D	E
Phase I												
1.	Shoot length cordon 1	211,04	133,80	137,29	87,18	165,56	145,09	107,57	89,68	75,44	76,78	124,28
2.	Shoot length cordon 2	109,66	116,48	110,69	77,92	109,67	135,17	129,31	61,24	64,63	87,81	112,28
3.	Total shoot length of both cordons	321,20	250,28	279,98	154,23	279,23	279,23	167,57	107,91	101,91	129,87	219,99
4.	Spurs per cordon 1	2,90	2,75	2,91	2,10	2,47	2,66	1,16	0,92	0,94	1,95	1,27
5.	Spurs per cordon 2	2,62	3,22	2,62	1,90	2,49	2,69	0,92	0,68	0,76	1,03	1,36
6.	Spurs per vine	5,60	6,07	5,55	4,06	4,96	5,35	1,48	1,28	1,37	1,42	2,11
7.	Number of leaves per shoot of cordon 1	25,08	26,53	21,55	24,92	28,46	25,29	15,71	13,25	12,23	19,73	23,79
8.	Number of leaves per shoot of cordon 2	20,88	20,45	26,87	7,92	25,10	20,53	14,26	12,01	14,28	12,28	24,00
9.	Total number of leaves of the shoots of varieties 7 and 8	46,32	47,04	48,43	32,86	53,56	46,05	27,47	19,27	19,70	24,15	31,57
10.	Total leaf area per shoot of cordon 1	513,41	1 406,07	1 172,51	1 560,27	1 851,02	1 470,09	1 151,98	754,25	715,96	232,49	1 711,11
11.	Total leaf area per shoot of cordon 2	263,37	1 120,78	1 625,16	422,86	1 777,97	1 244,34	875,72	728,73	829,73	600,35	1 665,91
12.	Total leaf area of both shoots	776,78	2 486,87	2 797,66	1 983,13	3 505,07	2 692,33	1 429,42	1 694,08	993,07	495,00	2 267,65
13.	Mean area per leaf of the shoots of cordon 1	49,96	51,11	51,69	41,33	47,79	48,79	33,95	17,46	15,72	20,39	30,87
14.	Mean area per leaf of the shoots of cordon 2	51,32	51,10	56,27	26,42	56,28	47,87	25,63	32,27	20,00	28,02	71,16
15.	Total mean area per leaf of both shoots	57,99	53,59	56,99	46,20	62,11	55,21	30,77	13,66	12,16	25,91	30,77
16.	Total leaf dry mass per shoot of cordon 1	6,96	7,11	6,29	7,87	9,29	7,43	4,89	4,01	3,74	6,82	8,25
17.	Total leaf dry mass per shoot of cordon 2	6,16	5,22	8,21	2,26	8,61	6,30	4,35	3,65	4,61	3,55	8,67
18.	Total leaf dry mass per shoot of both shoots	13,04	12,84	15,00	10,15	17,92	13,71	6,72	6,17	6,17	8,14	11,24
19.	Mean dry mass per leaf of the shoots of cordon 1	0,23	0,25	0,25	0,22	0,24	0,23	0,12	0,17	0,17	0,15	0,14
20.	Mean dry mass per leaf of the shoots of cordon 2	0,25	0,25	0,30	0,10	0,22	0,23	0,12	0,19	0,10	0,14	0,17
21.	Total mean dry mass per leaf of the shoots of both cordons	0,27	0,26	0,29	0,23	0,30	0,27	0,18	0,14	0,06	0,13	0,10
22.	Evaluation of the vines	466,00	2 454,54	2 689,65	2 586,00	2 393,10	2 511,76	761,57	641,64	560,37	641,74	727,28

1: Category A: Southwold series
 2: Category B: Avalon series
 3: Category C: Otencoe series
 4: Category D: Kananak series
 5: Category E: Heterogeneous group

Standard deviations in the data set are attributable to computer rounding off.

selected to be part of the homogeneous group showing that in this specific vineyard the four soil types had little or no effect on the growth parameters of the vines during this season.

Phase II: As a supplement to the existing data, five additional parameters including some grape quality parameters were determined. The Arthur programme was used on the data set including the five additional parameters and the results are listed in Tables 7 and 8.

Seven of the PCA factors have eigenvalues greater than one and were retained in the analysis. They account for 100% of the variance in the original variables. After the data was rotated by the Varimax rotation algorithm KAVARI, the first PCA factor explains 24,8% of the variance of the original variables (Table 8). This factor has relatively high factor loadings on leaf canopy (surface) and growth variables such as total number of leaves-, total leaf area per shoot on both cordons, similar to factor 1 in phase I where leaf cover and growth variables played an important role in the clustering of the vines. Factor 2 has an eigenvalue of 4,97 and explains 23,6% of the total variance. The variables with the highest factor loadings are total leaf area of the shoots on cordon 2, the total dry leaf mass of the shoots on cordon 2 and the average leaf mass per leaf of the shoots on cordon 2. This factor may therefore be interpreted to be relating to leaf cover in general and to growth parameters of the vines.

Fig. 5 represents the plot of the total leaf cover (factor 1, X-axis) to the total leaf cover of cordon 2 shoots (factor 2, Y-axis). In this plot the two

TABLE 7

The scale data of phase I & II with the mean, standard deviation, normalised standard deviation, and minimum and maximum values

Variable Number	Variables	Mean	Standard Deviation	Normalised Std. Deviation	Minimum	Maximum
Phase I						
1.	Shoot length cordon 1	146,00	104,10	0,71	0,00	514,00
2.	Shoot length cordon 2	134,70	101,20	0,75	0,00	664,00
3.	Total shoot length of both cordons	278,90	162,10	0,58	0,00	769,00
4.	Spurs per cordon 1	2,66	1,11	0,42	0,00	6,00
5.	Spurs per cordon 2	2,70	1,09	0,40	0,00	5,00
6.	Spurs per vine	5,36	1,70	0,32	0,00	10,00
7.	Number of leaves per shoot of cordon 1	25,51	16,99	0,67	0,00	93,00
8.	Number of leaves per shoot of cordon 2	20,39	16,22	0,79	0,00	72,00
9.	Total number of leaves of the shoots of variables 7 and 8	46,03	23,43	0,51	0,00	117,00
10.	Total leaf area per shoot of cordon 1	420,00	1 120,00	0,76	0,00	6 340,00
11.	Total leaf area per shoot of cordon 2	1 237,00	1 040,00	0,84	0,00	5 956,00
12.	Total leaf area of both shoots	2 693,00	1 539,00	0,57	0,00	8 419,00
13.	Mean area per leaf of the shoots of cordon 1	48,94	25,25	0,52	0,00	204,50
14.	Mean area per leaf of the shoots of cordon 2	48,00	30,70	0,61	0,00	452,10
15.	Total mean area per leaf of both shoots	55,41	20,98	0,38	0,00	200,60
16.	Total leaf dry mass per shoot of cordon 1	7,48	5,66	0,76	0,00	32,41
17.	Total leaf dry mass per shoot of cordon 2	6,25	5,33	0,85	0,00	26,70
18.	Total leaf dry mass of both shoots	13,71	7,84	0,57	0,00	39,75
19.	Mean dry mass per leaf of the shoots of cordon 1	7,05	116,60	16,55	0,00	2 000,00
20.	Mean dry mass per leaf of the shoots of cordon 2	0,23	0,14	0,60	0,00	0,61
21.	Total mean dry mass per leaf of the shoots of both cordons	7,29	0,09	0,32	0,00	0,53
22.	Evaluation of the vines	2 520,00	659,30	0,26	1 000,00	3 000,00
Phase II						
Phase I plus the following 5 variables						
23.	Total soluble solids	19,77	2,16	0,11	0,00	23,10
24.	Total titratable acids	7,90	1,16	0,15	0,00	11,80
25.	pH	3,45	0,30	0,09	0,00	3,69
26.	Yield per vine	4,99	2,25	0,45	0,40	11,10
27.	Number of bunches per vine	26,27	10,86	0,41	0,00	74,00

* Discrepancies in the data set are attributable to computer rounding off.

TABLE 8

Factor loadings for the first 4 Eigenvalues after rotation for 27 variables

(Programme used: Input, Utilit, Scale, Correl, Kaprin, Katran, Kavan, Varvar)

Variable Number	Variables	Factor 1	Factor 2	Factor 3	Factor 4
Phase I					
1.	Shoot length cordon 1	-0,1205	+0,1501	-0,0400	-0,0644
2.	Shoot length cordon 2	-0,1172	+0,0469	-0,0699	-0,0348
3.	Total shoot length of both cordons	-0,1516	+0,1236	-0,0642	-0,0606
4.	Spurs per cordon 1	-0,0323	+0,0456	-0,1120	-0,5037
5.	Spurs per cordon 2	-0,0227	+0,0500	-0,0171	-0,4559
6.	Spurs per vine	-0,0356	+0,0618	-0,0339	-0,6306
7.	Number of leaves per shoot of cordon 1	-0,4146	-0,0085	+0,0058	-0,0337
8.	Number of leaves per shoot of cordon 2	-0,0176	-0,4315	+0,0098	-0,0596
9.	Total number of leaves of the shoots of variables 7 and 8	-0,3583	+0,2921	+0,0202	-0,0728
10.	Total leaf area per shoot of cordon 1	-0,4119	+0,0279	-0,1210	-0,0224
11.	Total leaf area per shoot of cordon 2	-0,0687	+0,4189	-0,1554	-0,0189
12.	Total leaf area of both shoots	-0,3048	+0,2552	-0,1976	-0,0293
13.	Mean area per leaf of the shoots of cordon 1	-0,2646	-0,0291	-0,2209	-0,1828
14.	Mean area per leaf of the shoots of cordon 2	+0,0295	+0,2202	-0,1160	-0,0828
15.	Total mean area per leaf of both shoots	-0,1313	+0,0713	-0,1565	-0,0962
16.	Total leaf dry mass per shoot of cordon 1	-0,4183	-0,6172	-0,0500	+0,0042
17.	Total leaf dry mass per shoot of cordon 2	-0,0213	+0,4314	-0,0398	-0,0409
18.	Total leaf dry mass of both shoots	-0,3164	+0,2810	-0,0643	-0,0240
19.	Mean dry mass per leaf of the shoots of cordon 1	-0,0748	+0,0272	+0,0215	-0,0649
20.	Mean dry mass per leaf of the shoots of cordon 2	+0,0273	+0,3121	-0,2346	-0,1963
21.	Total mean dry mass per leaf of the shoots of both cordons	-0,1909	+0,1130	-0,3729	-0,1336
22.	Evaluation of the vines	+0,0129	+0,0062	-0,2258	+0,0049
Phase II					
Phase I plus the following 5 variables					
23.	Total soluble solids	-0,0124	+0,0446	-0,1170	+0,0279
24.	Total titratable acids	+0,0718	-0,3363	+0,1380	-0,0811
25.	pH	+0,0165	+0,0408	-0,0427	+0,0061
26.	Yield per vine	-0,0655	+0,0416	+0,0217	-0,0744
27.	Number of bunches per vine	-0,0540	+0,0358	-0,0009	-0,0777
Eigenvalues					
		5,4	4,9	2,4	2,3
Factor percentage responsible for variance					
		24,8	23,6	11,2	11,1
Cumulative percentage of variance					
		24,8	48,4	59,6	70,7

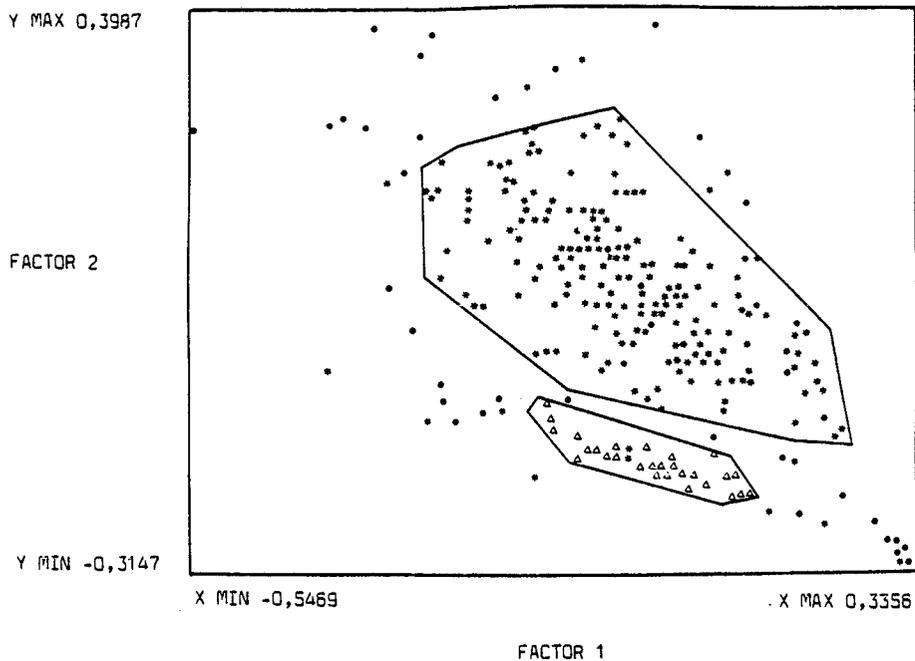


Fig. 5. PCA of 245 vines with 27 variables of a *Vitis vinifera* L. cv. Cape Riesling vineyard. Factor loadings for total leaf cover and total leaf cover of cordon 2 for PCA I and II (* vines considered homogeneous and • and Δ heterogeneous).

groups of vines which were present in cluster 1 (Fig. 1) of phase I is still evident although the cluster seems to be more compact with much smaller relative distances between groups (Fig. 6). This is because of the additional clustering effect of the added parameters measured.

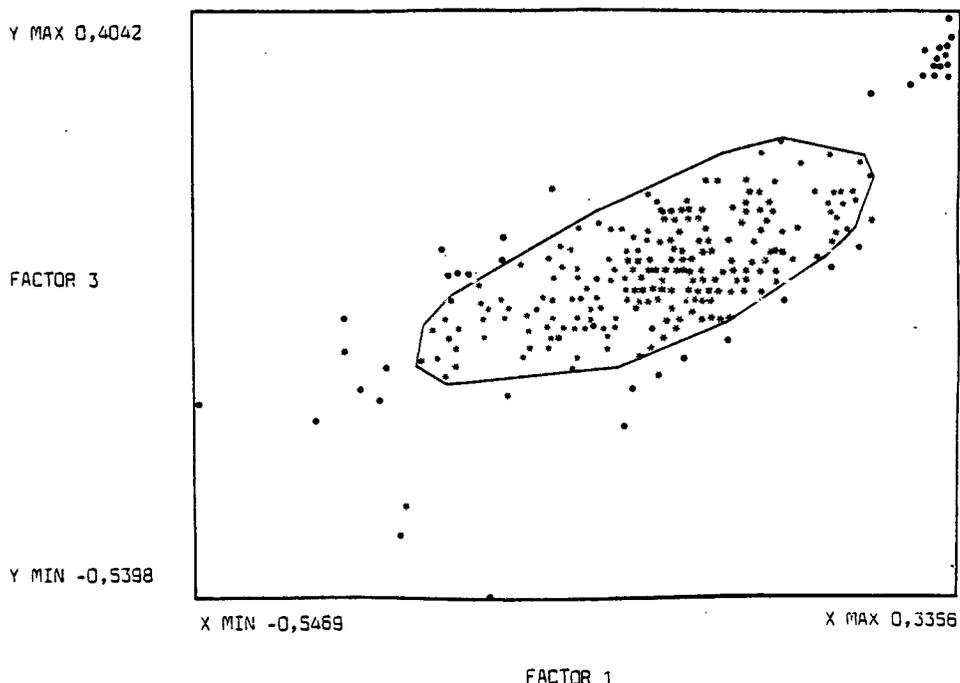


Fig. 6. PCA of 245 vines with 27 variables of a *Vitis vinifera* L. cv. Cape Riesling vineyard. Factor loadings for total leaf cover and average leaf cover for PCA I and III (* vines considered homogeneous and • heterogeneous).

Factor 3 has an eigenvalue of 2,35 explaining 11,2% of the total variance in the original data set. The highest factor loadings in this factor are the area per leaf of the shoots on cordon 1, cordon 2 and both cordons as well as the average leaf dry mass of the vine. This may once again be interpreted as being a growth factor.

In Fig. 6 factor 1 was plotted (X-axis) against factor 3 (Y-axis). Compared to cluster 1 (Fig. 1) the additional clustering effect of the five added parameters is evident. In Fig. 7 factor 2 (Y-axis) was plotted against factor 3 (X-axis).

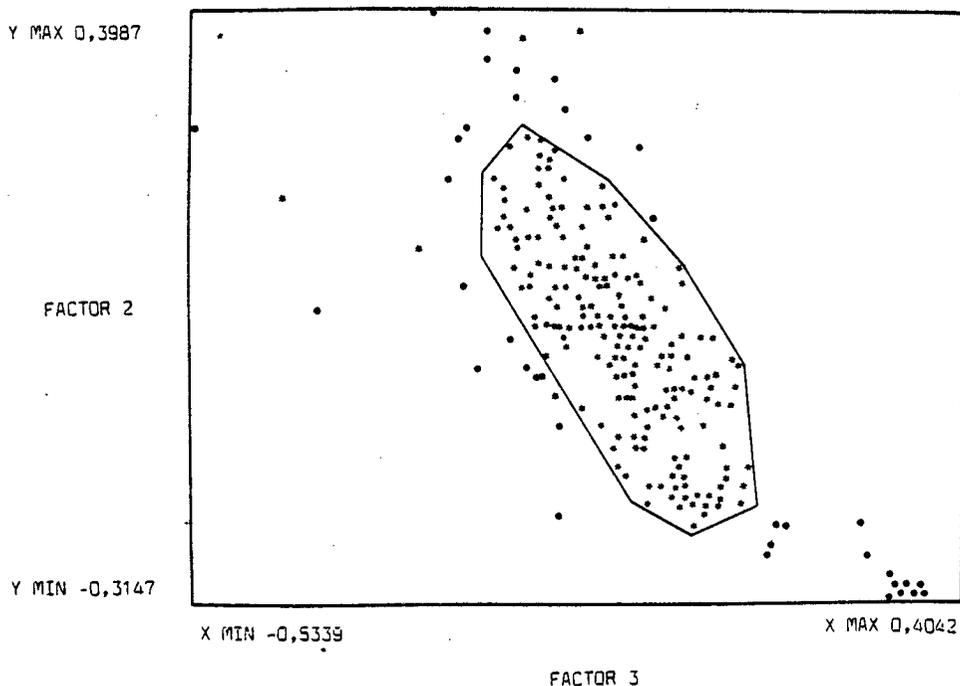


Fig. 7. PCA of 245 vines with 27 variables of a *Vitis vinifera* L. cv. Cape Riesling vineyard. Factor loadings for total leaf cover of cordon 2 and average leaf cover for PCA II and III (* vines considered homogeneous and • heterogeneous).

Factors 4 and 7 represent 11,1% and 9,4% (not shown) respectively of the variance in the original variables and have eigenvalues of 2,3 and 1,9. The highest factor loadings are on the growth parameters namely spurs and shoot length and may be interpreted as growth factors.

Factors 5 and 6 have eigenvalues of 2,2 and 2,0 (not shown) respectively with relatively high loadings on the parameters such as pH, yield per vine and number of bunches per vine.

CONCLUSION

In most PCA factors the leaf area played an important role in the clustering process although a number of factors affects the final selection. The more uniform vines were those with more or less the same leaf surface and growth variables, whereas those rejected for experimental purposes were those deviating from the above. In the selection of homogeneous vines it appears that instead of measuring 27 factors one could concentrate on variables for determining leaf canopy.

When all measured variables were taken into account, it is evident that the four soil types had little or no effect on the homogeneity of the different vines in the vineyard during this growth season. Quality variables such as $^{\circ}\text{B}$, TTA and pH resulted in better defined clusters and should therefore be used in future studies of this nature. The programme used in this study are powerful and handy tools in the hands of the viticulturist enabling him to simultaneously take into account groups of variables and their relationships with other groups and combined with the normal statistical tools such as randomisation and replication help to give a better understanding of the data.

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ABSTRACT

The effect of topping on a Vitis vinifera L. cv. Cape Riesling vineyard was investigated. Shoot and leaf growth of both the topped and untopped vines, respectively, can be described as sigmoidal. Shoot (cm) and leaf growth (cm^2) of the topped vines were significantly more than that of the untopped vines and are attributed to lateral shoot growth in the case of the topped vines. Topping had no effect on bunch development. The development of skin, pulp and seed with time of both the topped and untopped vines expressed as a percentage dry mass per berry can be described by a hyperbolic function for the skin, linear for the pulp and parabolic for the seed.

INTRODUCTION

Growth has been defined as "the advancement towards or attainment of full size of maturity; development; a gradual increase in size and the process whereby plants and animals increase in size by taking in food" (Bidwell, 1974; Salisbury & Ross, 1978). Growth may be evaluated by measurements of mass, length, height, surface area or volume (Noggle & Fritz, 1976). Growth curves of plants are generally sigmoidal (Bidwell, 1974; Noggle & Fritz, 1976; Salisbury & Ross, 1978) although double and triple sigmoids have been reported (Pratt & Reid, 1974; Coombe, 1976). Sigmoids and double sigmoids have been described for shoot growth and berry development for *Vitis* spp. and cultivars (Coombe, 1960; 1973; 1976; 1980; Nitsch *et al.*, 1960; Hale, 1968; Harris, Kriedemann & Possingham, 1968; Coombe & Hale, 1973; Kliewer & Schultz, 1973).

Removal of the distal 25 cm or more of the growing shoot tip is called topping (Winkler *et al.*, 1974) and is normally recommended to inhibit growth of vigorously growing shoots resulting in uniform and upright growth (Theron, 1944). By removing 25 cm of the apex, apical dominance is removed, resulting in the development of lateral shoots.

Results obtained in the northern hemisphere indicate that the juvenile leaves of the lateral shoots are the major sinks for nutrients (Hale & Weaver, 1962; Koblet, 1977) but after two to three leaves have matured, basipetal translocation of nutrients takes place (Koblet & Perret, 1971; 1972).

It is important that only vigorously growing vines should be topped because poor growth will be further aggravated by the effect of topping (Malan, 1935; Theron, 1944). The timing of topping is very important because the removal of leaves at the wrong time will result in insufficient grape nourishment. Le Roux & Malan (1945) and Coombe (1959) reported that repeated topping (three to four times or more during one season) decreased berry mass. Similarly El-Zeftawi & Weste (1970) found that a drastic decrease in leaf area usually causes a loss in berry mass and sugar concentration.

Since 1944 no work on the effect of topping on the vine was done in South Africa. It is therefore important that the effect of topping on the vine under South African climatic conditions should be investigated. The aim of this investigation was to determine the effect of topping on the growth characteristics of Vitis vinifera L. cv. Cape Riesling.

MATERIAL AND METHODS

Material: Fifteen year old V. vinifera cv. Cape Riesling vines grafted on 99 Richter rootstocks and trellised to a Perold trellising system (Zeeman, 1981) were used in this investigation. Homogeneous vines were selected as described by De la Harpe & Visser (1983) and were spur pruned to 16 buds per kilogram shoot mass. Rainfall was supplemented by two overhead sprinkler irrigations of 200 mm each during November 1981 and January 1982, respectively.

Methods: The selected vines were divided into two sections of 104 vines each.

One section was topped by removing the proximal 30 cm of each shoot of the vine at pea berry size development stage (56 days after bud break). For the purpose of this investigation bud break was defined as that stage at which 10% of the vines had two leaves. Topping was done at this developmental stage to ensure that the treatment was applied before the linear growth phase of the shoot. The other section was left untopped. Ten topped and 10 untopped vines were randomly selected and on each the two shoots on the second acropetal spur of each cordon were used for determination of shoot length and leaf area. Shoot lengths were determined frequently at irregular intervals. Leaf areas were determined with a model LI-3000 Li-Cor Portable Area Meter. Whole bunches on the second spur of both cordons of three topped as well as of three untopped vines were sampled 69, 76, 82, 92, 97, 110, 117, 131, 139, 145 and 152 days after bud break and stored at -20°C until subsequent analyses.

Sixty berries per vine from each of the topped and untopped vines were sampled 69, 76, 82, 92, 97, 110, 117, 131, 139, 145 and 152 days after bud break and the berry volume determined by measuring water displacement in a measuring cylinder. The fresh and dry mass of the berry, skin, pulp and seed separately were determined on 60 berries per bunch. Dry mass was determined by drying at 80°C to a constant mass.

One way analyses of variance were done by BMDP-IV and SPSS statistical programme and the regression analyses by a linear Least Squares Curve fitting programme (Wood & Gorman, 1971).

RESULTS AND DISCUSSION

The vegetative growth phase

Shoot growth: The average, final shoot length for the untopped vines was 267,8 cm (Fig. 1). The shoots started off with a slow elongation rate but shoot growth increased from 60 days after bud break i.e. shortly after topping (Fig. 2). This sharp increase lasted about three days after which the growth rate dropped to approximately three cm per day and declined steadily until no elongation could be measured at 135 days after bud break.

The average, final total shoot length of the topped vines was approximately 410 cm (Fig. 1) which is significantly more than that of the untopped vines mainly as a result of lateral shoot development. The shape of the growth curve (Fig. 1) of the topped vines was almost identical to that of untopped vines although at a much higher level from 60 days after bud break onwards. Two days after topping the elongation rate increased significantly and reached 37 cm per day for two days whereafter it declined sharply to about three cm per day (Fig. 2). Growth stopped 155 days after bud break in contrast to the 135 days of the untopped vines (Fig. 2).

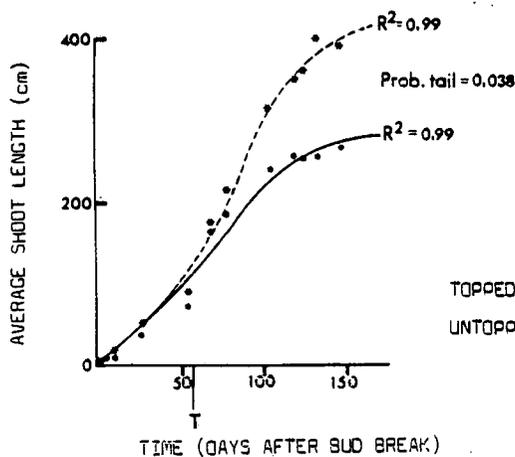


FIG.1. Fitted curves and observed data for topped (*) and untopped (●) *Vitis vinifera* L. cv. Cape Riesling vines. (T = time of topping).

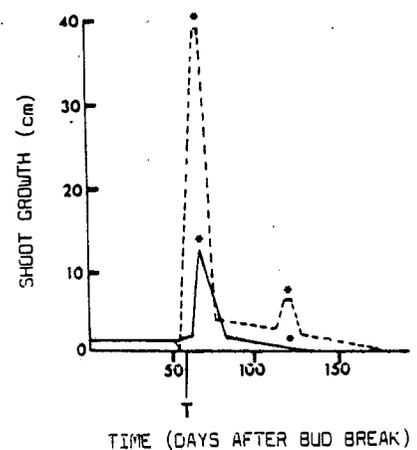


FIG. 2. Daily shoot elongation for topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines. (*significant differences ($\alpha = 0,05$) in the data set; T = time of topping)

The growth curve of the untopped vines reported here is very similar to those obtained by Van der Westhuizen (1974), Winkler et al., (1974) and Zelleke & Kliewer (1979).

Leaf growth: Before leaf fall began at 126 days after bud break, a total number of 129 leaves per shoot had differentiated on the untopped vines (Fig. 3). A maximum of 194 leaves per shoot for topped vines was obtained 134 days after bud break which amounted to a significant increase of 62% in the number of leaves per shoot per vine. This increase in number of leaves (65) from untopped to topped vines is statistically significant ($P = 0,05$). Increase in number of leaves per day during the growing season was expressed in at least three definite peaks namely at 30, 104 and 125 days after bud break (Fig. 4). The loss of leaves in the topped vines 56 days after bud break is attributable to topping.

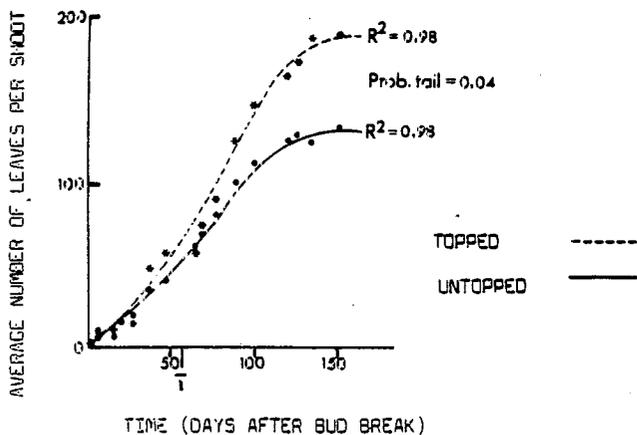


FIG. 3. Fitted curves and observed data for topped (*) and untopped (●) *Vitis vinifera* L. cv. Cape Riesling vines. (T = time of topping).

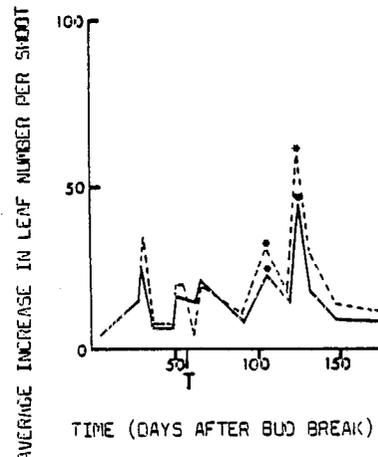


FIG. 4. Increase in number of leaves per day during the growing season for topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines (* significant differences ($p = 0,05$) in the data set; T = time of topping).

The total leaf area of 4728 cm^2 per shoot for the untopped vines was significantly less ($P = 0,05$) than the 7741 cm^2 for the topped vines (Fig. 5).

Three definite peaks in the rate of increase in leaf area were observed in the case of topped vines (Fig. 6). These peaks coincided with the bursts in shoot length. The weekly temperature from bud break to harvest is given in Figure 7.

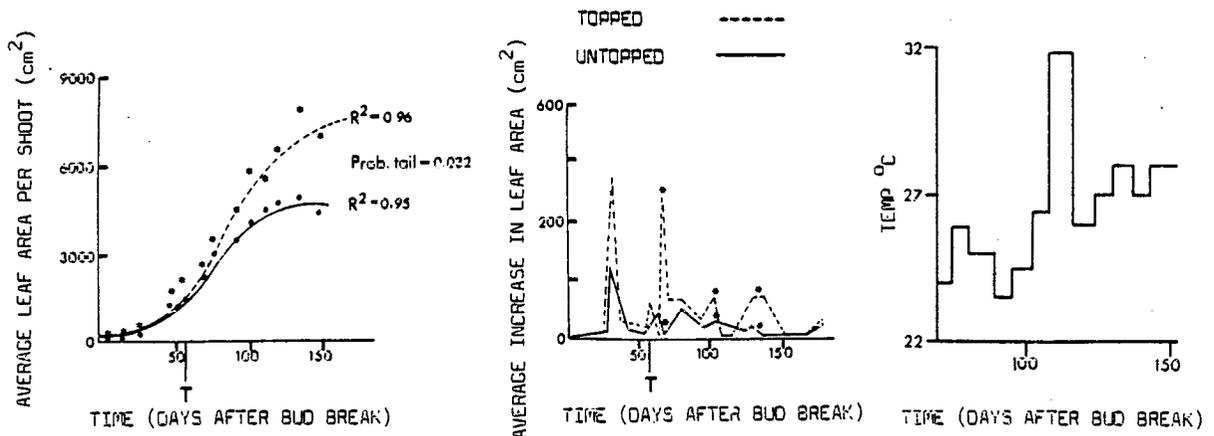


FIG. 5. Fitted curves and observed data for the average increase in leaf area from budbreak to harvest for topped (*) and untopped (•) *Vitis vinifera* L. cv. Cape Riesling vines. (T = time of topping).

FIG. 6. Increase in leaf area per day during the growing season for topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines (* significant differences ($p = 0,05$) in the data set; T = time of topping).

FIG. 7. Weekly temperature from bud break to harvest.

Reproductive growth phase

Bunches: The development of the bunches on topped and untopped vines is shown in Figs. 8, 9 and 10. No statistically significant differences ($P = 0,05$) were found between the fresh and dry mass per bunch of the topped and untopped vines (Figs. 8 & 9). It is clear that topping did not affect bunch development or that variation was so large that the effect of topping could not be shown statistically different.

Berries: The increase in berry volume showed a double sigmoid curve (Fig. 11) and could be divided into three stages as described by Coombe (1960; 1973; 1976; 1980), Harris, Kriedemann and Possingham (1968),

Coombe & Hale, (1973). The berry volume for both topped and untopped vines was $0,5 \text{ cm}^3$ 69 days after bud break, and attained a final value of 1,67 and $1,56 \text{ cm}^3$ respectively 152 days after bud break. According to Coombe (1976) the double sigmoid growth curve apart from its arbitrary nature giving rise to the three stage system, presents some logical and interpretational difficulties. One of the problems encountered is how and when a stage in the growth curve starts and ends. By fitting a linear regression curve $y = a + bt$ where $y =$ berry volume (cm^3); $a =$ constant on y axis; $b =$ coefficient constant and $t =$ time (days) on the data set, a fit of 87% was obtained by this regression equation and could be used for further reference.

The growth curves of the berry were obtained by plotting the accumulated dry mass against time for the topped and untopped vines (Fig. 12) and was similar to those reported by Nitsch et al., (1960) for "Concord" and "Concord Seedless", Hale (1968) for "Shiraz", Coombe (1973) for "Doradillo" and Kliever & Schultz (1973) for "White Riesling", "Cardinal" and "Carignan" grapes. As in the case of "Concord Seedless" grapes (Nitsch et al., 1960) the curve of the accumulated dry mass was more linear than those reported in the literature with the result that it became difficult to determine the different growth stages. A regression analysis showed a linear fit with R^2 values of 98% for both the topped and untopped vines. Topping had no statistically significant ($P = 0,05$) effect on the dry mass of the berries of the treated and control vines.

The accumulated dry mass of the skin, pulp and seed was linear if plotted against time (Fig. 13). No statistically significant differences were

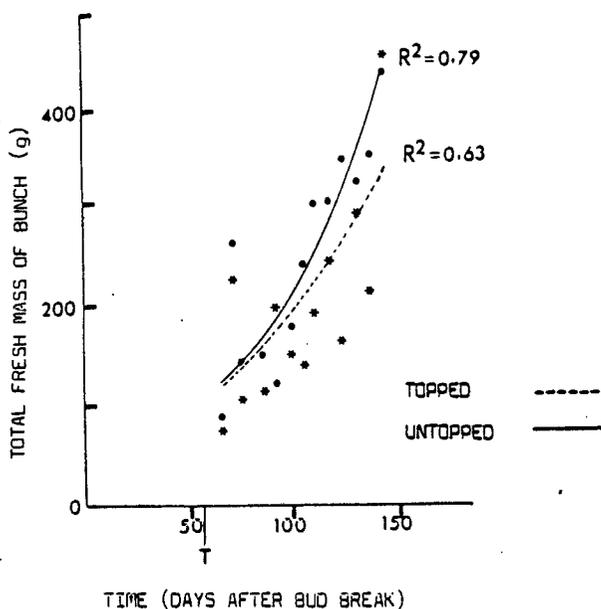


FIG. 8. Fitted curves and observed data for the increase in total fresh mass of the bunch from pea berry size to harvest for topped (+) and untopped (●) *Vitis vinifera* L. cv. Cape Riesling vines. (T = time of topping).

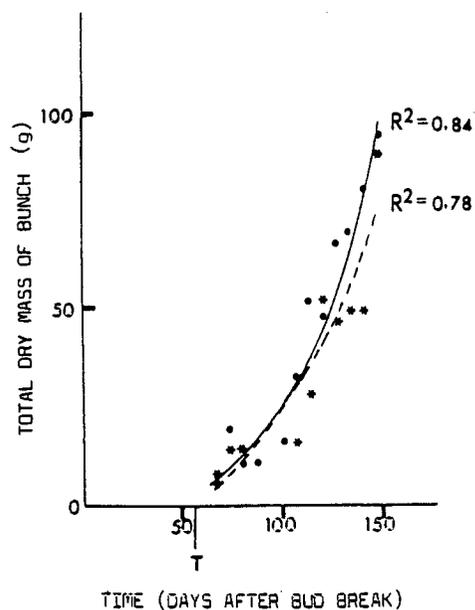


FIG. 9. Fitted curves and observed data for the increase in total dry mass of the bunch from pea berry size to harvest for topped (+) and untopped (●) *Vitis vinifera* L. cv. Cape Riesling vines. (T = time of topping).

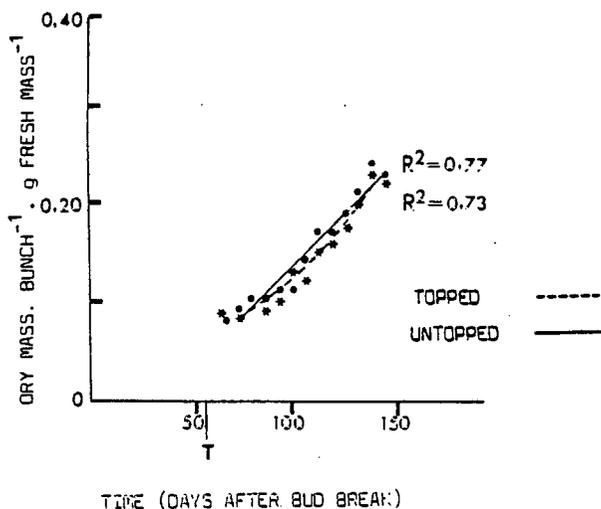


FIG. 10. Fitted curves and observed data for the increase in bunch mass for topped (+) and untopped (●) vines. (T = time of topping).

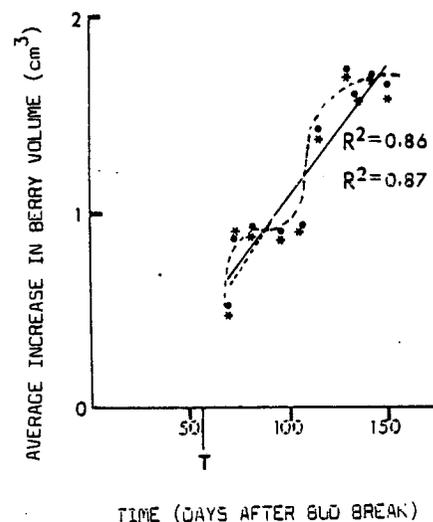


FIG. 11. A double sigmoid curve of volume versus time expressed on a cumulate bases for topped (+) and untopped (●) *Vitis vinifera* L. cv. Cape Riesling vines. (T = time of topping).

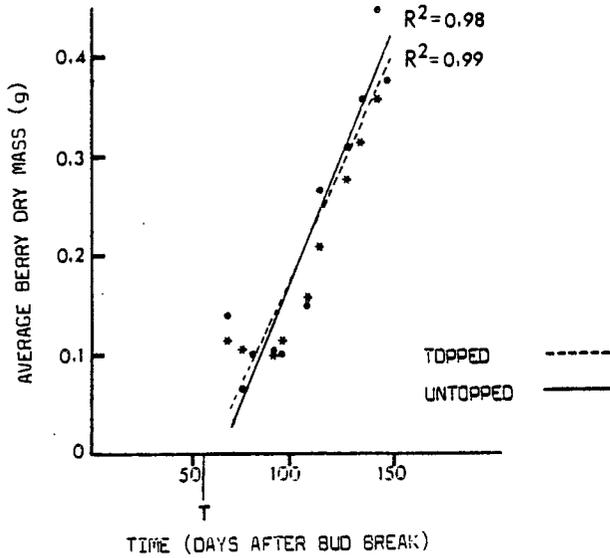


FIG. 12. Fitted curves and observed data for the average dry mass per berry from pea berry size to harvest for topped (*) and untopped (●) vines (T = time of topping).

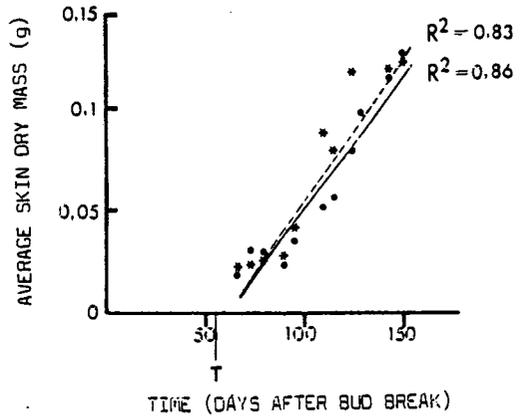


FIG. 13. Fitted curves and observed data for the average dry mass per skin from pea berry size to harvest for topped (*) and untopped (●) vines (T = time of topping).

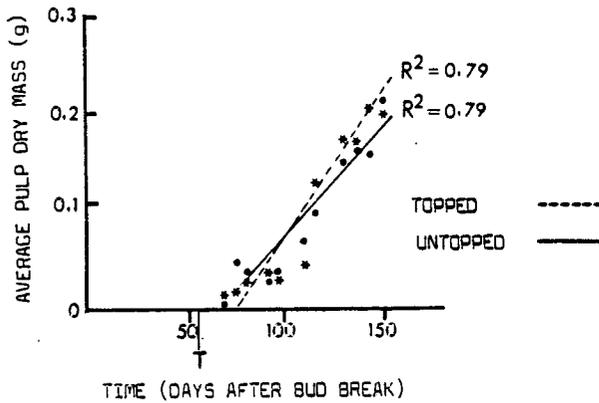


FIG. 14. Fitted curves and observed data for the average dry mass per pulp from pea berry size to harvest for topped (*) and untopped (●) vines (T = time of topping).

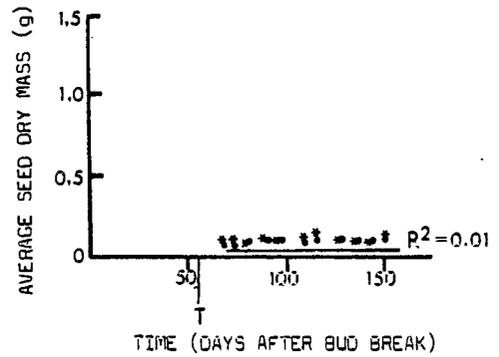


FIG. 15. Fitted curves and observed data for the average dry mass per seed from pea berry size to harvest for topped (*) and untopped (●) vines (T = time of topping).

found between these three components of the topped and untopped vines. During the early stages of the growth cycle the skin contributed more than the pulp and seed to total berry mass for both topped and untopped vines (Fig. 14). The percentage dry mass of the skin then declined rapidly whilst the percentage dry mass of the pulp and seed per berry accumulated. The percentage contribution of the seed contrary to the pulp then declined 92 days after bud break (Figs. 16, 17 & 18).

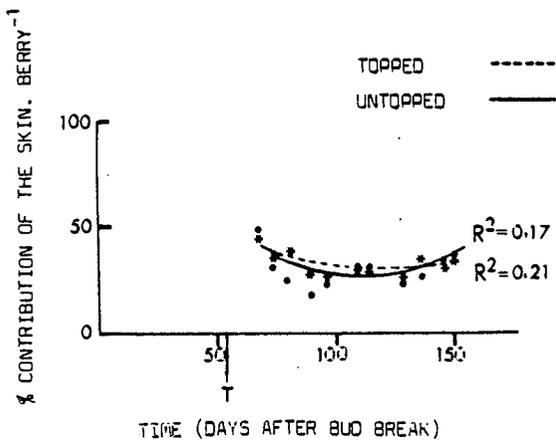


FIG. 16. Fitted curves and observed data for the percentage contribution of the skin to the dry mass of the berry for the topped (*) and untopped (●) vines (T = time of topping).

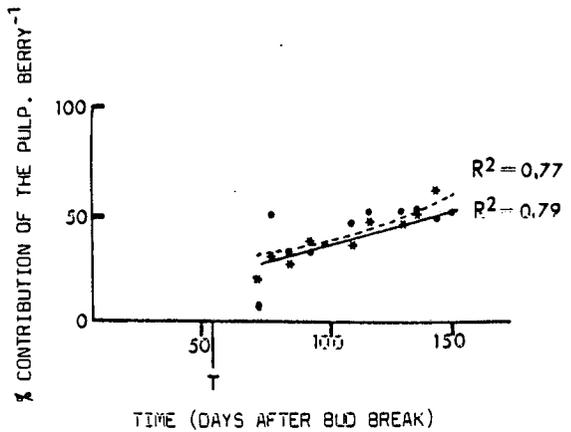


FIG. 17. Fitted curves and observed data for the percentage contribution of the pulp to the dry mass of the berry for the topped (*) and untopped (●) vines (T = time of topping).

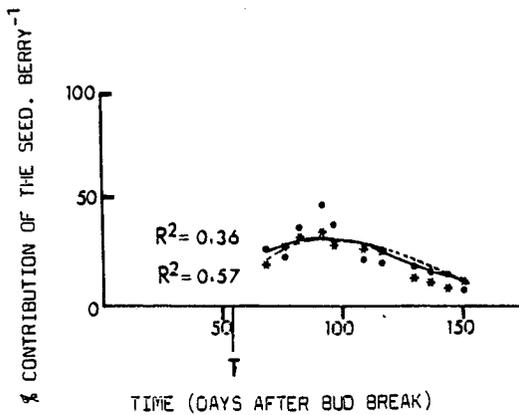


FIG. 18. Fitted curves and observed data for the percentage contribution of the seed to the dry mass of the berry for the topped (*) and untopped (●) vines (T = time of topping).

SUMMARY AND CONCLUSION

The shoot and leaf growth of *V. vinifera* cv. Cape Riesling can be described as sigmoidal. Significant differences were found between topped and untopped vines as far as rate of shoot and leaf growth are concerned. In the case of topped vines shoot development can be attributed to lateral shoot growth, enlarging the leaf area.

Topping had no effect on bunch development. The berry development is linear as far as volume and dry mass are concerned. The skin, pulp and seed development is hyperbolic in function whilst the development of the pulp is linear and that of the seed parabolic. No statistical differences were found between the bunches of topped and untopped vines.

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CHAPTER 4

Translocation of ^{14}C -labelled Photosynthetate in Vitis vinifera L. cv. Cape
Riesling

ABSTRACT

The contribution of lateral shoots, induced by topping, to grape composition was investigated. Multidirectional translocation of organic compounds took place. Bunches, shoots and leaves were sinks at veraison whilst leaves were the only sink at harvest. Bunches of topped vines had higher radio-active levels than bunches of the untopped vines at veraison and harvest after application of $^{14}\text{CO}_2$ to the leaves of the vines.

At veraison the shoot had the highest radio-activity in the neutral, anion and cation fractions indicating that organic compounds are being translocated from the leaves where the $^{14}\text{CO}_2$ was applied, to the rest of the vine. At harvest, however, the main activity remained in the leaves with no activity in the shoots and bunches indicating that very little export took place.

INTRODUCTION

Carbohydrates are translocated in the vine in the form of sucrose in the phloem (Stoèv, Mamaróv & Benchev, 1959; Swanson & El-Shishiny, 1959; Koblet, 1969; Weaver, Shindy & Kliewer, 1969; Winkler, et al., 1974; Saito & Kasai, 1978). Substances like higher sugars (Kliewer, 1965) organic acids, amino acids, hormones and inorganic nutrients are also translocated freely in the vine (Hardy, 1969; Winkler et al., 1974).

Climatic factors and viticultural practices play an important role in the translocation and distribution of these substances. One important factor in the translocation pathway of nutrients is the leaf canopy. Changes in the leaf area or exposition to the sun affects the rate of photosynthesis which could in turn affect the translocation of photosynthetate. Koblet (1969) reported that leaves start to export photosynthetate when they reach about 30% of their final size. However, import into the leaf also takes place simultaneously. The rate of export only exceeds the rate of import after the leaves had reached 50 to 75% of their final size. Koblet (1977) also reported on the movement of carbohydrates in young shoots, from bloom to fruit set, during berry maturation and the effect of shoot position on translocation. In young shoots the lowest leaves exported their assimilates mainly basipetally, the middle leaves showed a bidirectional export eg. to the shoot tip and inflorescences and the upper leaves to the shoot tip and younger leaves (Koblet, 1977). At bloom and fruit set the same pattern as young shoots were obtained. As growth proceeds, however, the predominant movement of photosynthetate became basipetal. During berry development and ripening Koblet (1977) reported a

basal movement of carbohydrates mainly into the bunches. Hale & Weaver (1962) were unable to detect movement of photosynthetate from the main shoot to the lateral shoots. Koblet (1969; 1975) and Koblet & Perret (1971; 1972) found that lateral shoots without grapes also export their carbohydrates to the grapes on the other shoots.

Since topping resulted in a doubling of leaf area as well as the creation of new sinks such as growing lateral shoot tips (Chapter 3), this investigation was done to determine the effect of topping on translocation and distribution of photosynthetate in the developing berry.

MATERIAL AND METHODS

Material: *Vitis vinifera* L. cv. Cape Riesling vines were selected as described by De la Harpe & Visser (1983).

Methods: Nine each of apical, middle and basal lateral shoots of nine topped vines as well as nine main shoots of nine untopped vines were exposed to $^{14}\text{CO}_2$ in polyethylene bags (Fig. 1) at veraison and harvest. The $^{14}\text{CO}_2$ was generated by addition of one cm³ 20% lactic acid to 0,5 cm³ 2,2 MBq NaH¹⁴CO₂, (carrier concentration = 0,01 mole NaHCO₃.dm⁻³). Fixation of $^{14}\text{CO}_2$ was allowed for 30 min after which the excess of $^{14}\text{CO}_2$ was removed by removing the polyethylene bags which covered the shoot. The shoots were then samples after three, six and nine h after the initial 30 min exposure to $^{14}\text{CO}_2$. At each sampling time three each of apical, middle and basal lateral shoots of topped vines as well as three main shoots, of untopped vines, containing the stem, leaves and bunch were sampled. The material was then frozen at -20°C before freeze-drying.

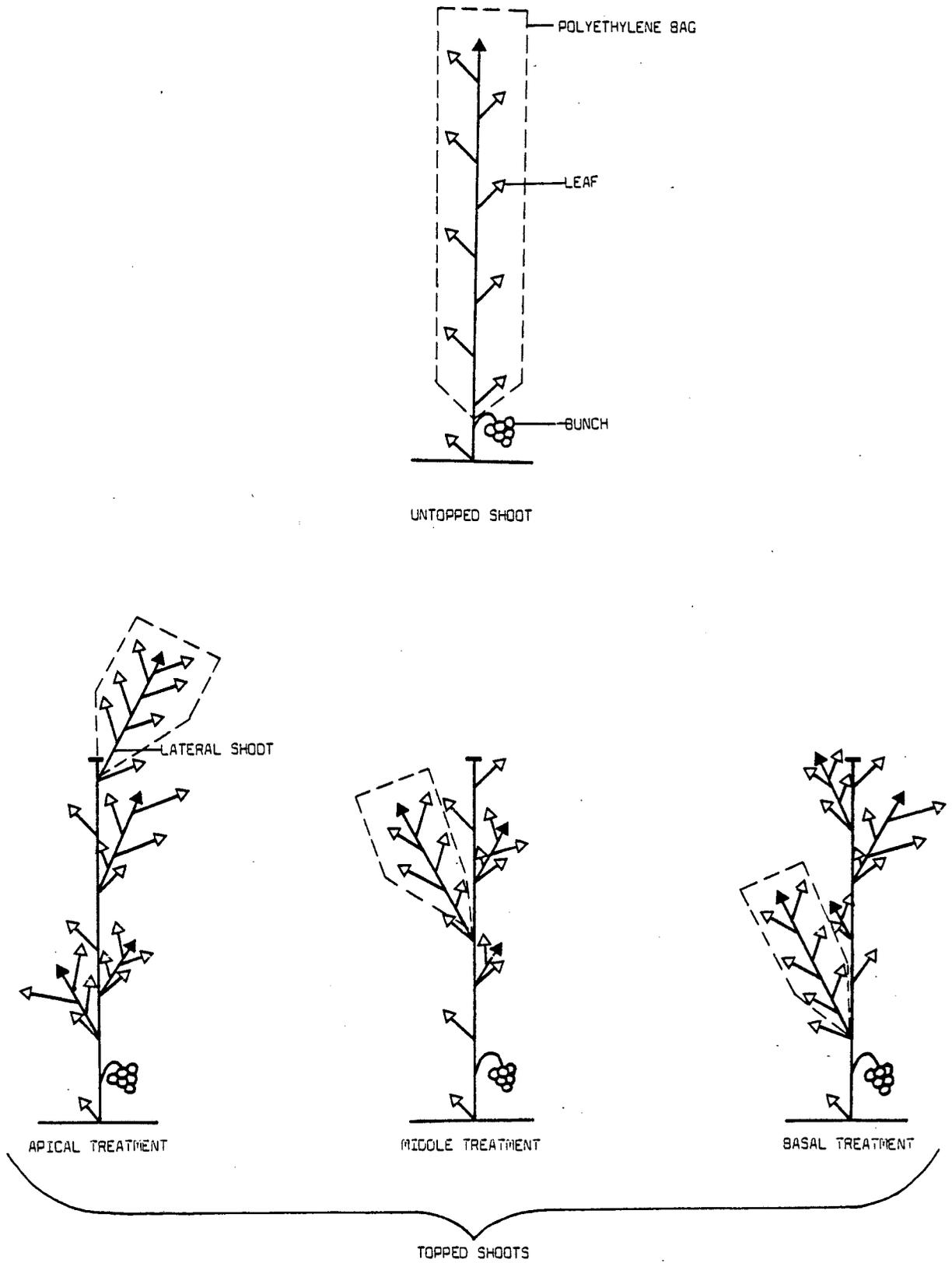


FIG. 1. Position of $^{14}\text{CO}_2$ application to the leaves of a *Vitis vinifera* L. cv. Cape Riesling vine.

For the determination of the total activity in the leaves, shoots and bunches, 0,5 g freeze-dried material was treated with 10% H_2O_2 for 48 h at $50^{\circ}C$ to oxidize pigments that may interfere with the counting of the radio-activity. Ten cm^3 Instagel scintillation liquid (Packard Ltd.) were added and vigorously shaken till the liquid turned to gel. Corrections for turbidity were made by means of a standard correction curve for turbidity. The turbid gel was then counted in a Packard Tricarb 460 scintillation spectrophotometer.

The question as to the nature of the translocated compounds arises when studying the distribution pattern of photosynthetate. In an endeavour to throw more light on this question, the distribution of the radio-activity among the main fractions of translocatable photosynthetate was investigated.

Neutral organic compounds, anionic and cationic compounds were extracted with $10\ cm^3$ 70% ETOH from one mg freeze-dried material. The anions were recovered from the $10\ cm^3$ extract by adsorption on Amberlite AG 400 (200 -400 mesh) anion-exchange resin in the Cl^- form. One cm^3 resin (determined by means of a standard curve) was then washed with $100\ cm^3$ deionised water and eluted with one cm^3 10% H_2SO_4 solution followed by distilled water to make up $25\ cm^3$. The cations were recovered from the extract (leaving only the neutral components in the extract) by adsorption on Amberlite CG-50 (200 - 400 mesh) cation resin in the H^+ form. One cm^3 resin (determined by means of a standard curve) was then washed with $100\ cm^3$ deionised water and eluted in one cm^3 10% HCl followed by deionised water to make up $25\ cm^3$. The remaining neutral fraction was also made up to $25\ cm^3$.

The data was statistically processed by an one way analysis of variance and

covariance BMDP statistical programme. Comparison of means was carried out using the Scott Knott multiple range test (Gates & Bilbro, 1978). Data was transformed using ARCSIN (Snedecor & Cochran, 1967).

RESULTS AND DISCUSSION

Movement of metabolites at veraison and harvest

Untopped vines: The percentage $^{14}\text{CO}_2$ activity (%A) representing metabolised $^{14}\text{CO}_2$ in the bunch, shoot and leaves after nine h are depicted in Fig. 2. At veraison 73% A was found in the leaves, 23% in the shoot and four% in the reproductive organs. At harvest, however, less than one % of the activity could be detected in the bunches while 10% of the activity was obtained in the shoot and 89% in the leaves (Fig. 2).

If the distribution of the activity after 30 min of exposure to $^{14}\text{CO}_2$, is determined after three, six and nine h, it is evident that translocation of $^{14}\text{CO}_2$ took place since activity is detected in the bunch (not exposed) (Fig. 3.). At veraison between 74 and 85% of the activity is found in the leaves, 12 to 20% in the shoot and three to six% activity in the bunch. Contrary to the situation at veraison, 78 to 96% of the activity is found in the leaves, three to 20% in the shoot and only 0,2 to 0,3% in the bunches at harvest. The results are statistically significant (Table 1).

The fluctuation in the %A detected between three, six and nine h might be attributed to a host-parasite relationship between exporting leaves and importing juvenile leaves depending on the ratio of exporting to juvenile leaves.

This would be in accordance with the results obtained by Koblet (1977) who showed that leaves only start exporting photosynthetate at about 30% of their final size and that they could be considered parasitic before export starts.

It is clear from the results depicted in Figs. 2 and 3 that at harvest only a small %A was detected in the shoot, while at least double the activity detected at harvest had been found at veraison. It is further important to note that in both cases (veraison and harvest) vegetativē growth (elongation) had stopped (Chapter 3) and the apical meristem would not, therefore, be expected to be a strong sink for photosynthetate. Hale & Weaver (1962) reported that the photosynthetate is translocated mostly in a basal direction with little or no acropetal movement. From the results obtained in this investigation it is evident that the photosynthetate in untopped vines were translocated from the leaves, where fixation took place, to the bunches, i.e. basipetally (Fig. 3). The experimental arrangement did not allow for measurement of activity in an acropetal direction.

Topped vines: The %A for the bunch, shoot and leaves at veraison and at harvest are depicted in Fig. 4 for the different treatments (i.e. apical, middle and basal). The statistical analyses of the data set is shown in Table 1.

Contrary to the untopped vines where most of the activity at veraison was situated in leaves (74 to 85%) with only 12 to 20% in the shoots and three to six% in the bunches, the activity was more evenly distributed in the topped vines (Fig. 4) especially at veraison. The %A in the shoots was higher in the topped than in the untopped vines, whilst the leaves of topped vines had

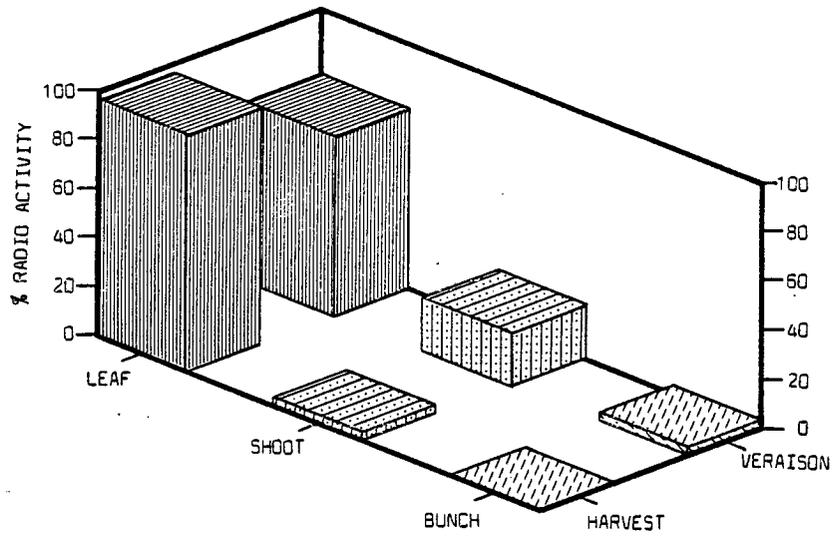


FIG. 2. The percentage $^{14}\text{CO}_2$ activity representing metabolized $^{14}\text{CO}_2$ in the bunch, shoot and leaves of untopped vines after nine hours after application of $^{14}\text{CO}_2$.

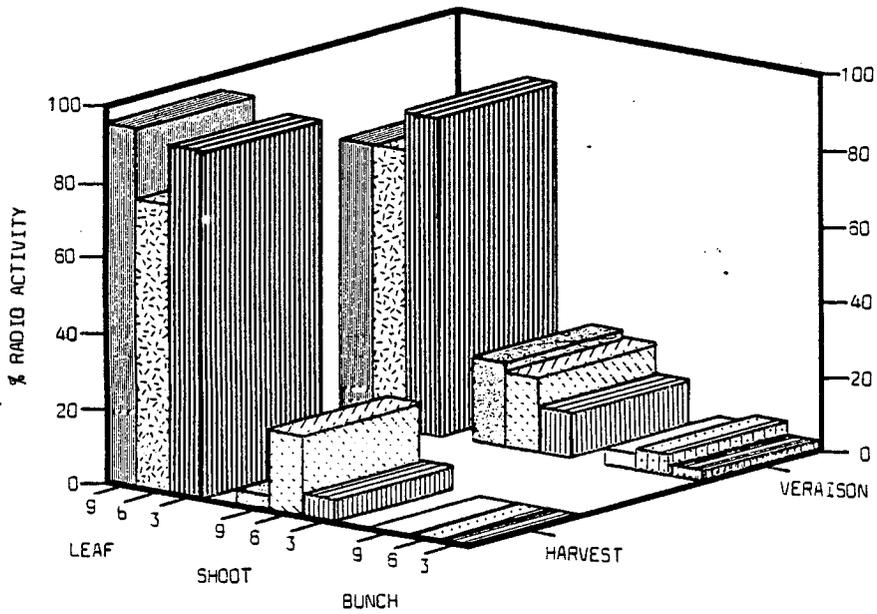


FIG. 3. The distribution of radio activity after three, six and nine hours after $^{14}\text{CO}_2$ application to untopped vines.

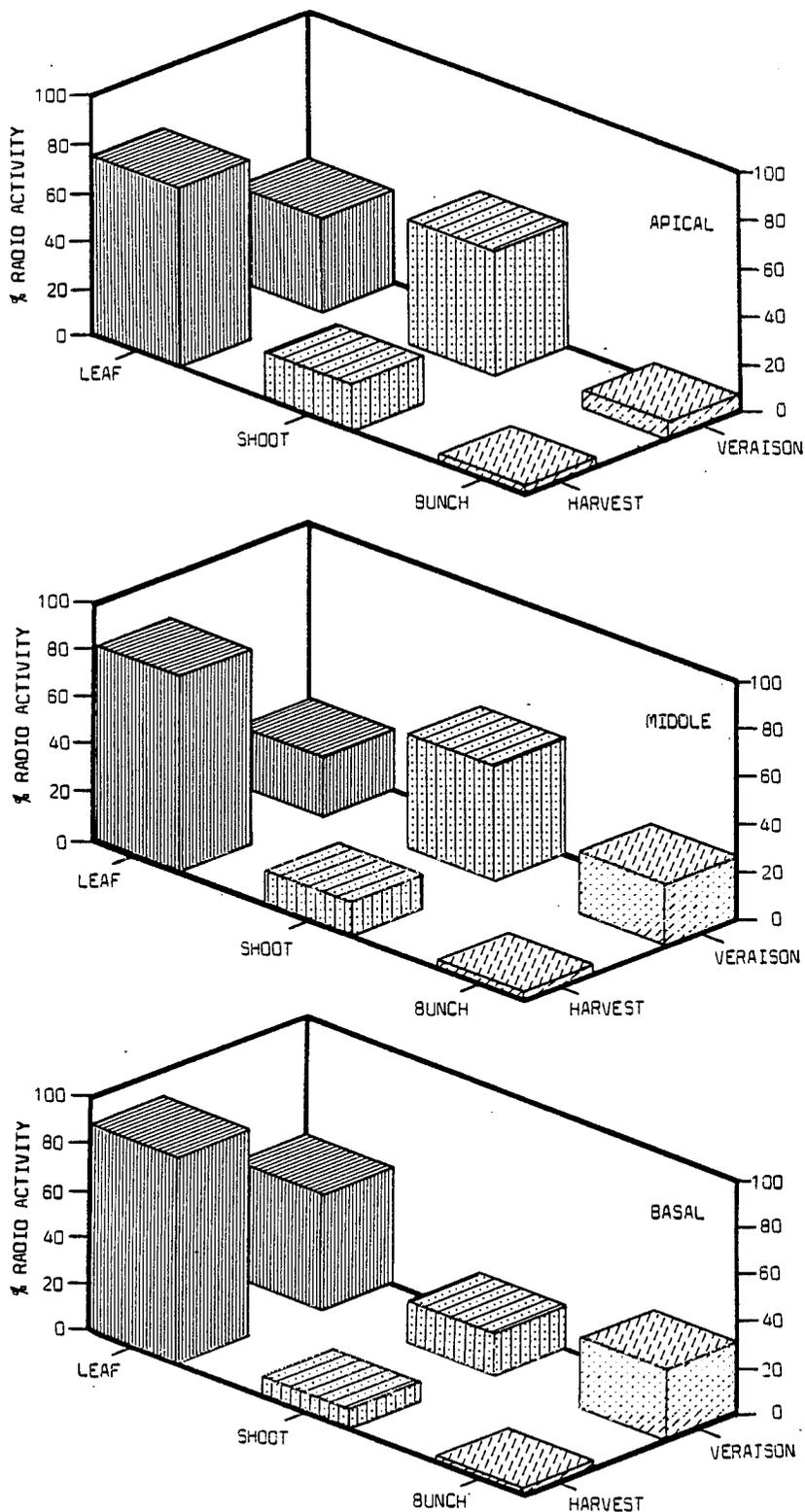


FIG. 4. The percentage $^{14}\text{CO}_2$ activity representing metabolised $^{14}\text{CO}_2$ in the bunch, shoot and leaves of topped vines after nine hours after application of $^{14}\text{CO}_2$. A) Apical treatment B) Middle treatment C) Basal Treatment.

less activity than those of untopped vines. These differences were statistically significant ($P = 0,05$) (Table 1).

The bunches of the untopped vines had statistically higher ($P = 0,05$) activity than those of the topped vines indicating an influx of activity into the bunch at veraison (Table 1). No statistical differences could be found between the results of the three, six and nine h treatments of the leaves, shoots and bunches.

In contrast to the export of photosynthetate at veraison, the topped vines showed very low export of activity at harvest (Fig. 4). The highest activity in the topped vines was in the leaves, whilst the shoot and bunches contributed only 20% of the total activity present. Since all vegetative growth had virtually stopped at harvest (Chapter 3), the bunch was left as the most important sink on the shoot and one would expect the bunch to have high %A as had been described by Hale & Weaver (1962). These authors found that at harvest the photosynthetate were translocated from the apical, middle and basal position of the shoot to the bunch. Although some activity was detected in the bunch in this investigation, the ratio of bunch: shoot: leaves was 3:15:82 showing that the bulk of the activity was not exported from the leaves to the bunches at harvest. The fact that only a small %A could be found in the shoot over a period of time indicated that at harvest a very slow rate of translocation of photosynthetate took place and that even the organs concerned with photosynthetate storage did not constitute a very strong sink at that stage.

Application of $^{14}\text{CO}_2$ to the apical and middle parts of the shoots at veraison resulted in high %A in the shoots ranging from 40 to 60% (Fig. 5). In

Table 1. Statistically significant differences between the untopped and topped vines (apical, middle and basal shoot ¹⁴CO₂ treatments) in the bunch, shoot and leaf of *Vitis vinifera* L. cv. Cape Disling

	BUNCH				SHOOT				LEAF							
	VERAISON		HARVEST		VERAISON		HARVEST		VERAISON		HARVEST					
	Untopped	Topped	Untopped	Topped	Untopped	Topped	Untopped	Topped	Untopped	Topped	Untopped	Topped				
¹⁴ CO ₂ Application time	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M	
¹⁴ CO ₂ treatment	Control M M M	Apical M M M	Middle M M M	Basal M M M	Control M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M
Veraison	Control M M M	Apical M M M	Middle M M M	Basal M M M	Control M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M
Harvest	Control M M M	Apical M M M	Middle M M M	Basal M M M	Control M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M	Untopped M M M	Apical M M M	Middle M M M	Basal M M M

M = p = 0,1
 MM = p = 0,05
 MMM = p = 0,01
 (continued)

contrast, the treatment of basal leaves resulted in %A ranging from 18 to 29% in the shoot and 30 to 76% in the bunches (Fig. 5). After application of $^{14}\text{CO}_2$ to the basal parts at veraison, activity was found in the shoots and leaves higher up on the shoot, indicating a direct flow to these parts. These results are in contrast to those reported by Hale & Weaver (1962) and Koblet (1977) who found only a basipetal movement of photosynthetate. In this investigation, activity was detected in all parts from the bunch to the apical meristem indicating multidirectional movement rather than basipetal movement alone.

At harvest the main activity was found in the leaves (Fig. 5). The %A in the shoots were the highest in the apical treatments and decreased with the middle and basal treatments. It is, however, important that the activity remained relatively constant in the bunches independent of the site of application, indicating a directed flow of photosynthetate to the bunches. These results are in accordance with those reported by Hale & Weaver (1962) and Koblet (1977).

As mentioned previously the activity in the untopped vines at veraison were mainly in the leaves even nine h after exposure (Fig. 2). Contrary to the untopped vines, different %A were obtained in the topped vines, depending on the treatment i.e. the apical, middle or basal application of $^{14}\text{CO}_2$ to the shoot (Fig. 4).

An important fact which emerged from this investigation, was the fact that irrespective of where $^{14}\text{CO}_2$ was applied, it could be detected along the length of the shoot within a three h period. Furthermore, all the lateral shoots contained activity, independent of exposure position on the shoots, both at verai-

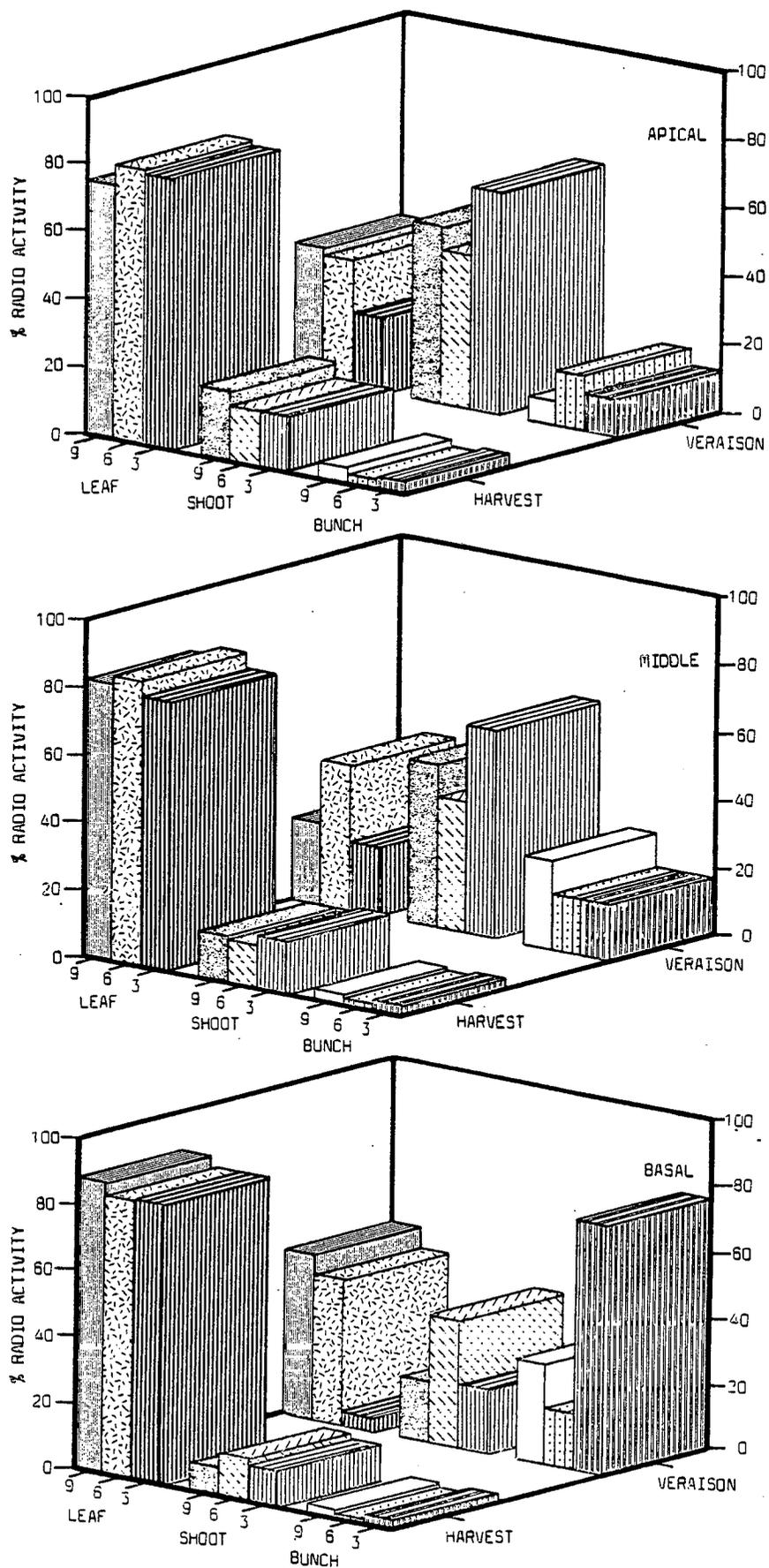


FIG. 9. The distribution of radio activity three, six and nine hours after $^{14}\text{CO}_2$ application to topped vines.

son and at harvest. This indicated that the lateral shoots acted as sinks throughout the ripening of the berries, contrary to the findings of Hale & Weaver (1962) who reported that no assimilate moved into the lateral shoots from the main shoot.

$^{14}\text{CO}_2$ applied to topped and untopped vines prior to harvesting remained in the leaves. The %A in the shoots decreased from the apical to the basal treatment, while the %A in the bunches stayed relatively constant for the different treatments.

A remarkable difference as far as the distribution and translocation of photosynthetate at veraison and at harvest exist. It is clear that the topped vines were metabolically more active than the untopped vines at veraison as judged by the fact that a higher %A was obtained in the shoots and bunches of the topped vines.

Distribution and movement of radio-activity among the neutral, anion and cation fractions at veraison and harvest

Untopped vines: The radio-activity in the neutral, anionic and cationic fractions of the alcohol extract of the various organs expressed as a percentage of the total activity in an organ are depicted in Fig. 6. The statistical analyses are given in Table 2.

At veraison the neutral fraction, representing mainly the sugars, had the highest %A in the leaves followed by the shoots and bunches (Fig. 6). The differences between the leaves on the one hand and the shoots and bunches on

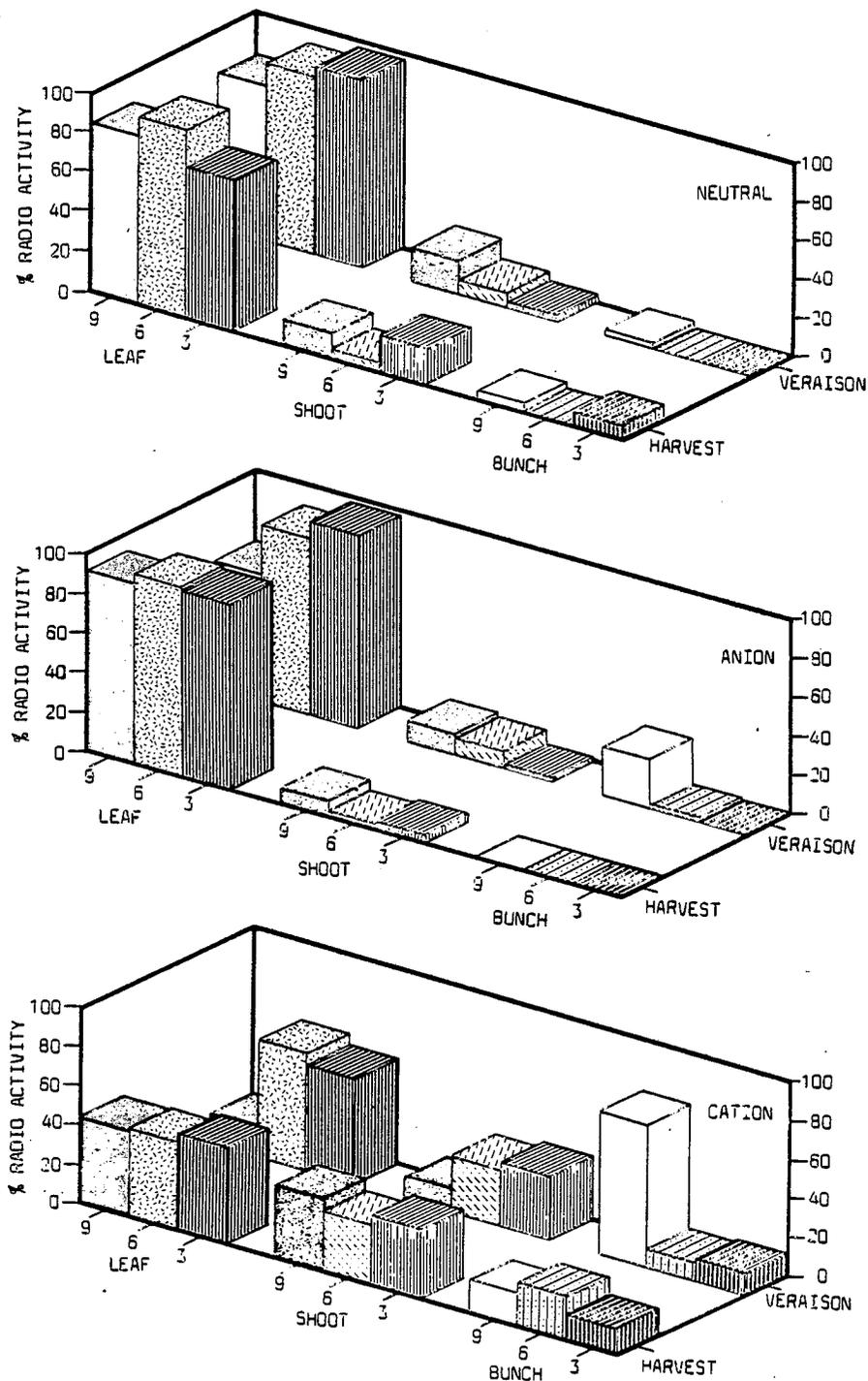


FIG.6. The distribution of radio activity three, six and nine hours in the neutral, anion and cation fractions of the alcohol extract of the leaf, shoot and bunch expressed as a percentage of the total activity in an organ of the untopped vines.

Table 2. Statistically significant differences based on the Scott Knott multiple range test, between the neutral, anion and cation fractions of the bunch, shoot and leaf at veraison and at harvest ($p = 0,05$)

Treatment	Neutral fraction			Anion fraction			Cation fraction		
	Bunch	Shoot	Leaf	Bunch	Shoot	Leaf	Bunch	Shoot	Leaf
Untopped	x	x	z	x	x	z	x	x	x
A	x	x	x	x	x	x	x	x	x
Topped M	x	x	x	x	x	x	x	x	x
B	z	x	x	z	x	x	x	x	x
Time		*	*		*	*		*	*

A = apical
M = middle
B = basal

| Treatment

Time = veraison and harvest

* = significant differences between veraison and harvest

the other hand were statistically significant (Table 2). As in the case of the neutral fraction, the main anion activity was situated in the leaves (Fig.6). After nine h, however, a decrease of anions were obtained in the leaves and at the same time an increase in the anions in the bunch were found indicating transport of anions. Contrary to the neutral and anion fractions a high %A for the cations were obtained in the bunches (Fig. 6). As in the case of the anionic fraction a high %A was obtained nine h after $^{14}\text{CO}_2$ application compared to the three and six h after application, indicating an accumulation of cations in the bunches.

At harvest, as in the case of veraison, the highest activity was situated in the neutral fraction of the leaf (Fig. 6). A slightly higher %A was obtained in the shoot in comparison to activity of the bunch at harvest. The anionic fraction was mainly situated in the leaves with little activity in the shoot and no activity in the bunches (Fig. 6). The cations were more or less evenly distributed between the leaves, shoots and bunches (Fig. 6) and no statistically significant differences could be found between these three organs.

Topped vines: At veraison, the bunches had the highest %A in the neutral fraction in the case of the basal treatment (Fig. 9). The neutral fractions of the shoots exposed to $^{14}\text{CO}_2$ in their apical (Fig. 7), middle (Fig. 8) and basal (Fig. 9) parts were high in comparison to the %A of the bunches and leaves. As far as the neutral fractions are concerned the leaves of topped vines had more than 50% less than the untopped vines (Fig. 6). This, together with the high %A of neutrals in the shoot indicated a high translocation rate of neutral components from the leaves to the shoots. A similar tendency was obtained for anions (Figs. 7, 8 & 9). The distribution ten-

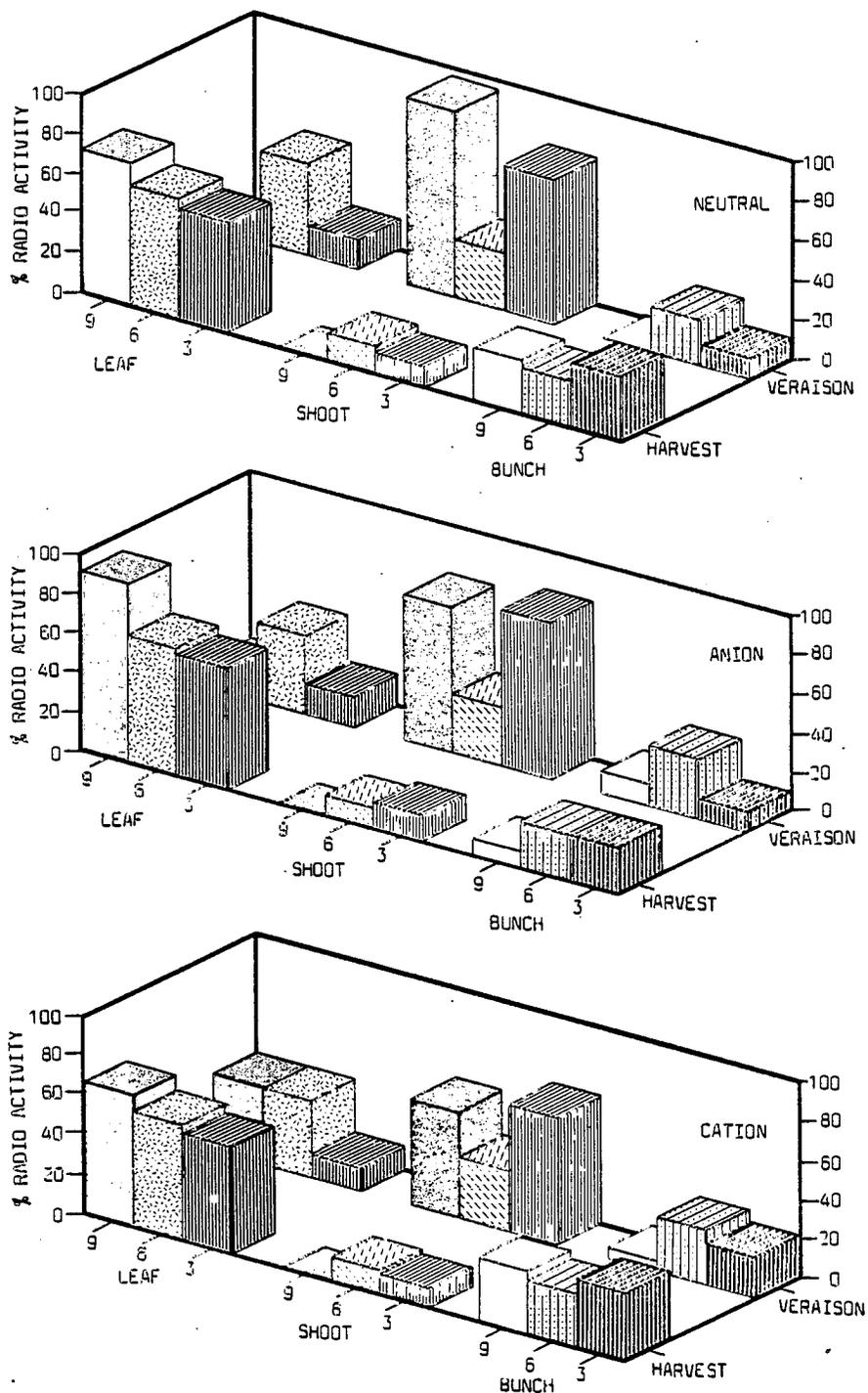


FIG.7. The distribution of radio activity three, six and nine hours in the neutral, anion and cation fractions of the alcohol extract of the leaf, shoot and bunch expressed as a percentage of the apical treatment of the topped vines.

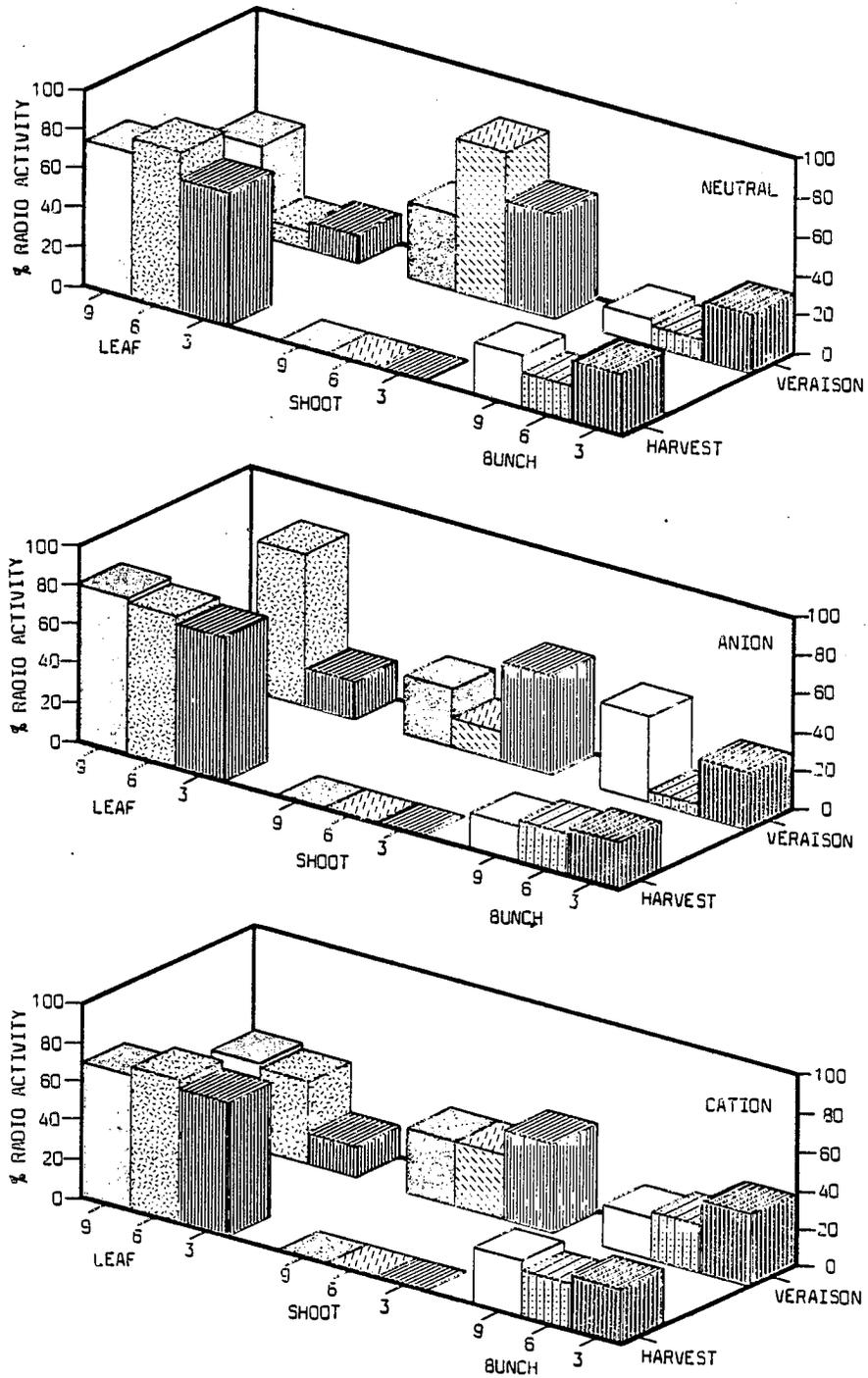


FIG. 8. The distribution of radio activity three, six and nine hours in the neutral, anion and cation fraction of the alcohol extract of the leaf, shoot and bunch expressed as a percentage of the middle treatment of the topped vines.

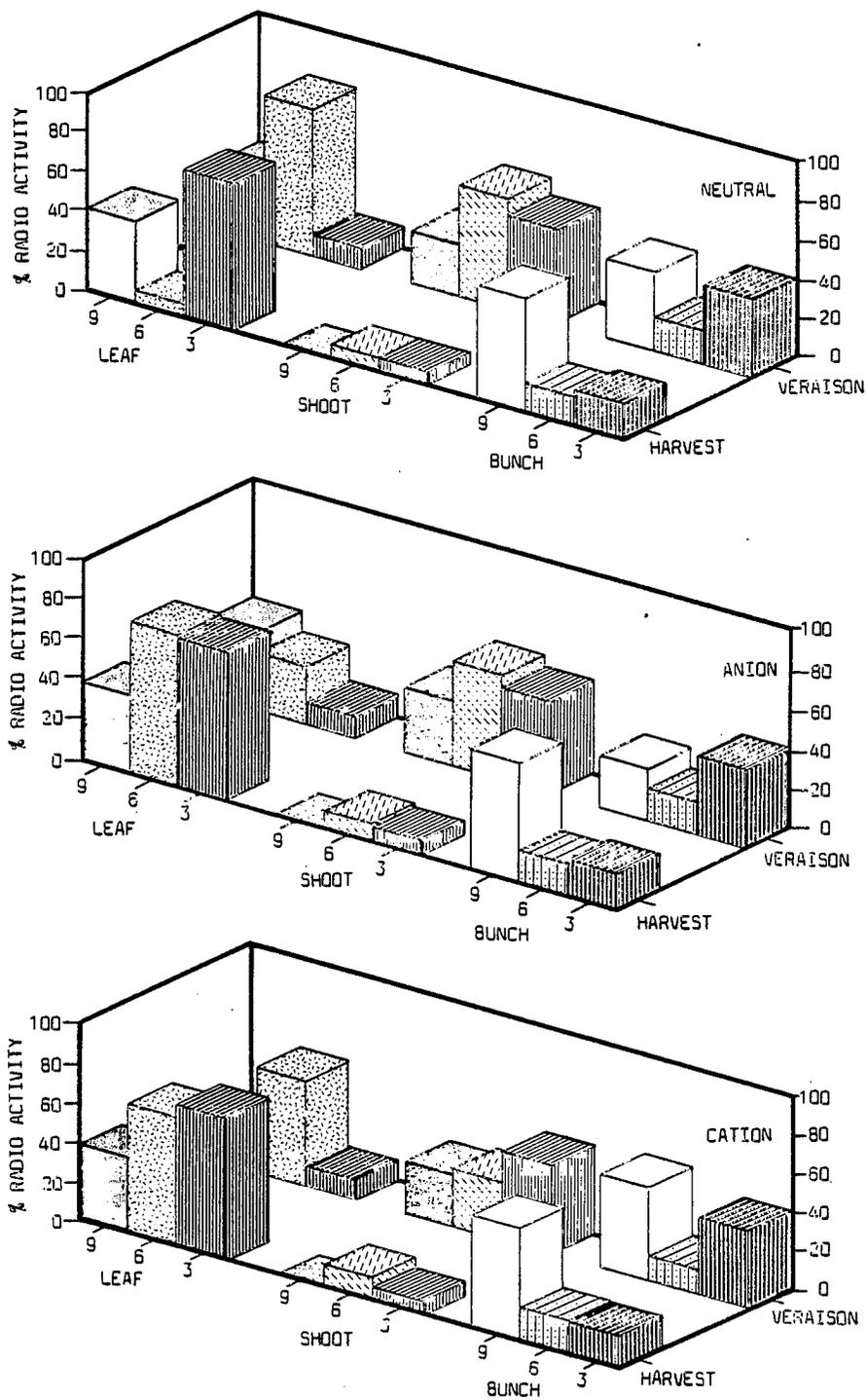


FIG. 9. The distribution of radio activity three, six and nine hours in the neutral, anion and cation fractions of the alcohol extract of the leaf, shoot and bunch expressed as a percentage of the basal treatment of the topped vines.

dency for the cation fraction was very similar, irrespective of the position of treatment. This indicated the cations to be a product of a metabolic process taking place in an organ rather than a translocation of cations.

At harvest, contrary to the situation at veraison, the main activity of all three fractions involved, were in the leaf, with very small %A in the shoots (Figs. 7 & 9). This indicated that translocation of all types of substances had virtually stopped at harvest. Nevertheless, some activity was still detected in the bunches. This activity was more or less evenly distributed between the three fractions and together with the fact that sucrose is the main photosynthetate being translocated (Amerine & Root, 1960), this might be explained as originating from respiratory and related pathways rather than from translocation of substances since little activity could be detected in the shoots.

CONCLUSION

At veraison translocation of photosynthetate takes place and organic compounds are freely moved in the vine. This is true for topped and untopped vines. At veraison more activity could be detected in the bunches of topped vines than in that of untopped vines. This would mean that topping affected a greater influx of metabolite into the bunches at veraison in comparison to the untopped vines.

At harvest translocation of photosynthetate was very slow. Although it is mentioned in Chapter 4 that little activity was detected in the berry at harvest time, the analyses of sugars showed an increase in the berries. This might be due to a redistribution of sugars that took place in the shoot. Contrary to the situation at veraison the main activity was situated in the leaves with a small percentage in the bunch. In contrast to the findings of some other

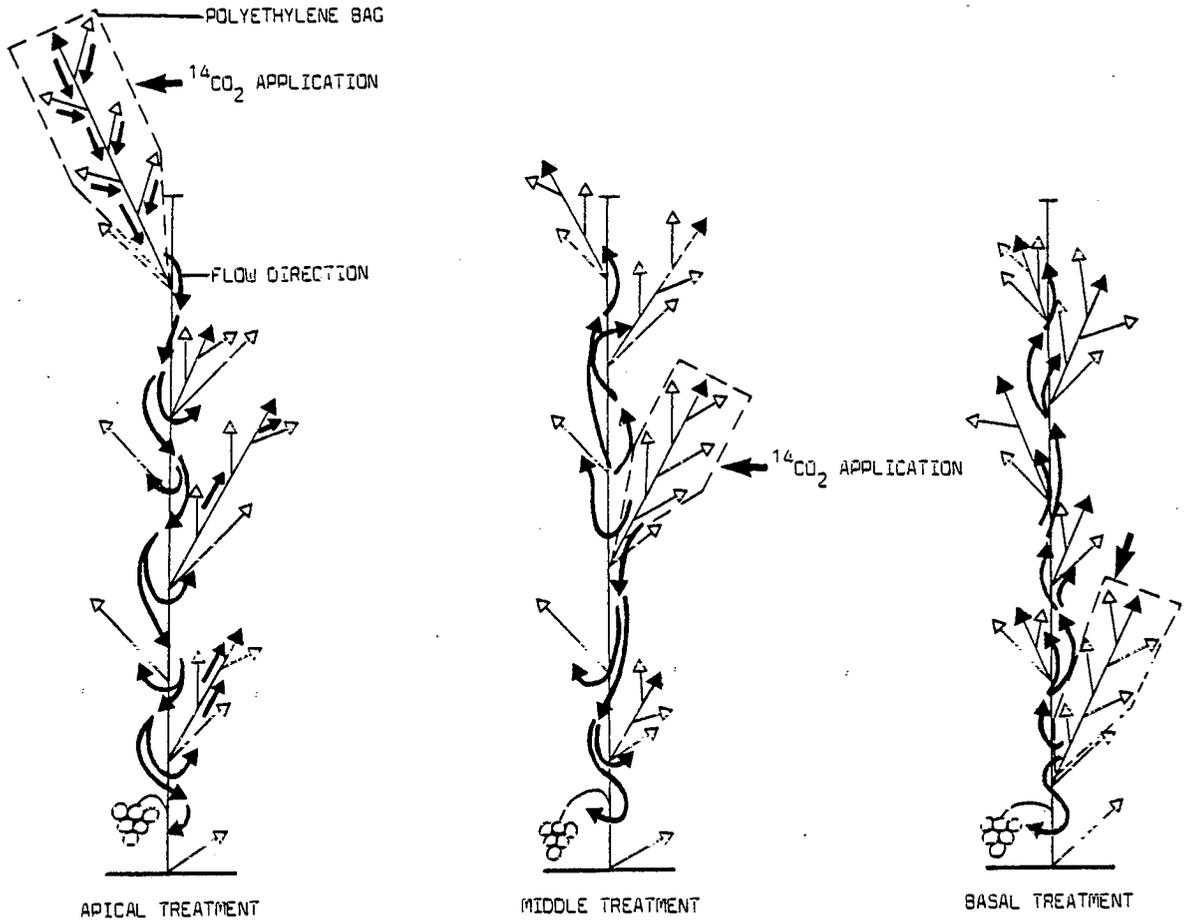


FIG. 10. The distribution of ^{14}C -photosynthetate in the leaf, shoot and bunch of *Vitis vinifera* L. cv. Cape Riesling after application of $^{14}\text{CO}_2$ to the apical, middle and basal lateral shoots and leaves.

authors multidirectional movement of photosynthetate took place independent of the position of exposure (Fig. 10).

The bunches of the topped vines still have a higher %A than the bunches from untopped vines, indicating the practical possibility of affecting grape composition and therefore quality, by a viticultural practice reasonably early in the season.

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CHAPTER 5

Accumulation of Sugars in the Berry of Vitis vinifera L. cv. Cape Riesling

ABSTRACT

The effect of topping on the sugar composition and accumulation in berries of Vitis vinifera L. cv. Cape Riesling during maturation was investigated. Glucose and fructose were found to be the major sugars in the berries. A glucose-fructose ratio of $1 \pm 0,1$ was obtained throughout the season. Topping had no effect on the total sugar concentration expressed on a dry mass basis. The total sugar concentration in the berries of topped and untopped vines increased initially and was followed by a decrease in fructose 76 days after bud break. Fructose started increasing again approximately 97 days after bud break and continued as the berries matured.

INTRODUCTION

As far as wine grapes are concerned sugar is the most important constituent of the fruit and is often used as a direct indicator of grape maturity (Amerine, 1956). Although galactose, sucrose, maltose, melibiose, raffinose and stachyose were found to occur in ripe berries, glucose and fructose are considered the predominant sugars in grapes (Amerine, 1956; Kliewer, 1965a; 1965b; Coombe, 1976).

Harris, Kriedemann & Possingham (1968) reported that a rapid decrease in acid concentration coincide with sugar accumulation in the berry. According to Coombe (1973) this accumulation of sugar is associated with the second rapid growth phase of the berry. The stage at which the sugar starts to accumulate is defined as veraison (Coombe, 1976). In contrast to peaches, grapes ripen from the outside inwards (i.e. from the skin towards the seed) resulting in a more intense accumulatory process of metabolites in the skin than in the pulp at the beginning of ripening (Coombe, 1976). Coombe & Matile (1980) reported that glucose is taken up more rapidly than fructose by the skin of the berry. These authors reported that these sugars might be transported into cell vacuoles of the skin. This was confirmed by Moskowitz & Hrazdina (1981) who identified glucose, fructose and sucrose in the vacuoles of the hypodermal layer of the berry.

Many papers have been published on the composition, concentration and seasonal development of sugars in the grape berry (Kliewer, 1965a; 1965b; 1967a; 1967b; Hardy, 1968; Kriedemann, 1969; Kliewer, 1970; Coombe, 1973; Patil & Gupta, 1973; Winkler et al. , 1974; Coombe, 1976; 1980). Concentrations of sugars in the berry vary considerably with variety, season, location, environment crop load

and stage of maturity (Kliewer, 1967a; 1967b).

Environment plays an important role in the development of grape quality (Rankine, *et al.*, 1971). Of these conditions it is mainly temperature which directly affects grape composition. Working with unripe berries Kliewer (1964) reported an increase in the concentration of glucose and a decrease in fructose concentration with an increase in temperature. Contrary to unripe berries, the glucose and fructose concentrations in ripe berries, were not affected by an increase in temperature.

Kliewer, Lider & Schultz (1969) found that by changing the environment of the vine by means of artificial shading, fruit maturation was delayed. Kliewer & Antcliff (1970) further found that leaf elimination treatments such as defoliation and darkening reduced berry mass and concentration of soluble solids. Klenert (1975) changed the climatic conditions of a vineyard by an artificial windbreak and shading during the period of berry set to fruit maturity. The windbreak did not have any measurable effects on grape composition. Shading, however, resulted in reduction of solar radiation and therefore, air, soil and vine temperatures were comparatively low in daytime. A decrease in temperature gave rise to a decrease in sugars in the berry.

The effect of topping on the vine was studied by Le Roux & Maian (1945). They reported a decrease in sugar concentrations in the berries of severely topped vines. These results were confirmed by Coombe (1959) who found that sugar concentration was decreased by one to two °B by topping or pinching. He attributed the decrease in sugar concentration to an increase in crop and/or a reduced leaf area. Since topping resulted in a doubling of leaf area one

would also expect to find a change in the micro climatic conditions near the bunch, and therefore possibly an effect on berry composition.

In Chapter 4 it is clearly illustrated that the topped vines transported more ^{14}C -labelled compounds to the bunch than the untopped vines. This might be caused by the enlarged leaf area due to topping (Chapter 3). This investigation was done to determine the effect of topping on the development of sugar in the grape.

MATERIAL AND METHODS

Material: Vitis vinifera L. cv. Cape Riesling vines were selected as described by De la Harpe & Visser (1983). Vines were topped and grapes sampled as described in Chapter 3.

Method: Whole bunches on the second spur of both cordons of three topped as well as three untopped vines were sampled 69, 76, 82, 92, 97, 110, 117, 131, 138, 145 and 152 days after bud break and stored at -20°C until analysed. Sugars were extracted from 30 randomly selected berries from each bunch with 70% ETOH and the extract centrifuged (50 000 g) for 15 min and subsequently determined by high performance liquid chromatography as described by Cloete (1980). Sugars were eluted from a Bio-rad HPX-87 column (300 x 7,8 mm) with water (flow rate $0,6 \text{ cm}^3 \cdot \text{min}^{-1}$) at 80°C .

The collected data was interpreted by a Vista 401 data handling system (Anon, 1980) and statistically analysed by applying an one way analysis of variance

BMDP-IV statistical programme on a Burroughs 7800 computer.

RESULTS AND DISCUSSION

As a result of the method used the only detectable sugars present in the berries of topped and untopped vines were glucose and fructose (Figs. 1a & b). Although Kliewer (1965a; 1965b) reported that in addition to glucose and fructose, galactose, sucrose, maltose, melibiose, raffinose and stachyose were also present in the berries, the method applied in this investigation did not reveal these sugars. Amerine & Root (1960) showed that sucrose is hydrolysed into glucose and fructose mainly in the pedicel, rachis and brush and is then transported into the berry which might explain the absence of sucrose in the berries during this study.

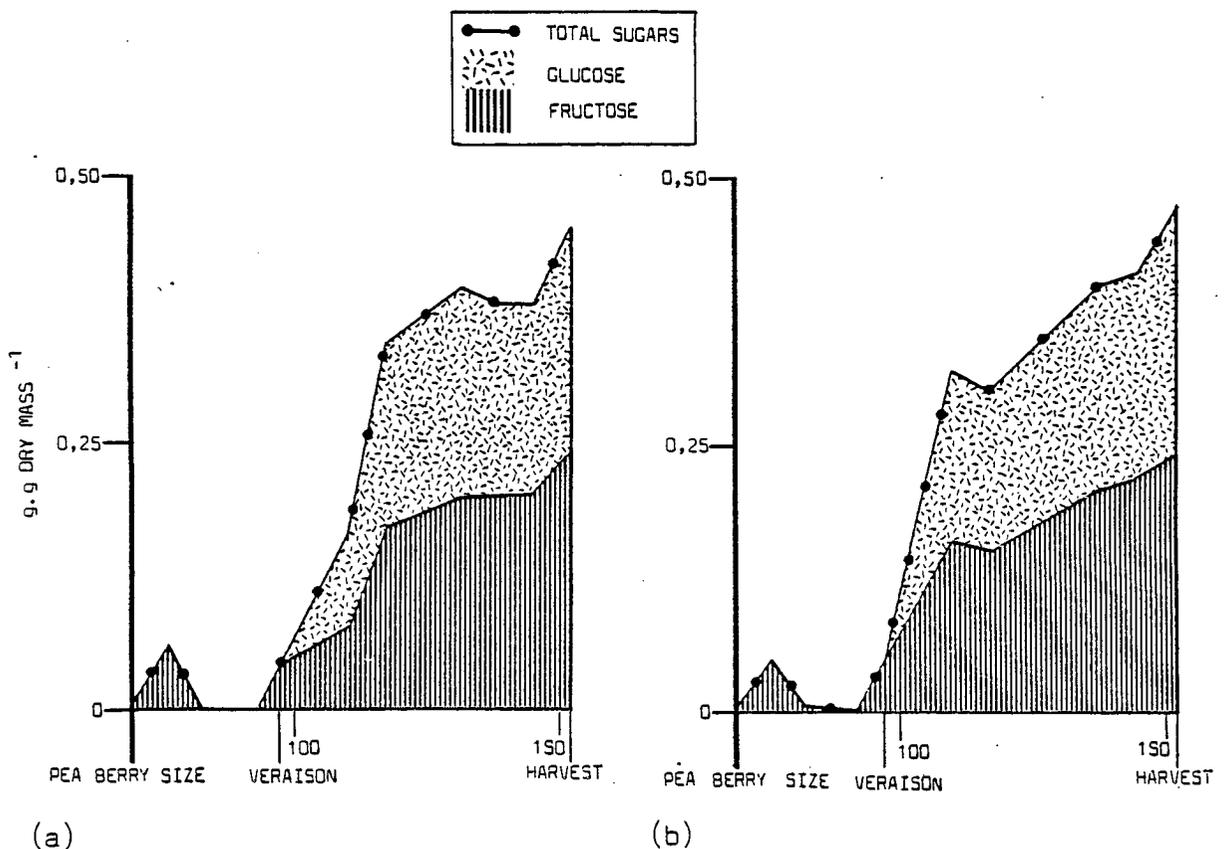


FIG. 1. Development of glucose and fructose in the berries of topped (a) and untopped (b) *Vitis vinifera* L. cv. Cape Riesling vines.

The berries started to accumulate glucose more or less 97 days after bud break. The accumulation is rapid for 14 days after which a levelling off became noticeable (Figs. 1a & b). Although the leaf area of topped vines were nearly double that of untopped vines during this stage of the season (Chapter 3) no significant differences in sugar concentration in the berries of topped and untopped vines could be found.

Although topping increased the leaf area by nearly 100% (Chapter 3), resulting in a higher influx of ^{14}C -labelled compounds to the bunches (Chapter 4) and a difference in the rate of sugar accumulation no difference could be found between the total sugar content of the berries from topped and untopped vines at harvest.

A possible explanation for this phenomenon might be the different sampling techniques used in the investigation. In the case of the ^{14}C -labelled compounds (Chapter 4) whole bunches were extracted for sugars whereas in this investigation only berries were used. This could mean that an appreciable amount of ^{14}C -sucrose might have been situated in the pedicels and rachis of the bunches (Amerine & Root, 1960) and not in the berries as such. Since the sucrose in the pedicel and rachis is hydrolysed to glucose and fructose and then translocated into the berry, only glucose and fructose were detected in the berry.

According to Meynhardt, De Villiers & Ireland (1974) sucrose may enter the berry, but the fast breakdown rate of sucrose to glucose and fructose by β -fructofuranosidase results in the observation of the latter only.

Glucose was found only in the skin and pulp and did not occur in the seed (Figs. 2a & b). In the pulp of berries from topped vines, glucose only started to accumulate after about 110 days. This delay of glucose accumulation in the pulp of topped vines cannot be explained at present.

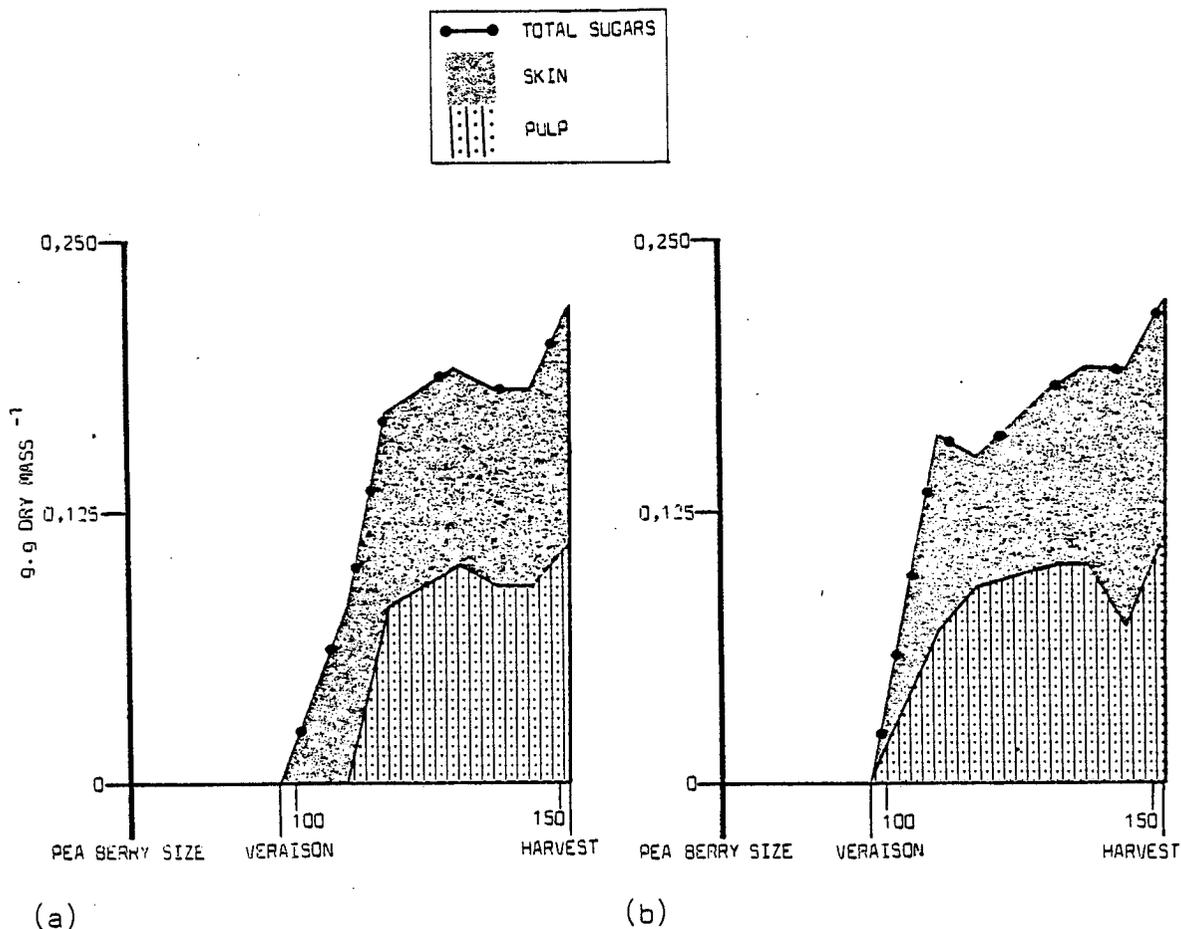


FIG. 2. Development of glucose in the skin and pulp of berries from topped (a) and untopped (b) *Vitis vinifera* L. cv. Cape Riesling vines.

In contrast to glucose, fructose started to accumulate at about 70 days after bud break to reach a peak six days later, then decreased till 92 days after bud break. Thereafter, a rapid increase took place (Figs. 3a & b). These results are in accordance with those of Coombe & Matile (1980) who reported that fructose concentrations are higher than glucose concentrations in the skin of pre-veraison berries. A possible explanation for the presence of fructose in the berry at this stage involves a possible preferential breakdown of glucose in the berry by way of the pentose cycle whilst the fructose accumulates (Kliewer, 1967b).

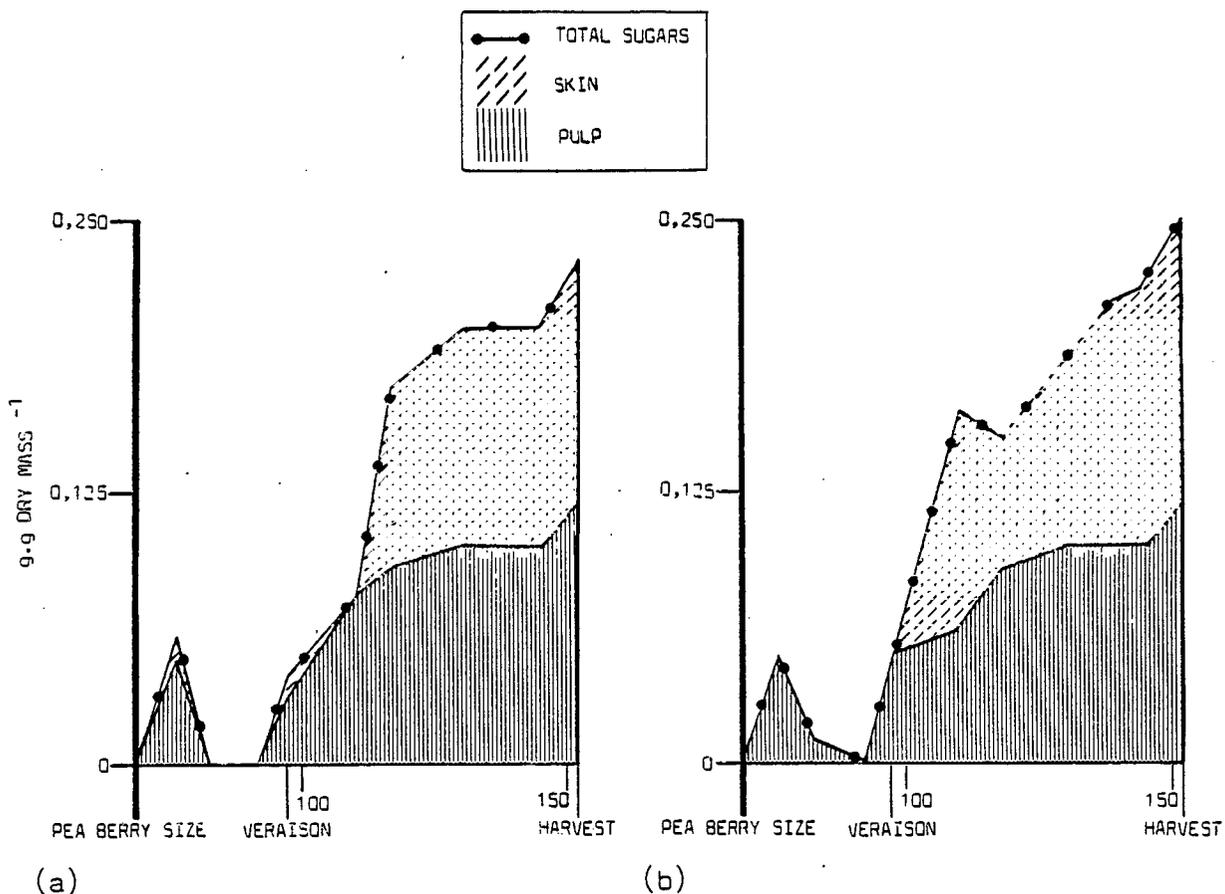


FIG. 3. Development of fructose in the skin and pulp of berries from topped (a) and untopped (b) *Vitis vinifera* L. cv. Cape Riesling vines.

As in the case of glucose in the skin, the increase in fructose concentration reached a peak 97 days and 110 days after bud break for topped as well as untopped vines (Figs. 3a & b). Topping had no significant effect on the composition and concentration of sugars in any part of the berries.

Except for the fructose peak 76 to 82 days after bud break in both the topped and untopped vines, accumulation of sugars started about 97 days after bud break (Table 1). The two sugars were evenly divided between the skin and pulp when

Table 1. The distribution of glucose and fructose in the berry expressed as a percentage of the total sugars and the glucose-fructose ratio over the season

Sampling Date	Berry		Skin		Pulp		Fructose		Glucose		Berry C/F	Skin C/F	Pulp C/F
	Fructose	Glucose	Fructose	Glucose	Fructose	Glucose	Skin	Pulp	Skin	Pulp			
69U	0	0	0	0	0	0	0	0	0	0	0,00	0,00	0,0
T	0	0	0	0	0	0	0	0	0	0	0,00	0,00	0,0
76U	100	0	0	0	100	0	0	100	0	0	0,00	0,00	0,0
T	100	0	100	0	100	0	14	86	0	0	0,00	0,00	0,0
82U	100	0	0	0	100	0	0	100	0	0	0,00	0,00	0,0
T	0	0	0	0	0	0	0	0	0	0	0,00	0,00	0,0
92U	0	0	0	0	0	0	0	0	0	0	0,00	0,00	0,0
T	0	0	0	0	0	0	0	0	0	0	0,00	0,00	0,0
97U	100	0	0	0	100	0	0	100	0	0	0,00	0,00	0,0
T	100	0	100	0	100	0	25	75	0	0	0,00	0,00	0,0
110U	50	50	49	49	49	51	63	37	56	44	1,00	0,90	0,9
T	50	50	0	0	50	50	0	100	0	100	1,00	0,00	0,0
117U	50	50	50	50	50	50	34	66	40	60	1,00	1,00	1,0
T	50	50	49	51	51	49	49	51	51	49	1,00	0,89	0,9
131U	51	49	50	50	50	50	49	51	49	51	0,95	1,00	1,0
T	51	49	51	49	50	50	50	50	49	51	0,95	0,90	1,0
138U	53	47	50	50	50	50	50	50	50	50	0,90	1,00	1,0
T	53	47	51	49	51	49	50	50	50	50	0,90	0,90	0,9
145U	54	46	51	49	59	51	55	45	65	35	0,86	0,90	0,7
T	53	47	51	49	51	49	50	50	50	50	0,90	0,90	0,9
152U	53	47	51	49	51	49	51	49	51	49	0,88	0,90	0,9
T	53	47	51	49	51	49	49	51	51	49	0,91	0,90	0,9

G/F = Glucose-fructose ratio.

U = Untopped vines.

T = Topped vines.

expressed as a percentage. A glucose-fructose ratio of $1 \pm 0,1$ was obtained throughout this investigation, from veraison to harvest (Table 1). This is to be expected since sucrose is hydrolysed in the pedicel, rachis and brush and then translocated as glucose and fructose into the berry (Amerine & Root, 1960). Topping did not affect this ratio.

An explanation for the different rates of accumulation of glucose and fructose in the berry might be due to limited translocation into the berry as well as accumulation in the pericarp cells (Coombe, 1973). These limitations might indicate that although topping in actual fact do increase the rate of sucrose influx into the bunch, the berry itself cannot store the additional available photosynthetate. Coombe & Matile (1980) showed that the rate of sugar accumulation from a glucose solution by a peeled skin was higher than that of whole berries and it seems that the experimental procedure of peeling and incubating of the skin segments plus the glucose medium in some way removed an inhibitor of the sugar transport process. Much more research needs to be done in this field to give a better understanding of the translocation pathways into the berry.

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ABSTRACT

A method for the determination of organic acids in grapes is described. Organic acids are extracted from the material with 70% ETOH, purified by anion exchange and analysed by High Performance Liquid Chromatography. The proposed method gives high percentages of recovery for the different acids and is reproducible. Further advantages are the relatively short extraction and analysis time as well as the high resolution obtained between the organic acids.

INTRODUCTION

Organic acids play an important role in viticulture and are of special importance in grape quality and wine making. The importance of organic acids in grapes and wine is emphasized by the large number of papers published on this subject over the past decades.

A number of methods for determining organic acids have been published. Amerine & Winkler (1942) determined the acids by means of titration using phenolphthalein as indicator while Harlik, Marshall & Lodge (1959) used a quantitative micro-diffusion technique to determine formic, acetic and butyric acid.

After the introduction of chromatography numerous methods were developed for determining organic acids. In 1946 Isherwood introduced partition chromatography and Busch, Hurlbert & Potter (1952) anion exchange chromatography as an aid to the determination of organic acids in foods. Thin layer (Bourzeix, Guitraud & Champagnol, 1970), paper (Bryant & Overell, 1953; Hardy, 1968; Williams & Loewus, 1978), ion exchange (Busch, Hurlbert & Potter, 1952), gas (Carwin, 1965; Saito & Kasai, 1968; Anderson *et al.*, 1970; Moskowitz & Hrazdina, 1981) and liquid chromatography (Donaldson, Tulane & Marshall, 1952; Marshall, Orten & Smith, 1948; Marshall, Donaldson & Friedberg, 1952; Zbinovsky & Burnis, 1954; Freeman, 1967) as well as electrophoresis (Gross, 1959; Michl & Högenauer, 1959) have all been used to determine organic acids, but with some restriction (Stahl, Schäfer & Lamprecht, 1972). Some of these determinations were carried out with chemically pure organic acids (Kaiser, 1973; Hyakutake & Hanai, 1975;

Ong & Nagel, 1978) while Palmer & List, (1973); Düring & Bachmann, (1975), and Moskowitz & Hrazdina (1981) applied the above mentioned techniques on foods, musts and wines. The results obtained by different workers, is virtually impossible to compare because the reproducibility of determinations as well as recovery percentages are seldom given.

The purpose of this study was to develop a simple reproducible technique with high recoveries to extract acids from different parts of the grape berry at different developmental stages for analyses by HPLC.

METHOD

Five replicates of one gram each of freeze-dried skin, pulp and seed of Vitis vinifera L. cv. Cape Riesling berries were extracted by vigorously shaking the individual samples for 20 min in 50 cm³ 80% ETOH. A duplicate set to which known quantities of pure organic acids were added, was extracted simultaneously. Percentage recovery was calculated as $\frac{A - B}{Q} \times 100$ where A is the concentration of individual organic acids extracted from a sample after addition of known quantities of the different organic acids, B the concentration of individual organic acids extracted from a sample before addition of known quantities of the different organic acids expressed and Q the concentration of organic acids added to sample.

All the extracts were filtered through 0,22 µm Millipore filters and the extraction repeated by washing the residue on the filter.

The acids were recovered from the extract by adsorption on Amberlite AG 400

(200 - 400 mesh) anion resin in the Cl^- form. One cm^3 resin was then washed with 100 cm^3 deionised water and eluted with one cm^3 10% H_2SO_4 solution followed by distilled water to make up 25 cm^3 .

The organic acids were then analysed on a Varian 5000 liquid chromatograph fitted with a UV- detector set at 200 nm on a Biorad HPX-87 column. As mobile phase 0,013 N H_2SO_4 was pumped at a rate of $0,3 \text{ cm}^3 \cdot \text{min}^{-1}$ at 65°C at a pressure of 8000 kPa. Data was collected and interpreted by a Vista 401 data handling system (Anon, 1980).

RESULTS AND DISCUSSION

A typical chromatogram of the organic acids present in the extract is depicted in Fig. 1.

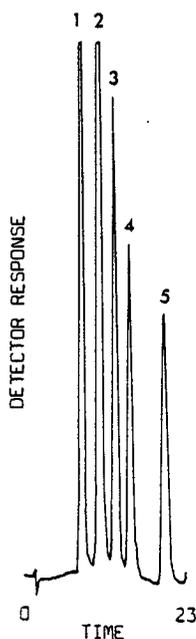


FIG. 1. Separation of organic acids with detection at 200 nm. 1 = citric acid; 2 = tartaric acid; 3 = malic acid; 4 = succinic acid; 5 = acetic acid.

Skin: It is evident from Table 1 that citric, tartaric and malic acid, are not completely extracted by the first extraction with alcohol. After a second extraction no acids could be found with further extractions. The percentage acid of the filtrate residue (FR) was never more than 10% and could be accounted for by a second extraction. The calculated loss of acid (i.e. after adding the PR (percentage recovery) and FR) may be due to insoluble salt formation and experimental losses eg. by washing, although different washing techniques did not improve the results.

Pulp: The PR for citric, tartaric and malic acid was $78 \pm 8,6\%$, $104 \pm 5,7\%$ and $97,4 \pm 19\%$ respectively. In the case of pulp only tartaric acid had an FR of $2,7 \pm 1,9\%$.

Seed: As in the case of the pulp PR values of $72,0 \pm 5,0\%$, $88,0 \pm 9,0\%$ and $84,0 \pm 14,0\%$ were obtained for citric, tartaric and malic acids respectively. Filtrate residue analyses of $1,8 \pm 4\%$ and $2,2 \pm 5,0\%$ were obtained for tartaric and malic acid.

The proposed method gives high percentages of recovery for the different acids and yields reproducible results, with the SD only in a few instances exceeding ± 10 . Further advantages of this method are the relatively short extraction (20 min) and analysis time (20 min) on the HPLC facilitating the handling of at least 50 samples in 24 hrs.

TABLE 1

The percentage recoveries and residual activities of the acids as obtained in the different parts of the grape berry

Replication	Citric Acid		Tartaric Acid		Malic Acid	
	Percentage Recovery	FR	Percentage Recovery	FR	Percentage Recovery	FR
	<u>Skin</u>					
1	85,0	0	85,0	10	78,0	2
2	70,0	0	66,0	9	81,0	2
3	78,0	3	84,0	10	78,0	3
4	88,0	3	88,0	8	99,0	3
5	92,0	8	91,0	10	85,0	4
Mean	82,6 ± 8,7	2,8 ± 3	82,8 ± 9,8	9,4 ± 0,9	84,2 ± 8,8	2,8 ± 0,8
	<u>Pulp</u>					
1	76,0	0	103,0	2,5	109,0	0
2	88,0	0	109,0	5,5	106,0	0
3	65,0	0	96,0	3,9	72,0	0
4	78,0	0	110,0	0,8	82,0	0
5	85,0	0	102,0	1,0	118,0	0
Mean	78,0 ± 8,6	0	104,0 ± 5,7	2,7 ± 1,9	97 ± 19	0
	<u>Seed</u>					
1	68,0	0	80,0	0	67,0	0
2	67,0	0	78,0	0	78,0	0
3	72,0	0	92,0	0	99,0	0
4	80,0	0	98,0	0	99,0	0
5	75,0	0	92,0	9	77,0	0
Mean	72 ± 5	0	88 ± 9	1,8 ± 4	84 ± 14	2,2 ± 5

FR = Filtrate residue.

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CHAPTER 7

Development of Organic Acids in the berry of Vitis vinifera L. cv. Cape Riesling

ABSTRACT

The effect of topping on the organic acid composition of the berries (skin, pulp and seed) of Vitis vinifera L.cv. Cape Riesling during ripening was investigated. The total acid concentration decreased as ripening progressed. Tartaric acid and malic acid were found to be the most important acids followed by citric, succinic and acetic acid. Topping had no effect on the total acid concentration in the berry expressed on a dry mass basis.

Major acids in the skin at harvest (21^oB) were citric, tartaric and malic acid while citric, tartaric, malic, succinic and acetic acid were present in the pulp. Except for citric acid the same acids as in the pulp were also found in the seeds.

Topping had no measurable effect on either the developmental pattern or the acid composition of the grapes during ripening in the same season in which it was performed.

INTRODUCTION

Organic acids are of great importance in the grape and has led to many studies on their presence and developmental pattern (Amerine & Winkler, 1942; Amerine, 1951 ; 1956; Amerine & Winkler, 1958; Kliewer, 1965 ; 1966; Kliewer & Nassar, 1966; Kliewer, 1967; Kliewer, Howarth & Omori, 1967; Du Plessis, 1968; Hale, 1968; Coombe, 1978; Takimoto, Saito & Kasai, 1976; Hale, 1977; Ruffner, 1982a ; 1982b). It is generally accepted that tartaric and malic acid are the predominant acids in grapes (Ruffner, 1982a ; 1982b). The acid content of the grapes is very high at the early stages of development but decreases rapidly during maturation of the berry. Normally, the berries have their highest malic and tartaric acid concentration just prior to veraison (Kliewer, 1965). These acids are not evenly distributed throughout the berry and it was shown by Amerine (1956) that at maturity the lowest concentration of acids is found in the hypodermis, the highest concentration around the seed, while the pulp has an intermediate concentration. In young developing berries, the highest concentration of acids is found in the hypodermis.

The effects of yield levels (Kliewer & Weaver, 1971; Balasubrahmanyam, Eifert & Diofasi, 1979), topping, girdling and defoliation (Le Roux & Malan, 1945; Coombe, 1959; Kliewer & Antcliff, 1970), irrigation (Kliewer & Schultz, 1973; Van Zyl & Van Huysteen, 1980) and the environment (Kliewer, 1964; Kliewer & Schultz, 1964; Klenert, 1975; Klenert, Rapp & Alleweldt, 1978) on the organic acid content have also been studied. Hennig & Burkhardt (1951) reported higher malic acid concentrations in cooler years but Amerine (1951) and Du Plessis

(1968) could not confirm this under their environmental conditions. In his review Ruffner (1982a ; 1982b) demonstrated clearly that the acid content of ripening grapes is mediated by exogenous factors such as temperature and pointed out that malic acid accumulation is favoured by cooler temperatures. Le Roux & Malan (1945) showed that severely topped vines (50% of the shoot removed) resulted in a decrease of the sugar content. Kliewer & Antcliff (1970) reported that leaf elimination treatments led to higher acidity in the grapes.

In South Africa little is known about the effect of summer pruning on the development and localization of organic acids in the berry. The aim of this study was to investigate the effect of topping on the seasonal development of organic acids and its localization in the berry.

MATERIAL AND METHODS

Material: *Vitis vinifera* L. cv. Cape Riesling vines were selected as described by De la Harpe & Visser (1983). Vines were topped and grapes sampled as described in Chapter 3.

Methods: Organic acids in grapes from three vines per treatment per sampling date were determined using liquid chromatography as described in chapter 6.

The collected data was interpreted by a Vista 401 data handling system (Anon, 1980) and statistically analysed by applying an one way analysis of variance BMDP-IV statistical programme on a Burroughs 7800 computer.

RESULTS AND DISCUSSION

Total organic acids in the berry: The total acid concentration in the grapes of topped and untopped vines is depicted in Figs. 1 and 2. Five organic acids namely citric, tartaric, malic, succinic and acetic acid were found in the grapes. The total acid concentration had high values early in the season for both the topped and untopped vines (Figs. 1). Approximately 90 days after bud break rapid decrease in total acid concentration occurred until 117 days after bud break at which an increase of acids took place before a final decrease to harvest. No statistically significant differences were obtained between the acid concentrations in the berries of topped and untopped vines.

Although the initial concentrations of the acids differ, they all follow similar patterns in that the concentration is high at pea berry size, starts decreasing at veraison and continues to decrease as the grape matures (Fig. 1).

Malic acid concentration increases from pea berry size to veraison whereafter it decreases for both the topped and untopped vines. Contrary to malic acid, tartaric, succinic, citric and acetic acid decreased from pea berry size to maturity.

The decrease in acid concentration occurred mainly during the warmer weeks (25°C - 32°C , Fig. 3), with the highest concentration at the lowest temperature (23°C) which is in accordance with reports of Ruffner (1982a ; 1982b). A very active vegetative growth phase (Chapter 3) preceded an increase in total acids from pea berry size to veraison indicating a sink-source shift from the vegetative growth to the reproductive growth phase.

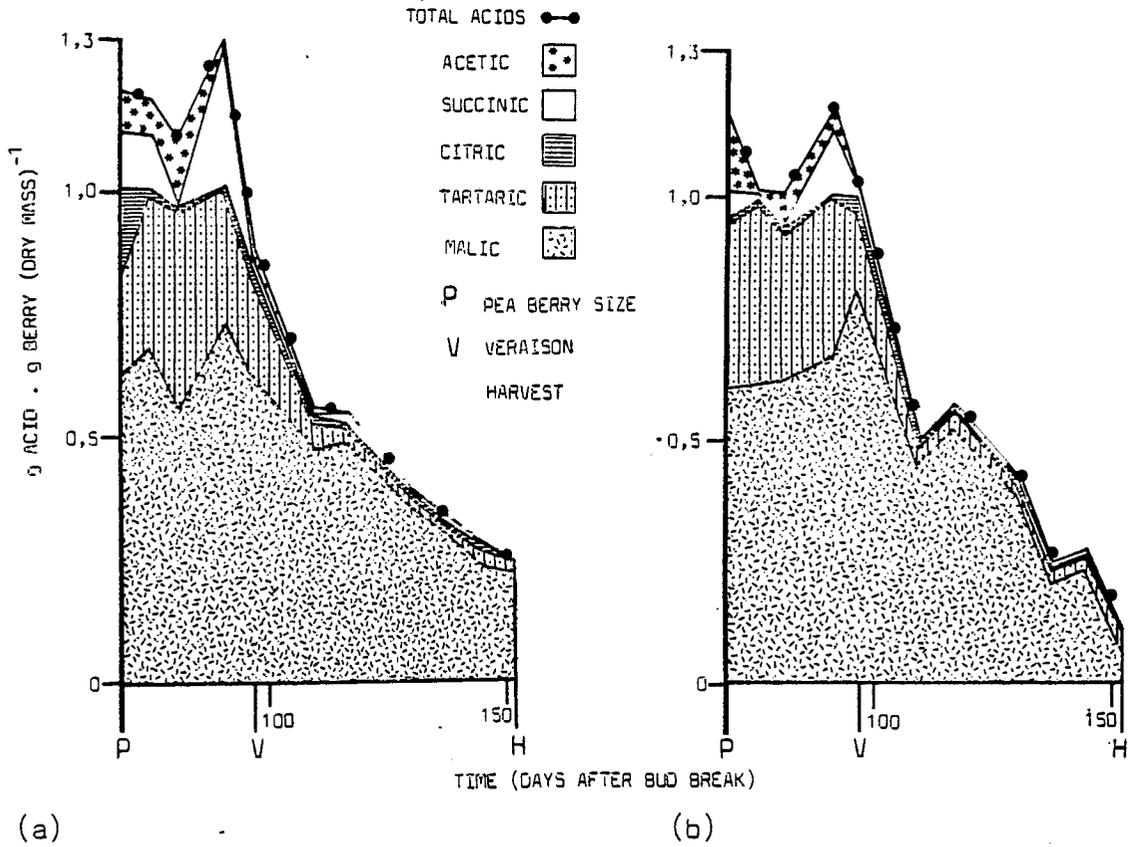


FIG. 1. Seasonal decrease of organic acids in the berries of topped (a) and untopped (b) *Vitis vinifera* L. cv. Cape Riesling vines.

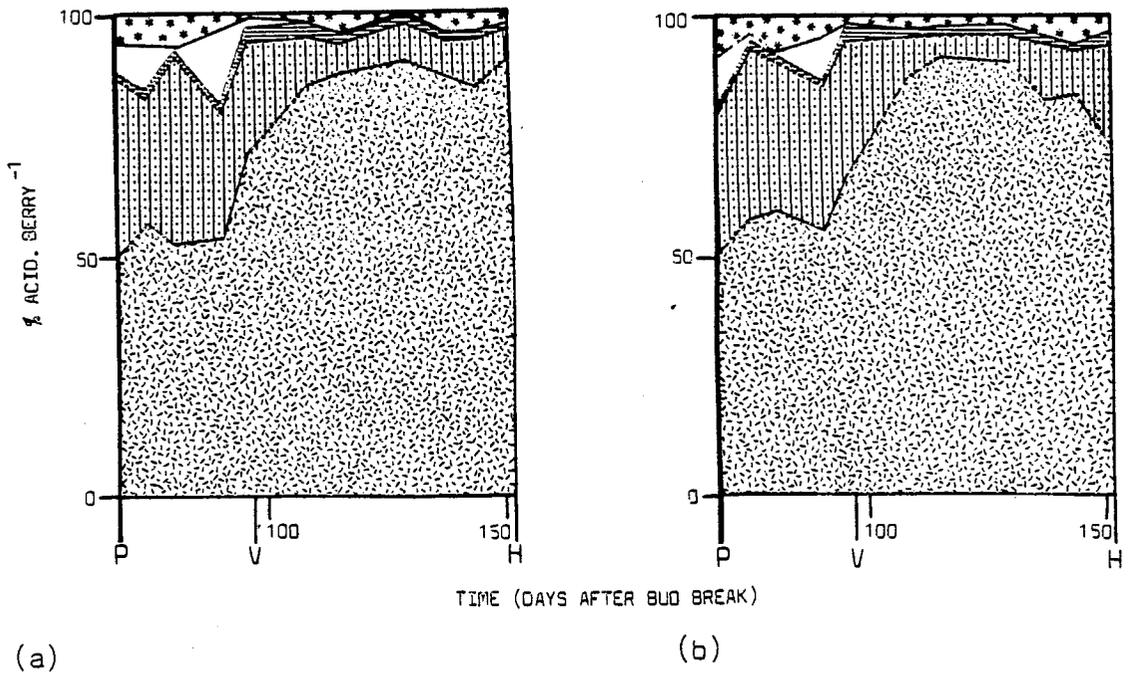


FIG. 2. The percentage acids in the berries of topped (a) and untopped (b) *Vitis vinifera* L. cv. Cape Riesling vines.

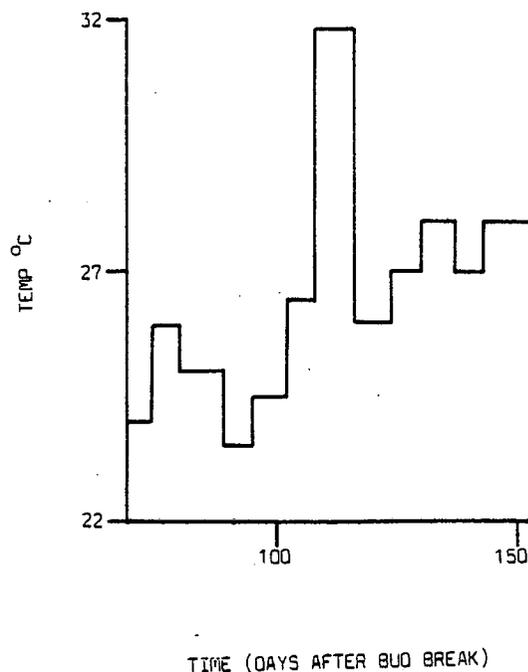


FIG. 3. Weekly temperatures from bud break to harvest.

The topped and untopped vines showed the same developmental pattern and although it was established that topping results in a doubling of total shoot length and leaf area (Chapter 3) and therefore theoretically enlarging the photosynthetic capacity, no statistically significant differences between the acids of the topped and untopped vines were obtained.

The fact that there were no significant differences gave rise to the postulation that the nutrients such as sugars and acids were metabolized by the vegetative growth, or that although these nutrients were translocated to the berries (Chapter 4) the berries themselves lacked the ability to store all the nutrients. Further factors affecting this phenomenon might be a possible increase in membrane permeability allowing acids to be respired and a reduced ability of the berry to synthesize acids as the berry ripens (Hardy, 1968). A further possibility is the formation of salts as reported by Kliewer (1971).

After an analyses of the acids it became evident that tartaric and malic acid were dominant and accounted for 90 to 95% of the acids in the grape

for both the topped and untopped vines. This confirmed the results on the acid composition of the berry reported by Amerine & Winkler (1942), Amerine, (1951 ; 1956), Kliewer, (1964), Kliewer & Schultz, (1964), Kliewer, (1967), Kliewer, Howarth & Omori, (1967), Du Plessis, (1968), Kliewer, (1971), Philip & Kuyker, (1973), Coombe, (1976), Cash, Sistrunk & Stutte, (1977) and Ruffner, (1982a ; 1982b). The concentration of citric, succinic and acetic acids were too low to have had an effect on the tendencies obtained with respect to total acid concentration.

The rapid loss in tartaric acid coincided with the last stage of growth phase II of the berry (Chapter 3).

It is well known that acids are not evenly distributed throughout the berry (Winkler, et al., 1974). In the present experiment it was found that the pattern of acid increase and decrease was similar for skin, pulp and seed (Figs. 4 & 5). The skin was characterised by the absence of succinic acid from veraison to harvest. The total acids in the skin showed a linear decrease from 0,44 g.g skin dry mass⁻¹ (untopped vines) and 0,38 g.g skin dry mass⁻¹ (topped vines) to 0,07 and 0,11 g.g skin dry mass⁻¹ respectively at full ripeness (Fig. 6a).

Tartaric and malic acids were the only acids present in concentrations of any practical importance and citric and acetic acid contributed less than one percent to total acid concentration in the skin. Contrary to tartaric acid, malic acid concentration initially increased in the case of both the topped and untopped vines, before decreasing as the berry continues to full ripeness (Fig. 6).

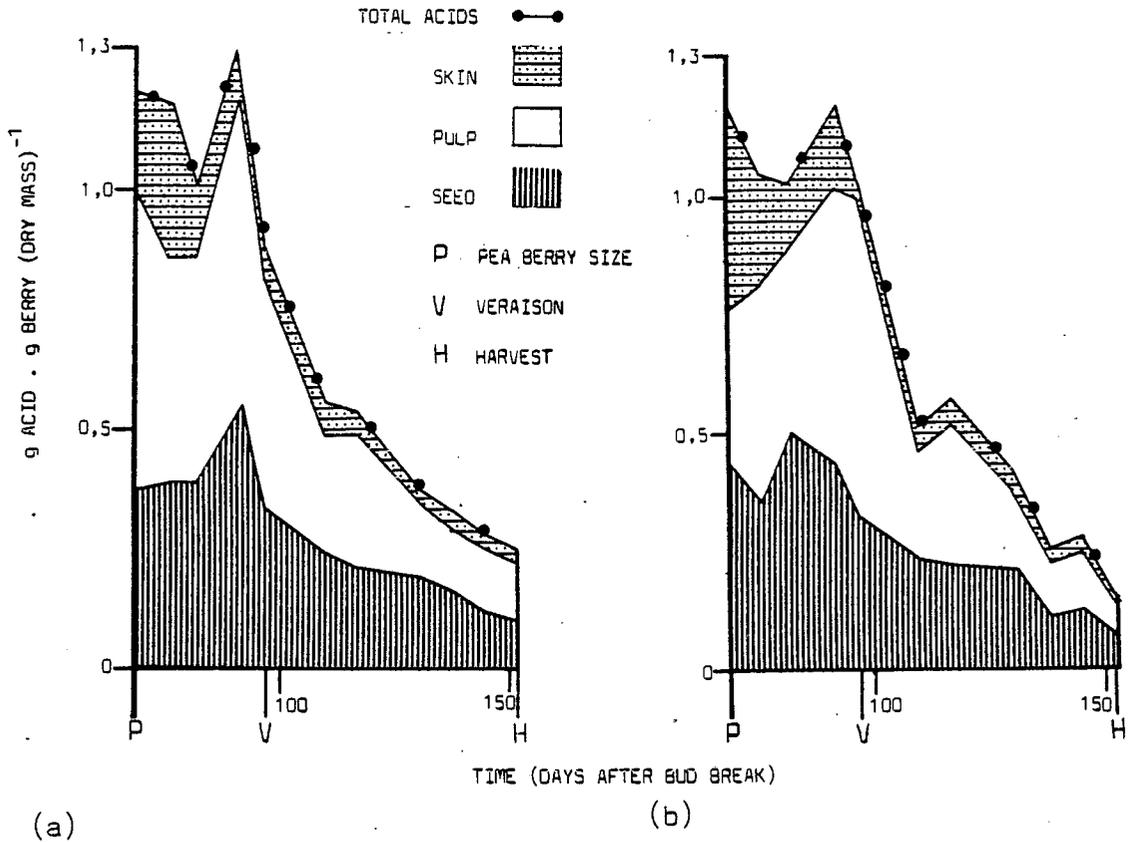


FIG. 4. Seasonal changes of the acid in the skin, pulp and seed of berries from topped (a) and untopped (b) *Vitis vinifera* L. cv. Cape Riesling vines.

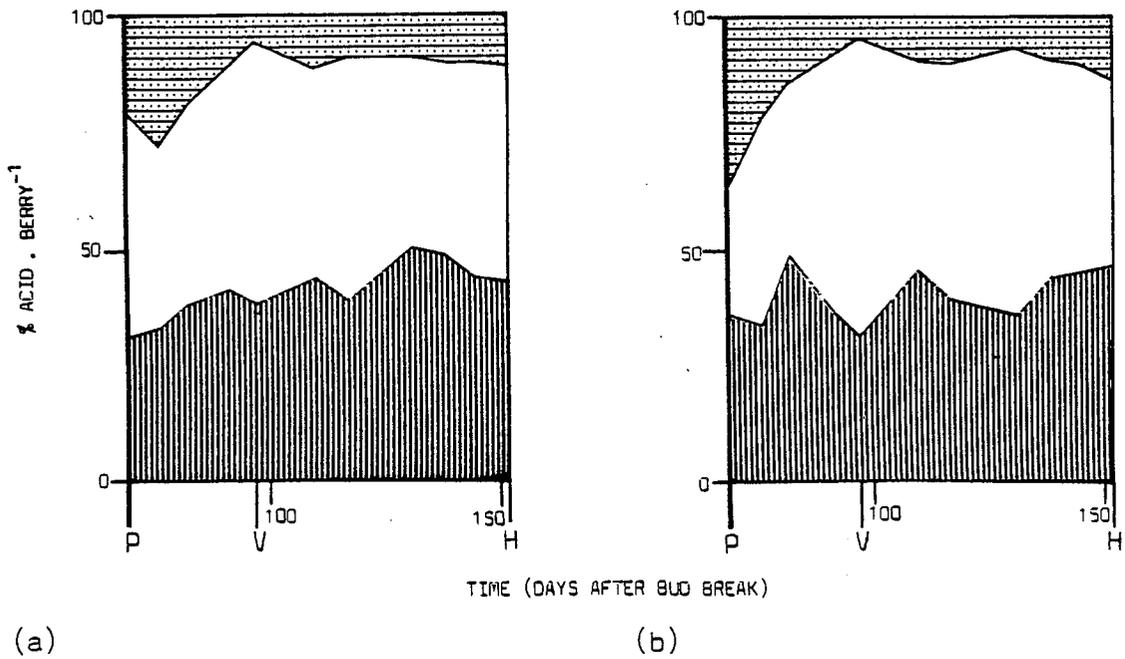


FIG. 3. The percentage acid in the skin, pulp and seed of berries from topped (a) and untopped (b) *Vitis vinifera* L. cv. Cape Riesling vines.

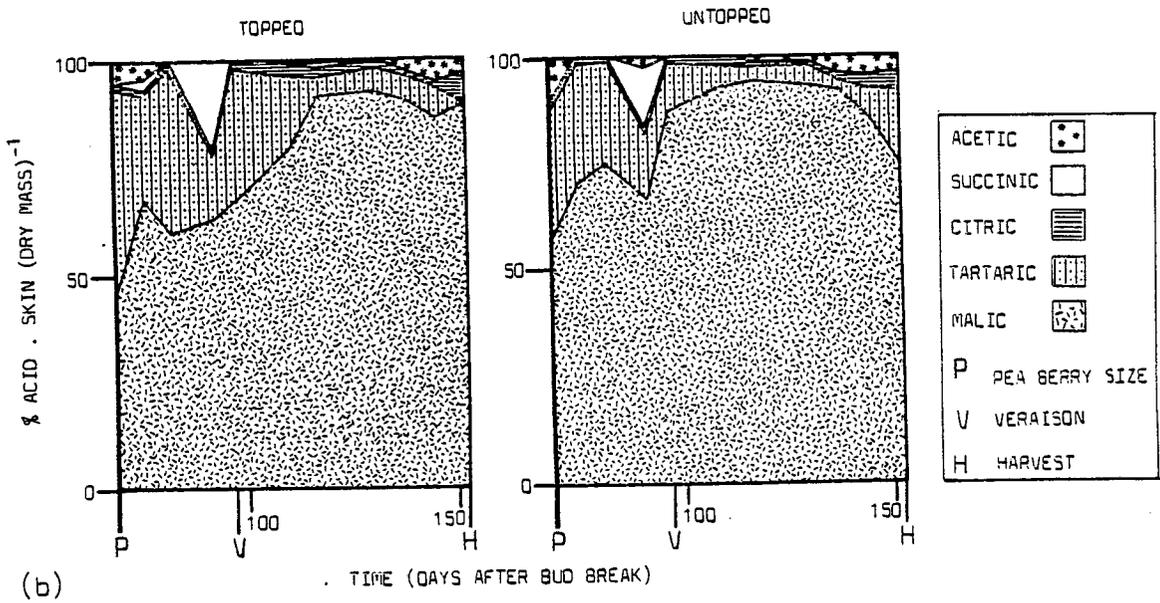
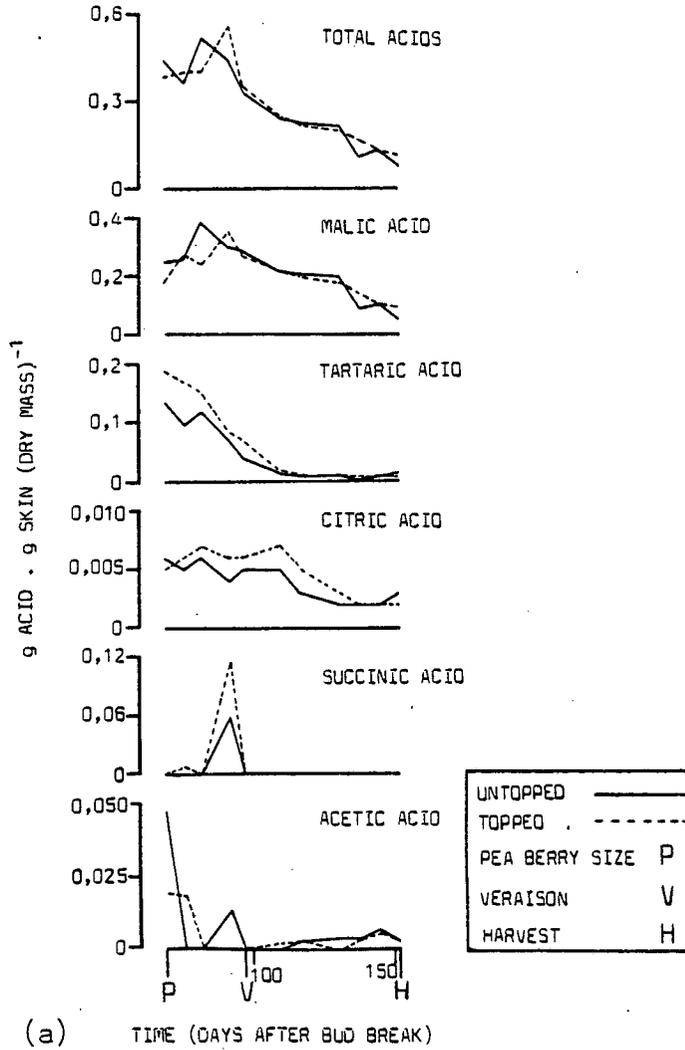


FIG. 6. Seasonal (a) changes and percentage contribution (b) of the organic acids in the skin of berries from topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines.

No statistical differences between topped and untopped vines could be found in the data set concerning the acid concentrations in the skin.

Citric, tartaric, malic, succinic and acetic acids were present in the pulp and showed an initial increase in concentration which is in contrast to the pattern found for the skin. After this initial increase, the acid concentration decreased from veraison to harvest (Fig. 7). Expressed as a percentage of total acids in the pulp, citric, succinic and acetic acids remained constant, tartaric acid decreased and malic acid decreased slightly at first followed by a rapid increase at veraison and then stayed constant until harvest (Fig. 7) for both the topped and untopped vines without any significant differences between them.

The seeds were characterised by the virtual absence of citric acid. The concentration of the other four acids in the seed were relatively high in both topped and untopped vines at pea berry size after which a rapid decrease at veraison occurred (Fig. 8).

Previously only tartaric and malic acid were regarded as the dominant acids but in this study acids like succinic and acetic were found to be prominent in the seed especially in the early stages of ripening (Fig. 8). At harvest, tartaric and malic acid were the most dominant acids. As in all the discussed cases, topping had no effect on the acid composition of the seed.

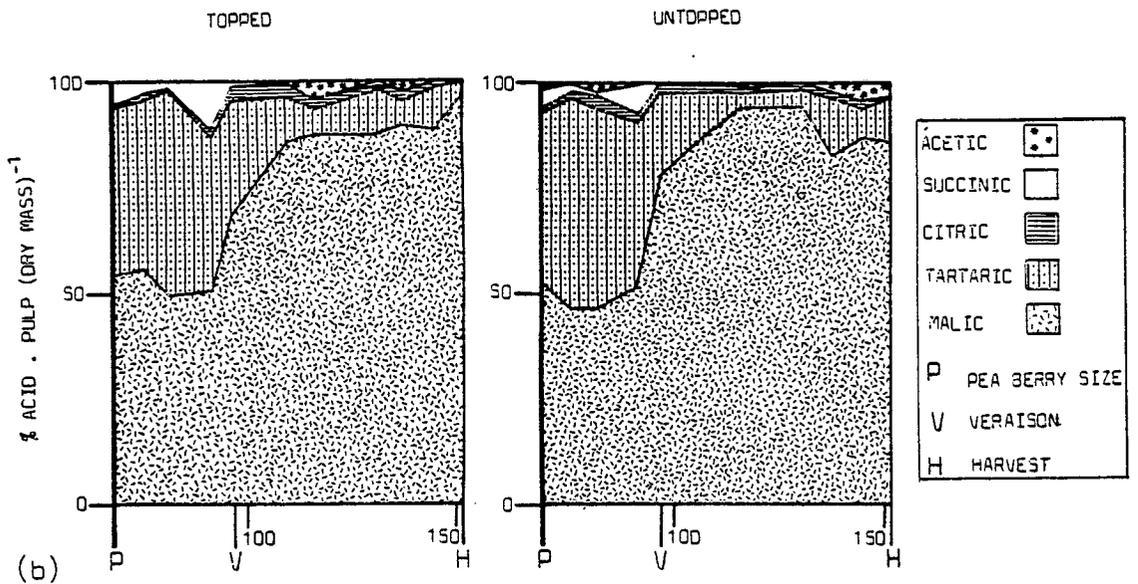
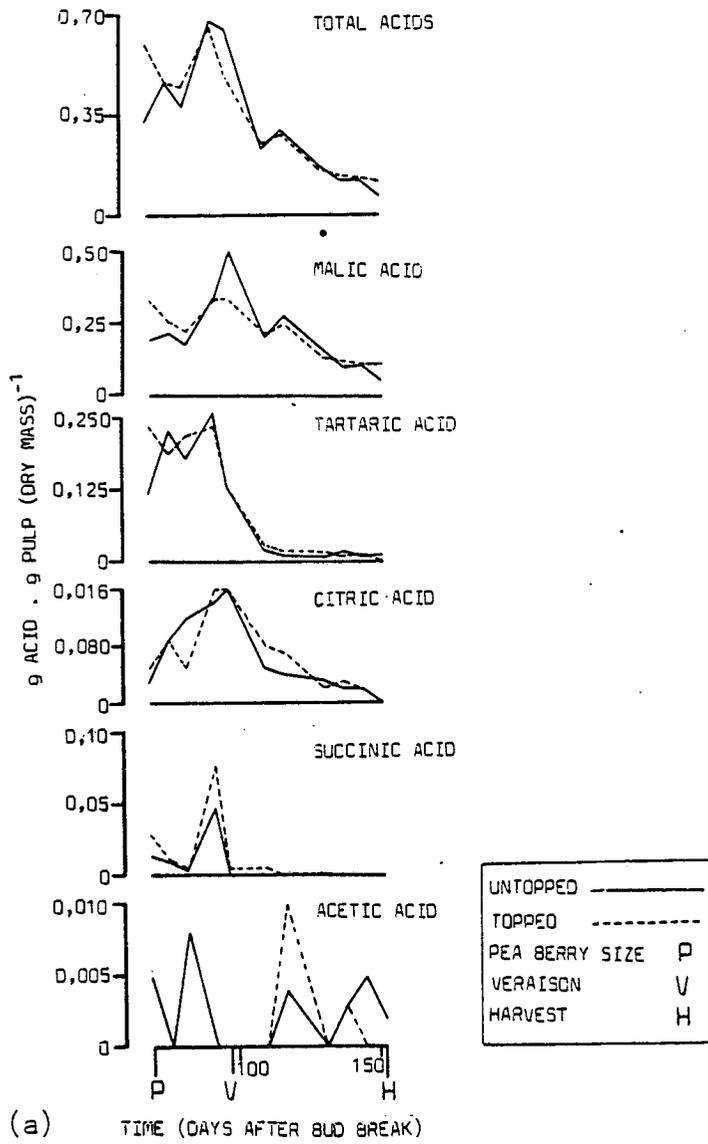


FIG. 7. Seasonal (a) changes and percentage contribution (b) of the organic acids in the pulp of berries from topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines.

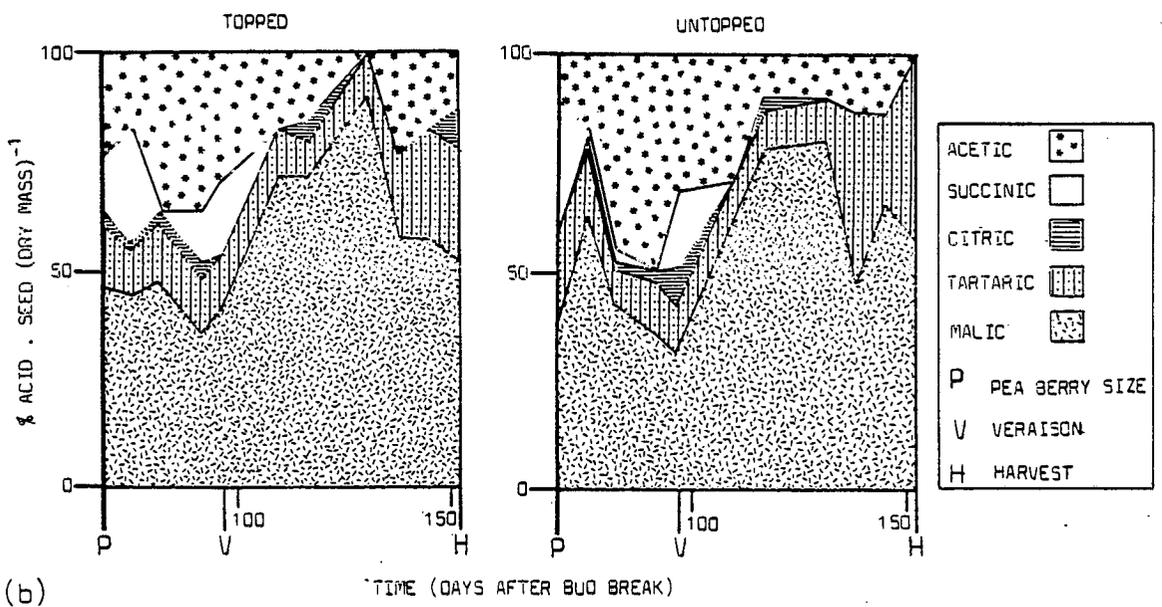
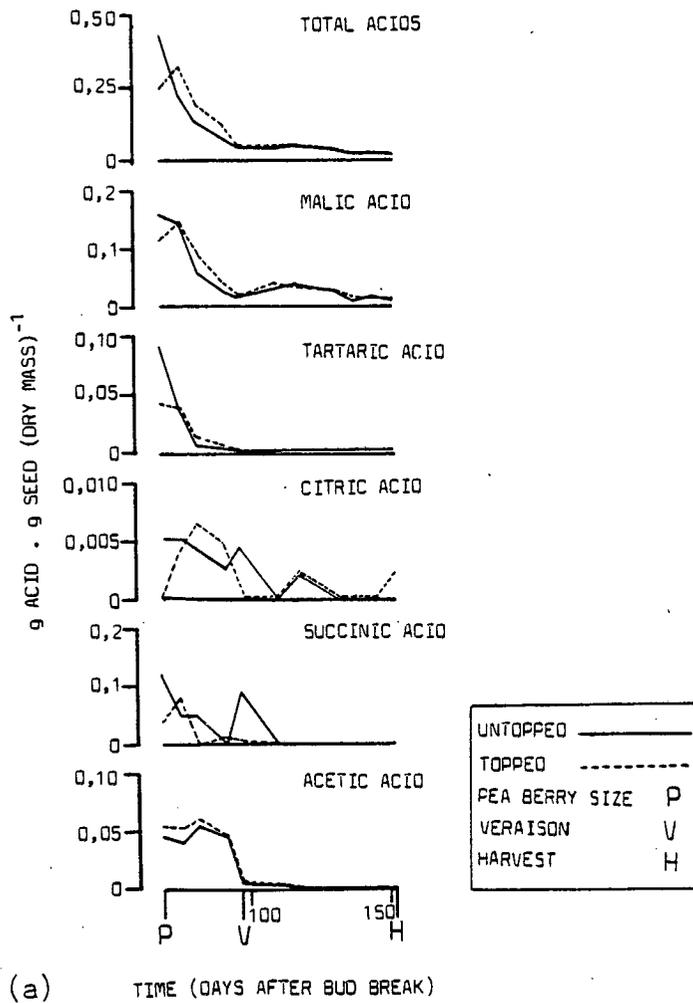


FIG. 8. Seasonal (a) changes and percentage contribution (b) of the organic acids in the seed of berries from topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines.

The same tendency as described above were obtained for tartaric and malic acid in the skin and pulp. Topping had no effect on the development of these acids in the skin.

The total acids in the skin expressed as g.g skin dry mass⁻¹ showed a decrease during ripening while in the pulp an increase occurred before a rapid decrease at veraison (Figs. 6 & 7). This coincides with the ripening of the berry from the skin on the outside to the pulp in the middle (Harris, Kriedemann & Possingham, 1968, Winkler et al., 1974). The total acids in the seed decreased rapidly until veraison and then stayed relatively constant until harvest (Fig. 8). This decrease might be due to the very active growth of the seed at that stage of ripening, where it represents more than 40% of the dry mass of the berry (Chapter 3).

CONCLUSION

It is evident that enlarging the leaf area by topping did not affect the acid composition of the grape. Although more photosynthetate had been translocated (Chapter 4), the excess were either metabolized or translocated to other parts of the vine. It is postulated that the limiting factor for high acid concentrations is not the photosynthetic capacity of the leaf canopy but rather a physical or chemical barrier in the storage capacity of the berry itself. This requires more intensive research because many other practical manipulations can be done to improve the photosynthetic activity of the vine to aquire improved quality of the grape.

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CHAPTER 8

Mineral Composition of the Berry of Vitis vinifera L. cv. Cape Riesling. I. Nitrogen

ABSTRACT

The total nitrogen concentration in the berries of topped and untopped Cape Riesling (Vitis vinifera L.) vines was determined. The total nitrogen increased as berries matured, and the topped vines reached a higher final concentration than the untopped vines. This difference is solely due to an increase in the nitrogen content of the pulp.

INTRODUCTION

Nitrogen is essential for plant metabolism (Bidwell, 1974). The role of nitrogen in viticulture has been extensively investigated and reported on (Cook, 1966; Taylor, 1967; Ough, Lider & Cook, 1968; Christensen, 1969; Kliever & Ough, 1970; Saayman, 1973; Winkler *et al.*, 1974; Conradie, 1980; Bains, Bindra & Bal, 1981; Conradie, 1981; Saayman, 1981).

Total nitrogen (TN) normally increases as the fruit matures. Kliever (1969) reported that concentrations of TN in the juice ranged from 38 to 162 mg N. 100 cm⁻³ juice for slightly to moderately ripe grapes (early harvest) and 43 to 220 mg N. 100 cm⁻³ juice of ripe to overripe grapes (late harvest) for table grape varieties. In the case of wine grapes the concentrations of TN ranged from 44 to 236 mg N. 100 cm⁻³ juice in early harvested grapes and 69 to 256 mg N. 100 cm⁻³ juice in late harvested grapes (Kliever, 1970). The same author also reported higher TN in juices of white cultivars than those of red cultivars and found no correlation between TN, titratable acidity, malate and tartrate. Ough, Lider & Cook (1968) showed significant differences in TN content between cultivars. These differences might be due to differences in leaf area per cane mass, leaf efficiency and pruning practices. Winkler (1958) pointed out that leaf area determines the composition and quality of the grape. Kliever & Ough (1970) reported that TN correlated positively with leaf area per unit mass of grapes. It is therefore clear from the literature that leaf area plays an important role in grape composition and quality.

Because topping resulted in virtually a doubling of leaf area (Chapter 3),

the effect of topping on the nutrient composition of the berry was investigated with special reference to nitrogen.

MATERIAL AND METHODS

Material: *Vitis vinifera* L. cv. Cape Riesling vines were selected as described by De la Harpe & Visser (1983).

Methods: Vines were topped and grapes sampled as described in Chapter 3. The berries were separated into skin, pulp and seed and freeze-dried. The TN was determined in a selenious acid/sulphuric acid digest by means of an automated colorimetric method described by Warner & Jones (1970). The data was statistically analysed by means of an one way analysis of variance done by a SPSS statistical programme (Nie, et al., 1975) on a Burroughs 7800 computer.

RESULTS AND DISCUSSION

Seasonal changes in TN concentration of the berries from topped and untopped vines are shown in Figs. 1 and 2. In both the topped and untopped vines, nitrogen accumulated rapidly and then remained relatively constant for four weeks. As the grapes matured the TN decreased, then increased again to reach a maximum shortly before harvesting and then decreased up to harvesting. The nitrogen concentration in the grapes of topped vines tended to be slightly higher than that of the untopped vines.

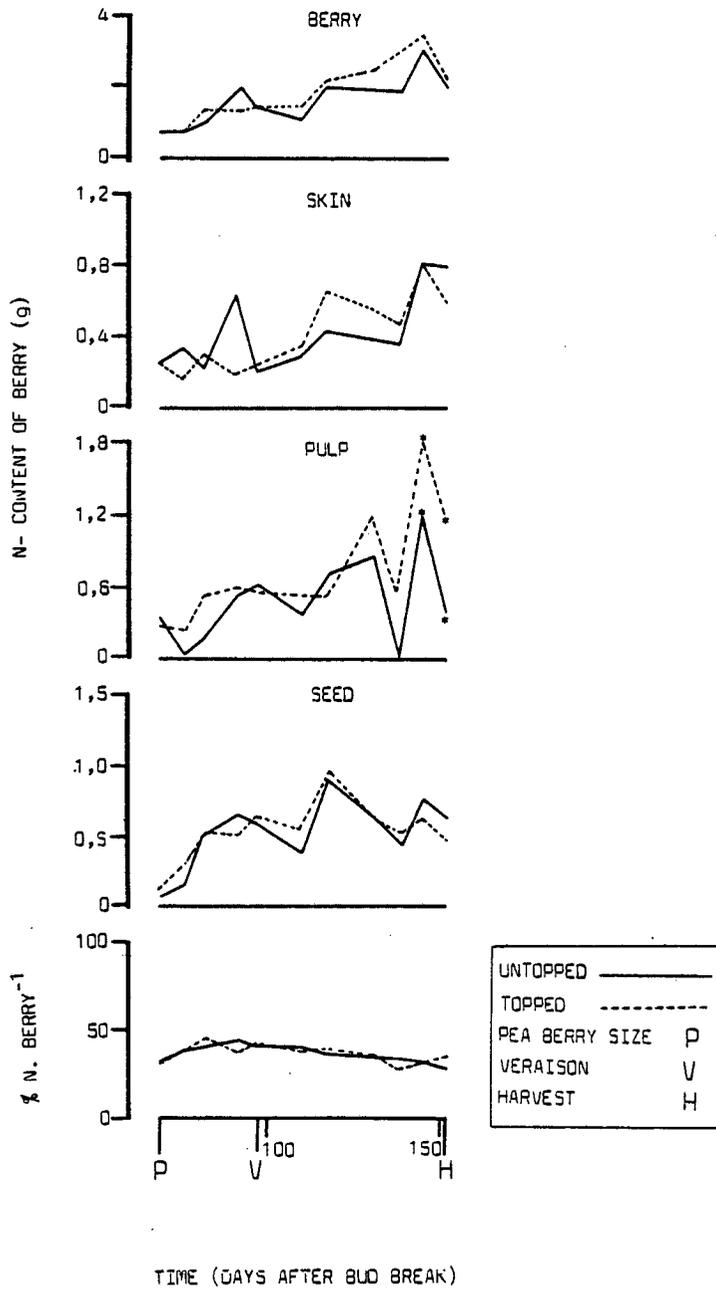


FIG. 1. Seasonal accumulation of nitrogen in the berries, skin, pulp and seed of *Vitis vinifera* L. cv. Cape Riesling vines. (* statistically significant).

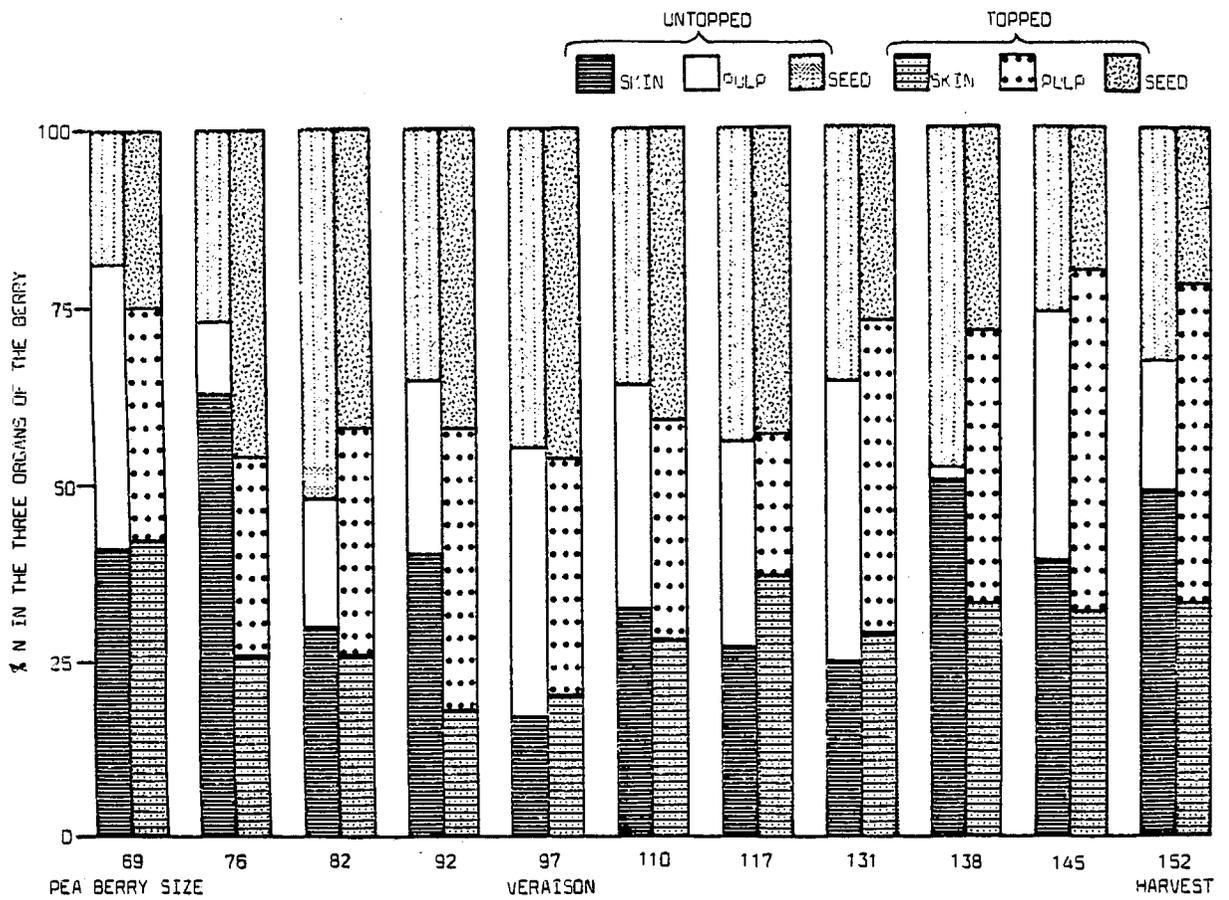


FIG. 2. Changes in the percentage nitrogen in the skin, pulp and seed of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines from pea berry size to harvest.

The TN in the berry exhibited two definite phases of rapidly increasing N-concentration namely, phase one from pea berry size to veraison and phase 2 which started two weeks before harvesting. These phases could possibly be related to growth phases distinguished in the vegetative and reproductive development of the berry (Chapter 3). Topping resulted in a larger increase in TN in comparison to the untopped vines. This phenomenon is not unexpected in view of results reported by Kliewer & Ough (1970) who correlated TN with the leaf area per unit mass of grapes. According to these authors an increase in the total leaf area is accompanied by an increased nitrogen concentration of the grape.

An increase in TN occurred in all three parts of the berry namely skin, pulp and seed as the berries ripened (Fig. 1). In the case of the skin and pulp a general increase was obtained after veraison whilst the TN of the seed increased rapidly around pea berry size and then stayed more or less constant until harvest. More or less 20% of the TN occurred in the pulp of untopped vines. The remaining nitrogen was found in the skin (50%) and seed (30%). This finding is in agreement with the results reported by Ribéreau-Gayon & Peynaud (1960-61). In the case of the topped vines 43% of the TN was found in the pulp, 34% in the skin and 23% in the seed.

Topping had no statistically significant effect on the TN concentration of the skin and seed with topped and untopped vines exhibiting similar tendencies throughout (Figs. 1 & 2). In contrast, the TN concentration of the pulp from the topped vines (Fig. 1) was statistically significantly higher one week before harvest (probability tail = 0,036) and at harvesting (probability tail = 0,040).

The distribution of nitrogen in the different parts of the berry can best be expressed as a percentage of the TN in the berry, as depicted in Fig. 2.

The percentage nitrogen in the skin increased as the berries ripened. The N-concentration of the pulp stayed more or less constant throughout the season whilst that of seed decreased towards harvest after an initial increase.

The pulp of the berries from the topped vines at harvest contained 43% of the total nitrogen, the skin 34%, and the seed 23%. This shows a drastic increase (26%) in nitrogen in the pulp of the topped vines compared to that of the untopped vines which was statistically confirmed. This might be of practical importance because one of the problems encountered in cellars is lagging fermentation due to low must nitrogen concentrations (Cantrelli, 1957; Ough & Kunkee, 1968; Van Rooyen & Tromp, 1982) and better quality at higher nitrogen. However, it must be stressed that the results obtained in this investigation is of a preliminary nature and much more research into this possibility needs to be done before explicit conclusion can be made.

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CHAPTER 9

Mineral Composition of the Berry of Vitis vinifera L. cv. Cape Riesling. II.

Phosphorus, Potassium, Calcium and Magnesium

ABSTRACT

The effect of topping on the seasonal pattern of phosphorus, potassium, calcium and magnesium concentrations in the berry of Vitis vinifera L. cv. Cape Riesling was determined. An increase in the concentrations of all cations were observed as the berries matured.

The pulp of the berries from the topped vines showed higher concentrations of phosphorus (22%), potassium (7%) calcium (8%) and magnesium (6%) than the untopped vines. Phosphorus and potassium were the most abundant in the skin with decreasing concentrations in the pulp followed by the seed. The highest calcium and magnesium concentrations were found in the seed followed by the skin and then the pulp.

INTRODUCTION

Seasonal trends of mineral elements in plants, especially leaves and petioles, have been widely investigated. Excellent reviews have been published by Oberley & Boynton (1966) on apple nutrition, Ballinger, Bell & Childers (1966) on peaches, Cook (1966) and Champagnol (1978) on grape nutrition.

The ranges in percentages by volume of phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg) in fresh juice are 0,02-0,05, 0,15-0,25, 0,004-0,025 and 0,01-0,025 respectively (Winkler, et al., 1974). Conradie (1981) reported an increase in P from veraison to harvest in the berries of Vitis vinifera L. cv. Chenin blanc so that at maturity the P in berries were 34,1% of the total P in the vine. As in the case of P, K increased as the berries matured. At harvest the grapes contained 86,1% of the total K in the vine (Conradie, 1981). This author further reported that the berries only accumulated small amounts of Ca (7,7%) and Mg (15,4%) of the total Ca and Mg contents of the vine in contrast to P and K.

Most of the research done on vine mineral composition was done on the leaf and petiole. In South Africa little is known about the effect of viticultural practices, such as topping, on the mineral composition and distribution in the berry. In Chapter 8 it is shown that topping significantly increased the total nitrogen of the pulp of the berry, which is the main part for wine making.

In this chapter the effect of topping on the mineral composition and distribution of the berry is discussed with special reference to P, K, Ca and Mg.

MATERIAL AND METHODS

Material: *V. vinifera* cv. Cape Riesling vines were selected as described by De la Harpe & Visser (1983).

Methods: Vines were topped and grapes sampled as described in Chapter 3 and the berries treated as described in Chapter 8. The freeze-dried samples (0,5g) were digested by five cm³ HNO₃ and two cm³ HClO₄ at 250°C in an aluminium block oven, then made up to 25 cm³. The K, Ca and Mg concentrations were determined by means of atomic absorption spectrophotometry (Technicon-Varian). Phosphorus was determined by means of the standard VORI automated colorimetric method (Anon, 1981). The statistical analyses were done as described in Chapter 8.

RESULTS AND DISCUSSION

Phosphorus: The P content of the berry and its parts, the skin, pulp and seed are shown in Figs.1 and 2. The P increased as the berries ripened. This is in accordance with the results of Conradie (1981) who reported that the P increased until harvest. The author is of the opinion that this increase might be due to translocation from the leaves since a decrease in P is observed in the leaves.

Although an increase in P content was obtained in the berries of the topped and untopped vines, the P content of the berries from the topped vines tended to be higher than the untopped vines. This might be due to the virtual doubling of leaf area per vine as reported in Chapter 3.

The distribution of P in the different parts of the berry from the topped and untopped vines expressed as a percentage of the total, is depicted in Fig. 2.

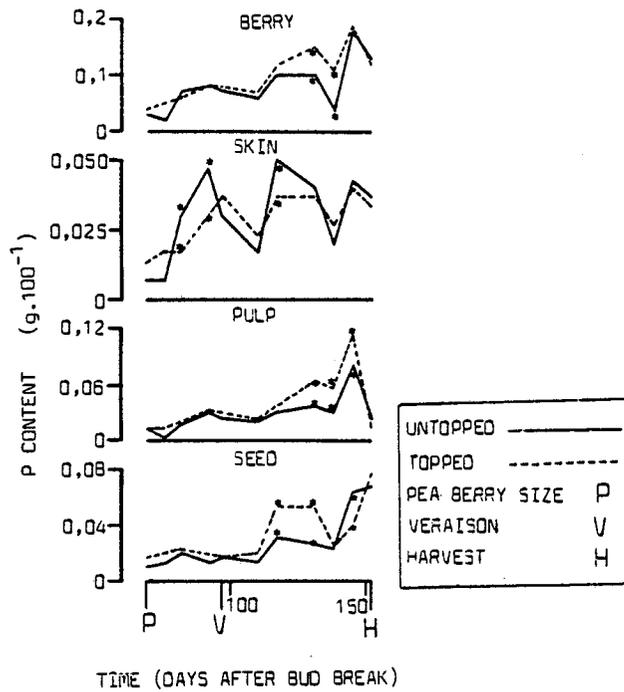


FIG. 1. Seasonal changes of phosphorus in the berries, skin, pulp and seed of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines. (* statistically significant).

It is apparent that topping affected the ratio of P percentages between the different parts. The pulp, which is mainly used in wine making, contributes more P to the must when subjected to a preceding topping treatment from more or less three weeks before harvest.

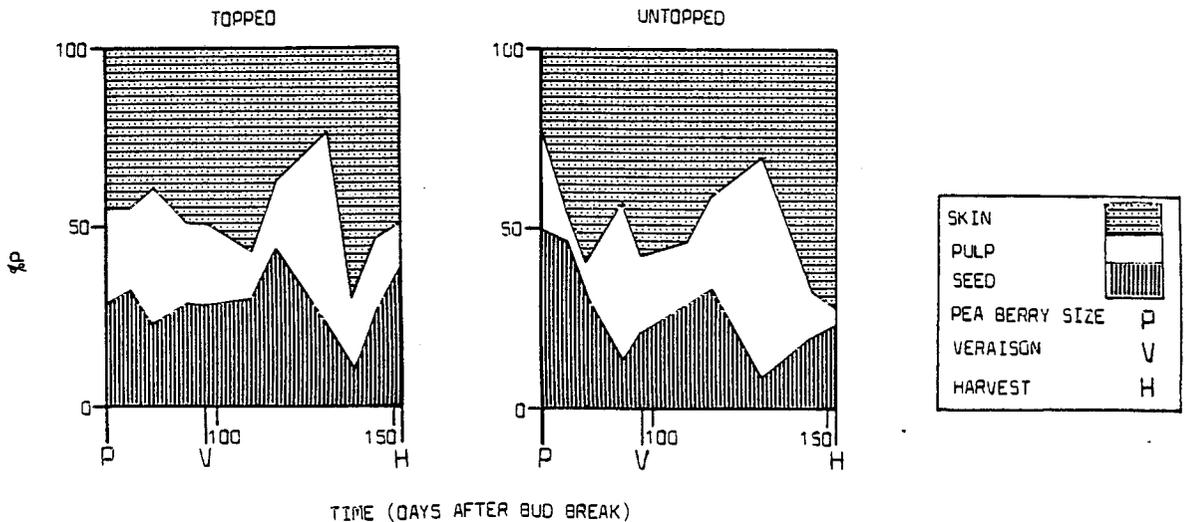


FIG. 2. The distribution of phosphorus in the berries of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines.

Potassium: Potassium was the element with the highest concentration in the berry (Fig. 3). Its concentration increased rapidly after veraison, which is in accordance with the results obtained by Conradie (1981) where a decrease in K in the rest of the vine with a corresponding increase in K concentration of the bunch was reported.

An increase in K in the skin and pulp were obtained as the berry matured (Fig. 3) whilst the K in the seed did not show the same pattern of increase. The percentage distribution of K in the berry for the untopped vines was 66% in the skin, 24% in the pulp and 10% in the seed (Fig. 4). The berries of the treated vines contained 68% (skin), 24% (pulp) and 4% (seed).

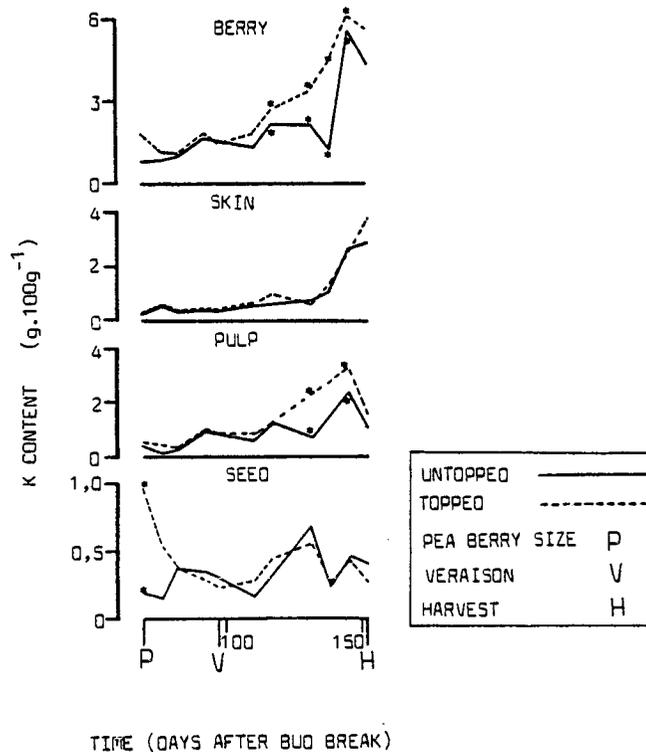


FIG. 3. Seasonal changes of potassium in the berries, skin, pulp and seed of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines. (* statistically significant).

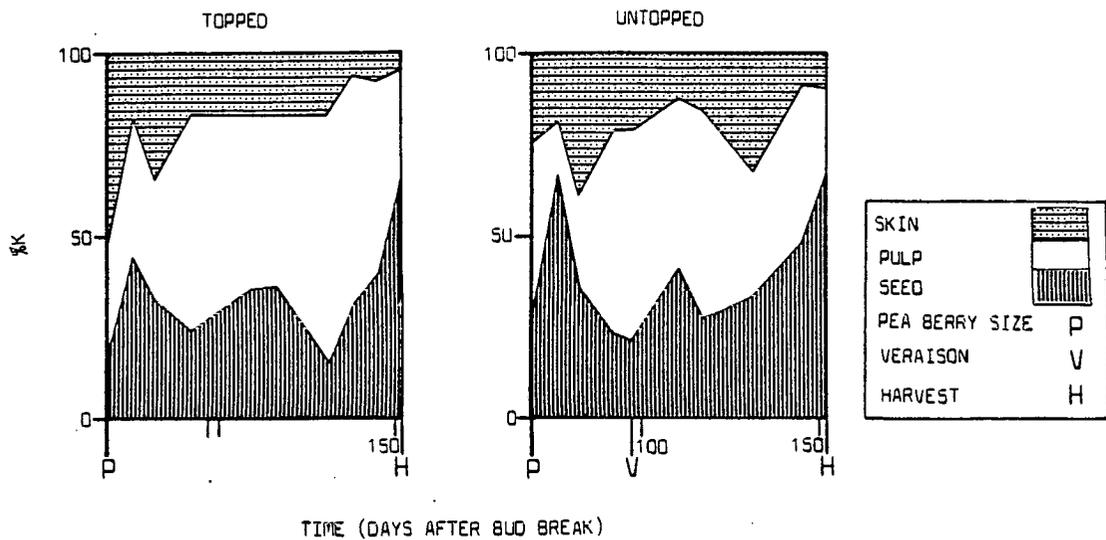


FIG. 4. The distribution of potassium in the berries of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines.

As in the case of N (Chapter 8) and P in the different components of berries it is apparent that topping affected the ratio of K between the different parts. Topping did not, however, affect the K content of the pulp at harvest. This is significant because an increase in pH due to an increase in K could be a negative quality factor.

Calcium: The results for Ca and Mg are depicted in Figs. 5 and 6. The Ca in the berry increased from pea berry size until one week before veraison after which it decreased rapidly. The increase at this stage was mainly due to an increase in Ca in the seed. The second increase in Ca in the berry found more or less two weeks after veraison, was the result of an increase in the skin, pulp and seed. In the case of the pulp the maximum Ca content was reached three weeks before harvest. However, the Ca concentration of the seed increased until harvest following the decrease in Ca at veraison.

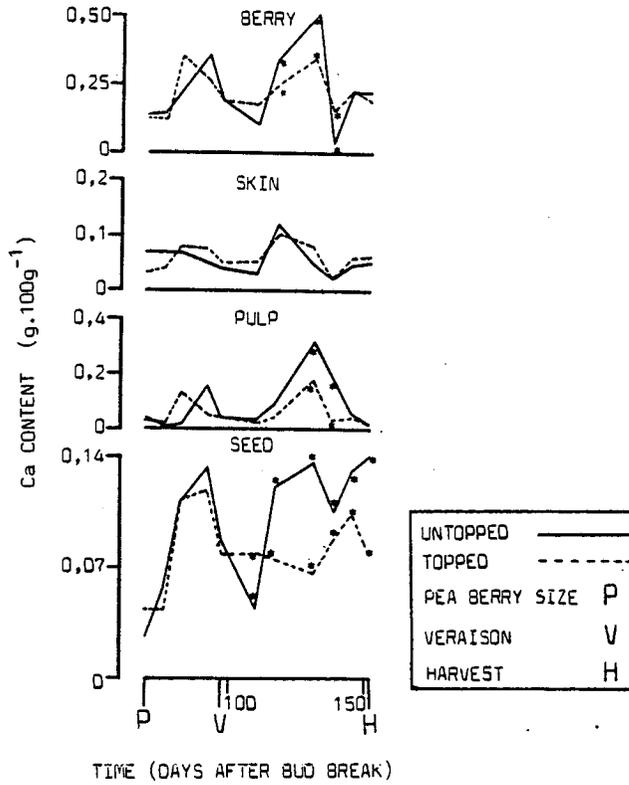


FIG. 5. Seasonal changes of calcium in the berries, skin, pulp and seed of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines. (* statistically significant).

The Ca concentration in the berries of the topped and untopped vines were conspicuously lower than the N, P and K and did not exceed 12% of the total mineral element concentration. Contrary to P and K the seed had the highest Ca concentration whilst the pulp had the lowest (Fig. 6).

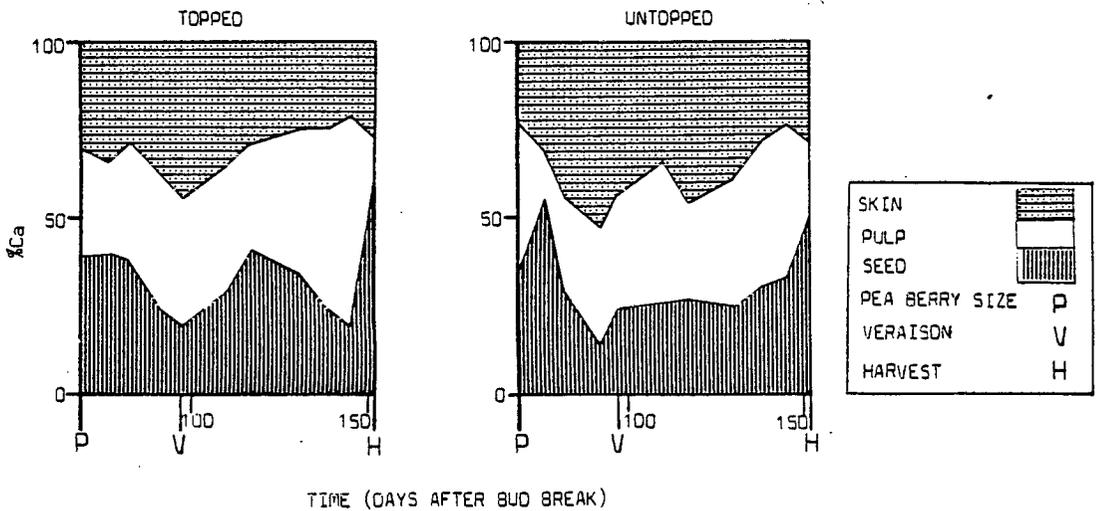


FIG. 6. The distribution of calcium in the berries of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines.

Magnesium: The Mg concentration of the berries from the topped and untopped vines increased as the berries matured (Fig. 7). In the skin a rapid increase

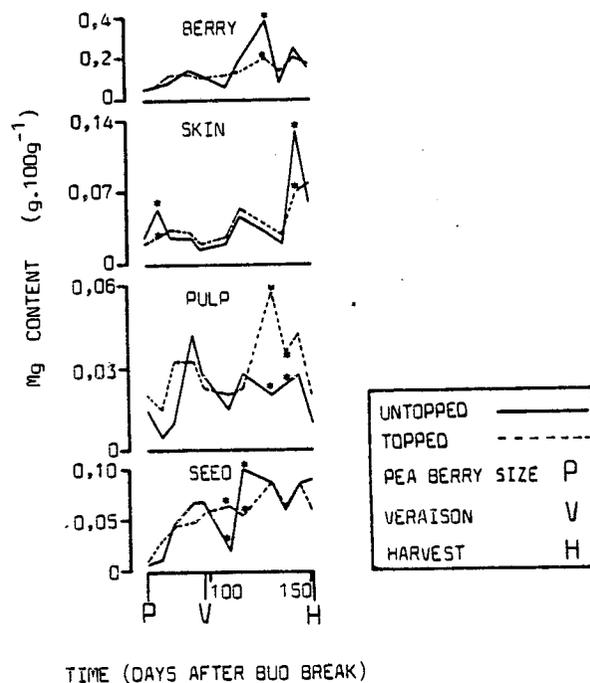


FIG. 7. Seasonal changes of magnesium in the berries, skin, pulp and seed of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines. (* statistically significant).

of Mg was obtained two weeks before harvest (Figs. 7 & 8). The Mg in the pulp of untopped vines increased from pea berry size to one week before veraison after which it decreased till harvest. Contrary to the untopped vines the Mg concentration of the pulp of berries from topped vines increased till one week before veraison, then decreased for three weeks after which it rapidly increased for two weeks before finally decreasing to harvest. Although the decrease in Mg concentration was rather rapid, the Mg content was still significantly higher in the pulp of berries from the topped vines than in that of the untopped vines. As in the case of Ca the highest Mg concentration was in the seed whilst the pulp had the lowest concentration (Fig. 8).

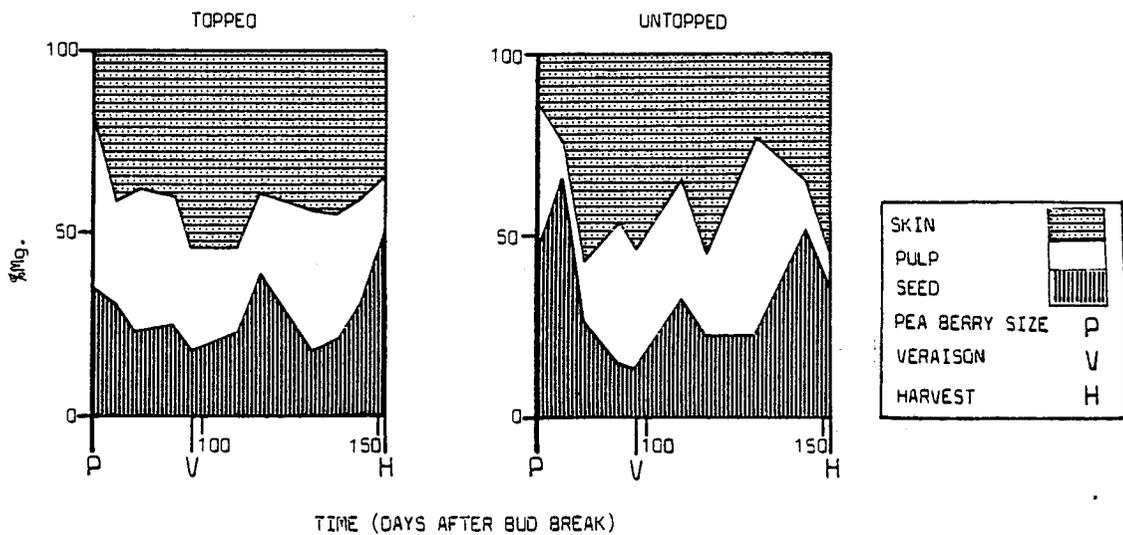


FIG. 8. The distribution of magnesium in the berries of topped and untopped *Vitis vinifera* L. cv. Cape Riesling vines.

Total mineral elements: The total mineral contents (N,P,K, Ca, Mg) expressed as a percentage dry mass per berry tissue was 12% higher in the pulp of berries from topped vines than untopped vines at harvest. The increase in mineral content in the pulp, which is mainly due to N, might be of importance in wine making, since the pulp contains the juice which is the must from which the wine is made. It is known that a higher N concentration improves wine quality (Van Rooyen & Tromp, 1982), whilst a high K concentration negatively effects wine quality (Somers, 1977). As far as Ca and Mg are concerned little is known about their effect on wine quality. Topping resulted in a higher N concentration in pulp (Chapter 8) and did not significantly effect the K concentration in the pulp indicating that topping might have a positive effect on wine quality.

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CHAPTER 10

SUMMARY AND CONCLUSIONS

SUMMARY AND CONCLUSIONS

The effect of topping on the vegetative and reproductive growth as well as the chemical composition and concentration of sugars, organic acids and mineral elements in berries of a Vitis vinifera L. cv. Cape Riesling was investigated. It was found that topping doubled shoot length and leaf area, increased the influx of photosynthetate to the bunch and increased the N content of the berry. The sugars and acids in the berry itself, however, did not show any significant ($p = 0,05$) differences.

Vegetative and reproductive growth

Shoot and leaf growth of both the topped and untopped vines, respectively, can be described as sigmoid. Total shoot length (cm) and total leaf area (cm^2) of topped vines were significantly more than that of untopped vines ($P = 0,05$), attributable to lateral shoot growth in the case of the topped vines. Topping had no effect on bunch development. The skin, pulp and seed development with time of both the topped and untopped vines expressed as a percentage of total berry dry mass can be described by a hyperbolic function for the skin whilst the development of the pulp is linear and that of the seed parabolic. The leaf area correlated positively with the different sugars and mineral elements but were negatively correlated with the acids (Appendix A). A very low correlation coefficient ($r = 0,05$) was obtained between the leaf area and dry mass of the berry. The berry volume was highly correlated with glucose, fructose and total sugars indicating that not only did the sugar concentration increase but an increase in berry volume also took place. Contrary to sugar, acid were highly negatively correlated ($r = -0,90$) with berry volume which meant that as the berry volume increased a decrease in concentration of acids was evident (Appendix A). The dry mass of the berry was positively

correlated ($r = 0,85$) with the berry volume as well as with fructose ($r = 0,91$), glucose ($r = 0,87$) and total sugars ($r = 0,89$). With increased dry mass per berry a decrease in all acids measured was obtained (Appendix A).

Translocation of photosynthetate

Since topping resulted in a doubling of leaf area as well as the creation of new sinks such as growing lateral shoot tips, the effect of topping was determined on the translocation and distribution of photosynthetate in the developing berry (Chapter 4). Multidirectional translocation of organic compounds took place. Bunches, shoots and leaves were sinks at veraison whilst leaves were the only sink at harvest. Following exposure to $^{14}\text{CO}_2$, bunches of topped vines had higher radio-active levels than bunches of untopped vines at veraison and harvest.

The shoot exhibited the highest radio-activity in the neutral, anionic and cationic fractions at veraison indicating that a wide range of organic compounds are being translocated from the source (leaves where $^{14}\text{CO}_2$ was applied), to the rest of the vine. At harvest, however, the main activity remained in the leaves with no activity in the shoots and bunches indicating that very little export took place.

Sugars

Since topped vines transported more ^{14}C -labelled compounds to the bunch than untopped vines, which might be caused by the enlarged leaf area, the effect of topping on the accumulation of sugar in the berry was investigated. Glucose and fructose were found to be the major sugars in the berries. A glucose-fructose ratio of $1\pm 0,1$ was obtained from veraison to harvest. Absence of sucrose was a conspicuous feature of this investigation. Topping had no effect on the

total sugar concentration in the berry expressed on a dry mass basis. The total sugar concentration in the berries of topped and untopped vines increased initially and was followed by a decrease in fructose 76 days after bud break. It started increasing again approximately 97 days after bud break and continued as the berries matured. High correlations were found between glucose, fructose and the total sugars (Appendix A). A negative correlation ($r = -0,96$) was obtained between the sugar and acid concentrations. This meant that as the sugar increased from veraison to harvest the acids decreased.

Organic acids

Topping had no effect on the total acid concentration in the berry expressed on a dry mass basis which steadily decreased in both the topped and untopped vines as ripening progressed. Tartaric and malic acid were found to be the most abundant acids followed by citric, succinic and acetic acid. Topping had no effect on the total acid concentration in the berry expressed on a dry basis. Major acids in the skin at harvest (21°B) were citric, tartaric and malic acid while citric, tartaric, malic, succinic and acetic acid were present in the pulp. Except for citric acid the same acids as in the pulp were also found in the seeds. Topping had no measurable effect on either the developmental pattern or acid composition of the grapes during ripening in the same season in which it was performed. All the acids measured were negatively correlated with the parameters determined in this investigation, indicating that as the berries matured a decrease in acids took place while the other parameters increased (Table 1).

Mineral elements

The total nitrogen increased as berries matured, and the topped vines reached

a higher final concentration than the untopped vines. This difference is solely due to an increase in the N content of the pulp. An increase in P, K, Ca and Mg were also obtained as the berries matured. The pulp of the berries from the topped vines showed higher concentrations of P, K and Mg than the untopped vines. Phosphorus and K were the most abundant in the skin with decreasing concentrations in the pulp followed by the seeds. The highest Ca and Mg concentrations were found in the seed followed by the skin and then the pulp. Nitrogen increased as sugar accumulated and the acids decreased. Nitrogen was found to be highly correlated with P ($r = 0,96$) and K ($r = 0,86$) (Appendix A).

The fact that leaf area and shoot growth were increased by topping led to an increased influx of labelled photosynthetate to the bunch. Although this increase was found in the bunch, the berry, however, did not show a corresponding increase. Only N was found to increase due to topping.

From this investigation it would seem that judicious topping could have beneficial effects on berry composition and could be recommended in vigorously growing vineyards under local conditions. It is quite conceivable that the severity and timing of topping will greatly affect bunch and berry composition as well as various aspects of vegetative growth.

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APPENDIX A

Table 1. The correlation matrix for 17 parameters determine on the berries and vines of *Vitis vinifera* L. cv. Cape Riesling

	F	G	TS	C	T	M	S	A	TA	N	P	Mg	K	Ca	L	B	BD
F	1,00	0,96	0,99	-0,04	-0,80	-0,91	-0,69	-0,79	-0,96	0,65	0,68	0,58	0,76	0,05	0,65	0,94	0,91
G	0,96	1,00	0,99	-0,86	-0,74	-0,90	-0,53	-0,72	-0,94	0,64	0,66	0,68	0,71	0,19	0,63	0,93	0,86
TS	0,99	0,99	1,00	-0,86	-0,77	-0,91	-0,57	-0,76	0,96	0,65	0,68	0,64	0,74	0,12	0,65	0,94	0,89
C	-0,84	-0,86	-0,86	1,00	0,78	0,69	0,51	0,44	0,84	-0,50	-0,53	-0,40	-0,68	-0,01	-0,42	-0,75	-0,78
T	-0,80	-0,74	-0,77	0,78	1,00	0,57	0,54	0,53	0,79	-0,41	-0,42	-0,33	-0,61	-0,12	-0,40	-0,72	-0,80
M	-0,91	-0,90	-0,91	0,69	0,57	1,00	0,61	0,86	0,93	-0,65	-0,67	-0,59	-0,65	-0,09	-0,73	-0,87	-0,79
S	-0,60	-0,53	-0,57	0,51	0,54	0,61	1,00	0,62	0,70	-0,38	-0,36	-0,33	-0,35	-0,06	-0,43	-0,56	-0,51
A	-0,79	-0,72	-0,76	0,44	0,44	0,86	0,62	1,00	0,79	-0,59	-0,58	-0,56	-0,55	-0,13	-0,70	-0,70	-0,68
TA	-0,96	-0,94	-0,96	0,84	0,79	0,93	0,70	0,79	1,00	-0,65	-0,67	-0,55	-0,73	-0,03	-0,69	-0,90	-0,89
N	0,65	0,64	0,65	-0,50	-0,41	-0,65	-0,30	-0,59	-0,65	1,00	0,96	0,67	0,86	0,30	0,67	0,71	0,64
P	0,68	0,66	0,68	-0,53	-0,42	-0,67	-0,36	-0,50	-0,67	0,96	1,00	0,65	0,89	0,30	0,66	0,73	0,69
Mg	0,58	0,68	0,64	-0,40	-0,33	-0,59	-0,33	-0,56	-0,55	0,67	0,65	1,00	0,56	0,73	0,39	0,69	0,43
K	0,76	0,71	0,74	-0,60	-0,61	-0,65	-0,35	-0,55	-0,73	0,86	0,89	0,50	1,00	0,03	0,68	0,73	0,83
Ca	0,05	0,19	0,12	-0,01	0,12	-0,09	-0,06	-0,13	-0,03	0,30	0,30	0,73	0,03	1,00	0,05	0,22	-0,05
L	0,65	0,63	0,65	-0,42	-0,40	-0,73	-0,43	-0,70	-0,60	0,67	0,66	0,39	0,68	0,05	1,00	0,70	0,72
B	0,94	0,93	0,94	-0,75	-0,72	-0,87	-0,56	-0,78	-0,90	0,71	0,73	0,69	0,73	0,22	0,70	1,00	0,85
BD	0,91	0,86	0,89	-0,78	-0,80	-0,79	-0,51	-0,60	-0,89	0,64	0,69	0,43	0,83	-0,05	0,72	0,85	1,00

F = fructose; G = glucose; TS = total sugars; C = citric acid; T = tartaric acid; M = malic acid; S = succinic acid;

A = acetic acid; TA = total acids; N = nitrogen; P = phosphorus; Mg = magnesium; K = potassium; Ca = calcium; L = leaf

area; B = berry volume; BD = dry mass. berry⁻¹