GRAPE AND WINE RESEARCH COUNCIL

PROJECT NUMBER UA4GW

PROJECT TITLE: STUDIES ON FRUIT-SET IN WINEGRAPES

PROJECT SUPERVISOR: P. MAY

FINAL REPORT

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SUMMARY

Comparisons over three seasons of Chardonnay vines in vineyards of the Adelaide Hills (H, at about 500 m above sea level) and the Southern Vales (P, at about sea level) showed that differences in per cent set between vines pruned to spur, cane or by simulated mechanical hedging were small on both sites. Site H had temperatures below 16° for two weeks during the flowering period in spring 1988 and per cent set was only about half that of site P. In the next two seasons, percentage set was similar on both sites. Yield per vine at P was six times that of H in season 1 and about three times bigger in the other two seasons when the differences were due mainly to causes unrelated to berry set. At H, berries had fewer seeds than on P in all seasons.

Cap fall per day was closely related to temperature, and occurred in the main at daily maxima above 20° Pollen and ovules appeared normal in the second season, while in the third season some ovules lacked embryo sacs. Pollen germination *in vitro* was erratic even at higher temperatures; only pollen from P produced pollen tubes. Pollen germination on the stigma and pollen tube growth within the pistil was better at P than at H. In particular, some pollen tubes branched within the H- pistils, indicating some growth inhibition.

Studies of the distribution of flowers and berries within the inflorescence and the bunch are not yet complete. Observations of early bunch stem necrosis showed large seasonal variation. Applications to inflorescences of boron and proline in 1989/90 and 1990/91 were without effect.

It is concluded that in cultivars such as Chardonnay, whose sexual apparatus is fully functional under favourable weather conditions, the development and/or functioning of these organs become abnormal under unfavourable weather conditions, leading to the abscission of flowers before ovary expansion has commenced.

INTRODUCTION

Poor berry set is one of the major hazards of grape growing, particularly in "cool" regions. Much information is available for cultivars in which this condition is due to genetic defects of the sexual apparatus. However, we know little about cultivars which are not deficient in this respect but which set poorly in some seasons. This study was designed to increase our knowledge on these aspects of the problem and, if possible, to suggest ways to reduce the likelihood of crop losses caused by it. Chardonnay vines were used for (a) testing the effects of vine pruning, vine vigour and environment on berry set and for (b) studying the development of flowers as influenced by their position on the inflorescence and by weather conditions.

In a main experiment, the effect of three methods of pruning on berry set was tested in two vineyards varying in climate and other vineyard characteristics. The course of cap fall was observed on individual inflorescences to examine the relationship between weather and flower opening. The development of the floral organs and the process of pollination were examined. The possible effects on berry set of applications of boron and of the amino acid proline were tested in spot trials. Some observations were also made on the occurrence of early bunch stem necrosis.

The study extended over the seasons 1988/89 to 1990/91. Observations were concluded by July 1991, but evaluation of the results has continued since and is not yet complete. This report is therefore not to be regarded as "final"; publication of some results should be possible in the near future. A summary of the general problem of berry set was prepared (see Appendix).

MATERIAL AND METHODS

The main experiment

Chardonnay vines (clone I10 V1), less than 10 years old, own-rooted and trickle-irrigated,.were used in two vineyards, one in the Adelaide Hills at 500 m altitude (Hills, H), the other several kilometres inland at about sea level near Willunga, Southern Vales (Plain, P). The vines of vineyard P are vigorous and produce a dense foliage canopy, particularly with "mechanical" pruning. Their trellis consists of two superimposed wires, with the upper wire carrying part of the cordon. The management of the canopy is restricted to some shoot topping. The H-vines, pruned to fewer buds, have an upright, much thinner canopy held in position between two pairs of foliage wires; the shoots are topped at about 2 m height. Except for the pruning treatments, the experimental vines were maintained as the rest of the vineyard.

Each experiment consisted of 12 randomized blocks, each with three plots consisting of a single vine pruned to spur, cane or by simulated

mechanical "hedging". In the first season, all vines were pruned in mid-winter. In the next season, six blocks were pruned in mid-winter, three at the first sign of bud burst and three when the terminal shoots of the unpruned canes had about three young leaves. In the next season, six blocks each of the P-vineyard only were pruned at the earliest and latest stage. The H- and P-vines were given 18 or 28 spurs and 3 or 4 canes respectively. Bud numbers on hedging were not controlled and varied considerably from season to season; To determine berry set, four inflorescences per vine in 1988 and two per vine in 1989 and 1990 were enclosed in paper bags just before anthesis. They were harvested before veraison and frozen for later counting. These counts were done separately for each branch of the bunch rachis by grouping the berries and non-developing ovaries visually into seven size classes. The harvested crop consists of the berries in classes 1-4; therefore, per cent set is calculated as the percentage of the numbers in these classes per total number of flowers. The remaining three classes contain abscissed flowers or "pinheads". Many of them remain in their position on the bunch while others become detached.

Weather data

The best available weather records for the Hills vineyard come from the Lenswood Research Centre of the South Australian Department of Agriculture and for the Plain vineyard from interpolation of the Commonwealth Bureau of Metereology data collected at Adelaide Airport and Victor Harbour. One month' values from a data logger installed at the Hills vineyard showed Lenswood to be about 1° warmer than the Hills site.

The course of cap fall

On a site of H some distance from that used for the main experiment, twelve Chardonnay inflorescences were enclosed in muslin bags in each of the three seasons and the detached caps were counted daily or less often towards the end of the flowering period. Measurements of shoot elongation, berry set and distribution of berries were made also, including counts of seeds.

Development of ovules and pollen grains

The third branches of Chardonnay inflorescences of the H- and P-experiment were collected at two- to three-day intervals after late October. Whole ovaries in spring 1989 and ovules only in 1990 were prepared for microscopic examination by standard techniques, i.e. fixation, embedding in glycol methacrylate, sectioning, and staining with periodic acid-Schiff reagent and Toluidine Blue.

Pollination

The presence or absence of pollen grains on the stigma and of pollen tubes in the pistil was determined on flowers of the third inflorescence-

branches (see above) by observing their fluorescence under UV illumination. The whole ovaries, fixed in Carnoy's fluid, were hydrated, softened in 0.8N NaOH, stained with aniline blue, bisected longitudinally and examined in 80% glycerol. In spring 1989, the specimens were scored according to the number of pollen grains or -tubes, using a scale from 1-5 arranged in ascending order while actual counts were made in 1990.

Pollen germination

Pollen was collected from newly opened anthers and placed into hanging drops containing 10% sucrose, 0.01% H_3BO_3, 0.03% $Ca(NO_3).4H_2O$, 0.02% $MgSO_4.7H_2O$ and 0.01% KNO_3. Two microscope slides with two hanging drops each were placed onto small plastic blocks in petri dishes. They were the four replications of one of the treatments, the combinations of one of five temperatures (5°, 10°, 15°, 20°, 25° C) and one of the periods of incubation (varying from 2h to 72h). Several samples of 1990 pollen were incubated with fluorescein diacetate added to the bathing solution to test their viability by fluorescence.

Application of Boron

In spring 1989, inflorescences of the Hills vineyard were dipped just before the commencement of flowering into solutions of boron (Spray boron at 0.5% and 0.1% concentration) or water with Agral being added as wetting agent. When dry, they were bagged for subsequent counts of berries and ovaries.

Application of Proline

Solutions of proline (concentrations 0.1M, 0.5M, 1M) or water as control, in each case with Agral as wetter, were applied to Chardonnay inflorescences of the Hills vineyard just before anthesis, in 1989 by a single spray and in 1989 by a dip repeated three times at two-day intervals. The inflorescences were then bagged for later counts of berries and ovaries.

RESULTS AND DISCUSSION

Weather conditions before and during flowering

Average 5-day temperature maxima and minima during the period prior to and during flowering for the Hills and the Plain region are shown in Fig. 1.

Flowering was estimated to commence in the H-vineyard of the main experiment in the three seasons on about November 6, 11 and 16 and to end on about December 6 in 1988 and one week earlier in the other seasons. At P, flowering commenced and ended at about the same time in 1988 and 1990 and lasted from about October 25 to about November 15. In 1989, flowering started later on about November 6 but ended by about

November 20. The first appearance of free anthers can be determined accurately without guessing, but subjective estimates of the percentage of open flowers and even of the end of the flowering period must be regarded with suspicion because flowering time varies considerably between bunches and guessing the proportions of still closed flowers among the confusing mass of anthers displayed by even small numbers of open flowers is difficult.

The temperature data shown for spring 1988 do not fully express the unfavourable conditions in the Hills during that season: a period of high temperatures at the beginning of November induced early commencement of flowering; it was followed by a period of 14 days with maxima of 16° or less interrupted by only two days of 20° and 24°.

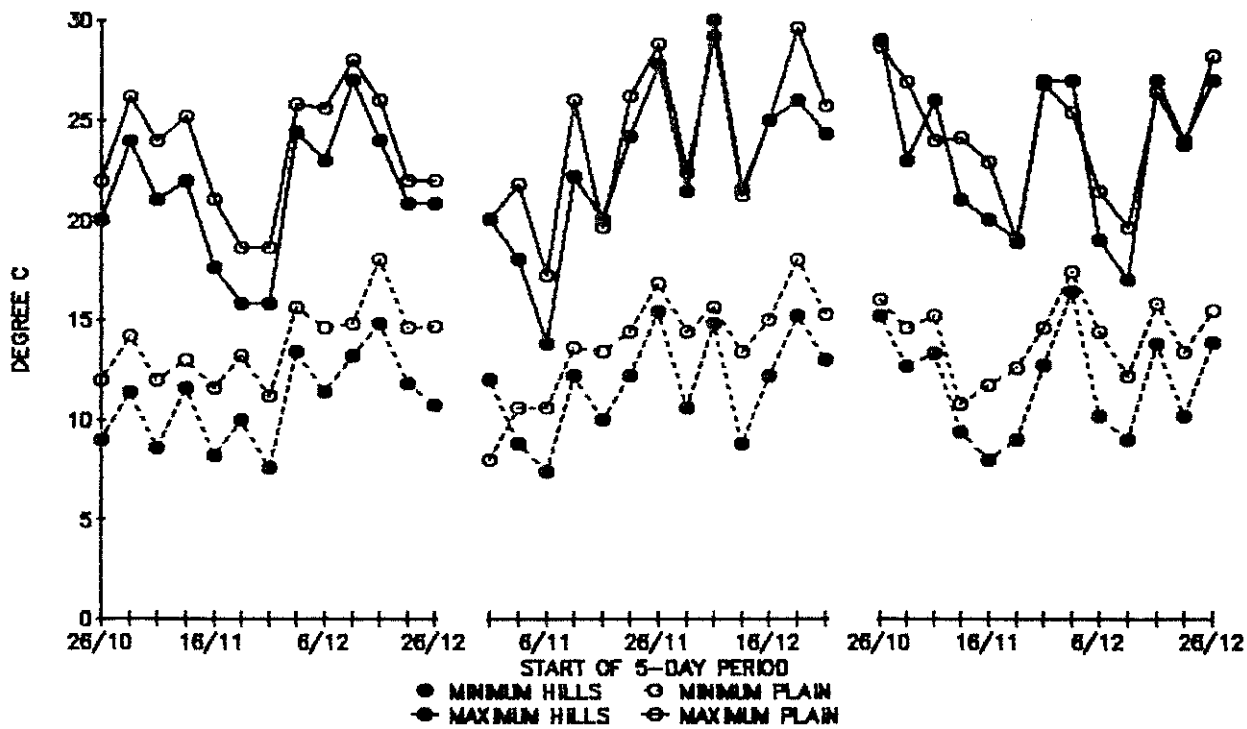


Figure 1: Maximum and minimum temperatures (averages for 5-day periods) for October 26 to December 31, seasons 1988/89 (left), 1989/90 (centre) and 1990/91 (right), Hills and Plain region.

Effect of pruning treatments on fruit set

Berry numbers and per cent set for the three pruning treatments spur, cane and simulated mechanical hedging at the two sites are shown in Table 1. Overall, the inflorescences of cane-pruned vines tended to have more flowers than those of the other two pruning treatments, particularly in the P-vineyard.

TABLE 1: Berry number / bunch and per cent berries / total flowers of bunches bagged at flowering, from vines of three pruning treatments in the Hills (H) and Plain (P) experiments in three seasons.

	1988/89		1989/90		1990/91	
BERRY NUMBER	H	P	H	P	H	P
Spur	39	106	49	99	54	65
Cane	48	123	74	116	56	114
Mechanical	46	107	64	114	46	94
Significance	NS	NS	*	***	NS	***
PER CENT BERRY SET						
Spur	20	48	27	39	35	37
Cane	18	36	31	34	26	30
Mechanical	21	43	30	36	37	40
Significance	NS	***	NS	NS	*	**

NS = not significant; *,**,*** significant at p > 5%,1%,0.1%

This was due to the inclusion by random selection of bunches on shoots more distal than those on short or even long spurs. Distal bunches are bigger as fruitfulness increases along the cane. Reduced set follows as the result of the general trend for larger inflorescences to have lower per cent set. The treatment differences were much smaller in the H-vines whose inflorescences had fewer flowers. Overall, these differences did not affect the response to seasonal conditions. They were of little consequence considering the very large overall differences between 1988/89 and the later seasons. The density of the foliage canopy of the hedged vines which resulted in increased shading with earlier and more intense yellowing of basal leaves, did not lead to reduced berry set. Time of pruning did not affect per cent set and data are not presented.

Table 2: Harvest data for vines of three pruning treatments in the Hills (H) and Plain (P) experiment in three seasons.

YIELD/VINE (kg)	1988/89		1989/90		1990/91	
	H	P	H	P	H	P
Spur	1.50	11.77	4.23	13.10	2.99	9.87
Cane	1.57	9.75	5.13	14.85	3.38	13.88
Mechanical	2.73	15.34	6.71	15.71	4.52	14.38
LSD 1%	0.52	2.58	1.26	3.10	0.97	3.32
BUNCHES/VINE						
Spur	49	97	49	92	49	78
Cane	55	74	58	106	56	92
Mechanical	94	147	88	164	79	145
LSD 1%	12	17	13	13	15	24
WEIGHT/BUNCH (g)						
Spur	30	122	87	143	59	128
Cane	29	132	92	139	63	151
Mechanical	29	104	76	96	58	98
LSD 1%	NS	16	3	13	9	12

LSD = Least significant difference between means; NS = not significant

The effects of the pruning treatments on yield, bunch number and calculated bunch weight are shown in Table 2. In 1988, the small per cent set of the H-vines led to very low yields. The P-vines, which had almost finished flowering when they experienced a period of low temperatures, which was also less severe than at H, yielded about six times as much. In 1989/90 and 1990/91 when per cent set on the two sites was similar, yields differed by factors of 2.7 and 3.5. It may be assumed that in 1988/89 the yield difference per vine between the two sites was in about equal parts due to differences in pruning level, planting distance, vineyard management and general environmental conditions on the one hand and to poor set in the Hills on the other hand. The question then remains whether the poor set was caused by partial failure of flowers to be fertilized or by insufficient nutrition of fertilized flowers leading to the abscission of the slightly expanded ovaries soon after fertilization. The higher yields of hedged vines were due to more bunches and not to altered per cent set.

Seed number per berry

Success or failure of ovule fertilization depends on the pre-anthesis development of the sexual organs and on the process of pollination. Seed number per berry is the combined result of the two phenomena. Table 3 presents mean seed number per berry for classes of berry size arranged in descending order. Each value is the mean of the three pruning treatments whose mean seed number did not differ significantly.

TABLE 3: Seed number per berry in four size classes of berry size from the Hills (H) and Plain (P) experiment in three seasons.

SIZE CLASS	1988/89		1989/90		1990/91	
	H	P	H	P	H	P
1	1.03	2.33	1.97	2.50	1.58	2.27
2	0.93	1.59	1.38	1.97	1.23	1.67
3	0.60	0.97	1.12	1.27	0.98	1.12
4	0.50	0.61	0.91	1.06	0.53	0.81

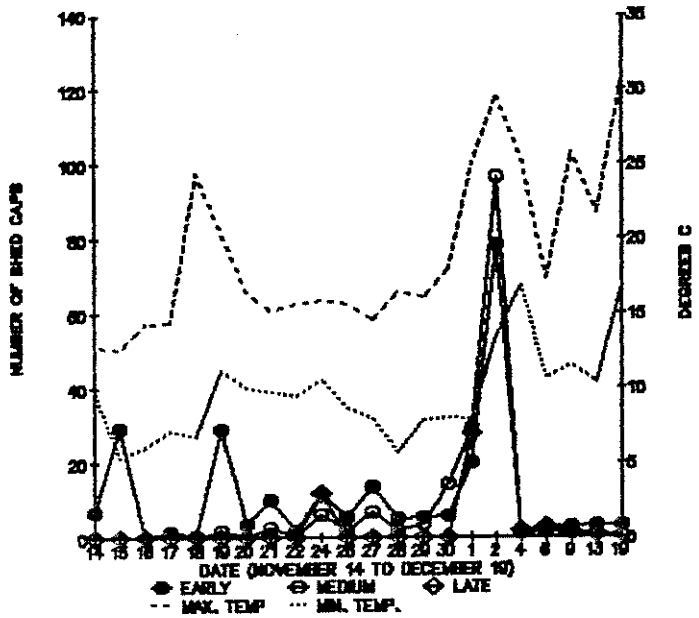
Berries of each size class contained more seeds at P than at H, not only in 1988/89 when set at H was poor but also in the subsequent seasons when per cent set was similar on both sites. On average for the four classes, seed number at H was 62%, 80% and 74% of that at P in the three seasons. This indicates reduced efficiency of the sexual apparatus in the Hills vineyard in all seasons.

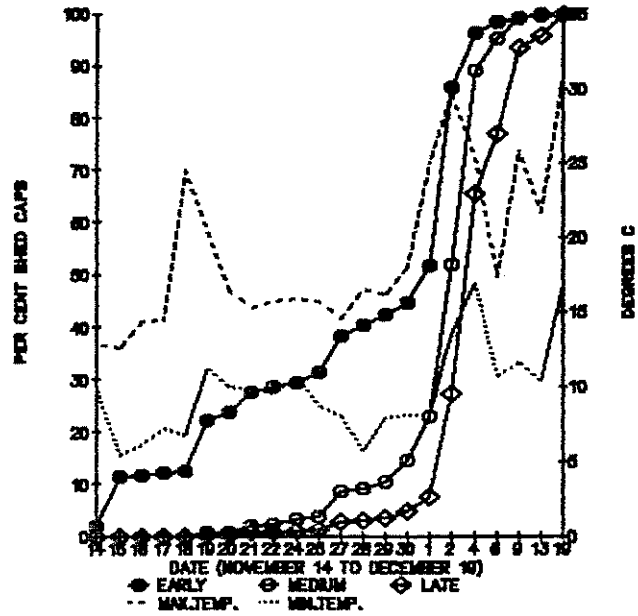
The course of anthesis

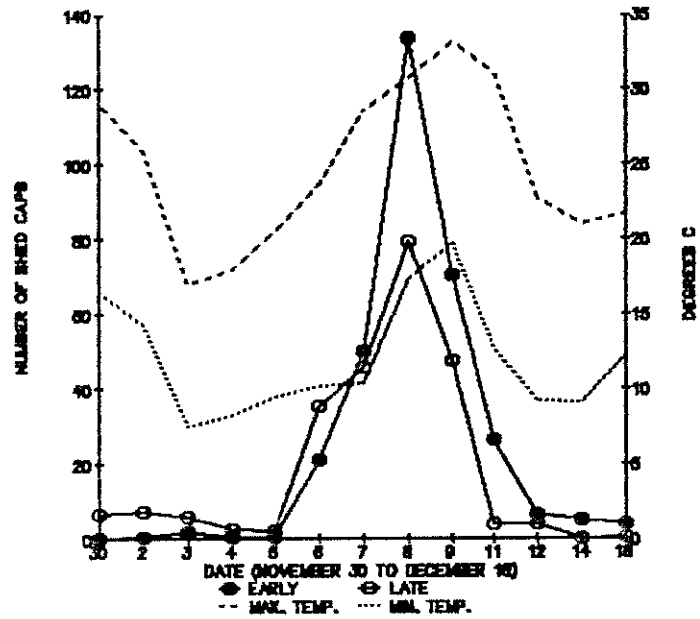
Fig. 2 summarizes the course of the daily shedding of flower caps (the fused petals) from H-inflorescences which were grouped according to the start of their flowering into "Early" / "Medium" / "Late" in 1988 and "Early" / "Late" in the following two seasons. The left-hand graphs show numbers of caps shed per day, either as actual counts or as averages for two or more days. Cumulative percentages of shed flowers are plotted on the right-hand graphs. Each diagram contains also the daily temperature maxima and minima.

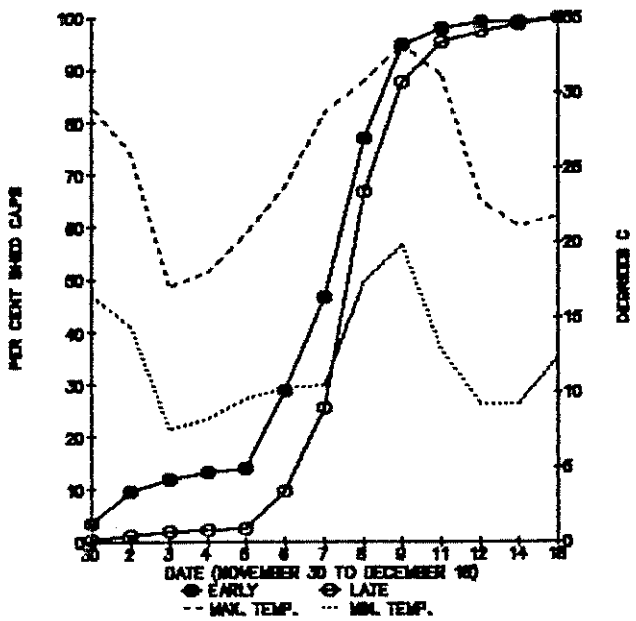
The close relationship between temperature and cap fall is obvious. Few caps fell at maximum temperatures below 20° and none below 15°. The start of flowering on "late" inflorescences must have been delayed by factors unrelated to pre-anthesis weather conditions; once flowering had started, the flowers of the late inflorescences shed their caps rapidly at suitable temperatures and completed flowering at the same time as "early" inflorescences. Evidence from the studies discussed below indicates that pollination does not occur before the cap is shed, even though some anthers dehisce and deposit their pollen grains on the stigma while the cap is still in position. Cap fall therefore indicates when pollination can commence but further studies are needed to determine how pollen deposition and stigma receptivity are correlated and how weather conditions can affect this relationship.

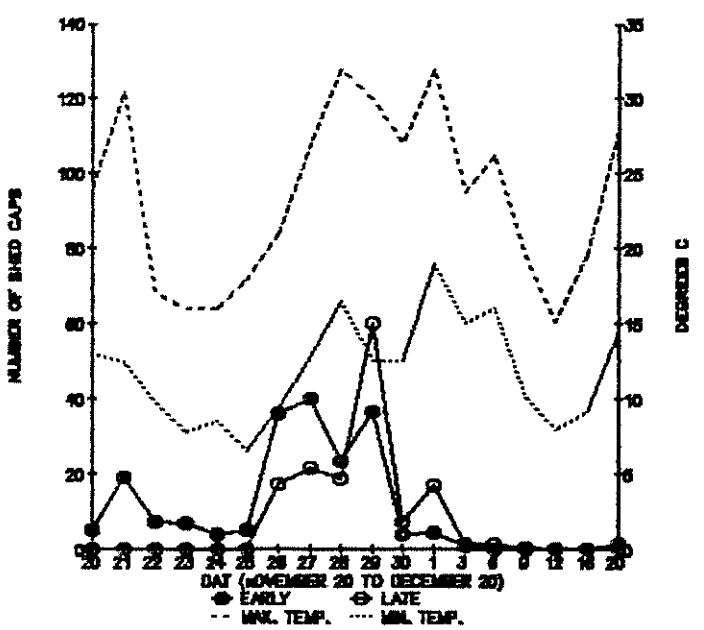
Fig. 2: Mean number of caps (left) and cumulative percentage of caps (right) shed daily on inflorescences with early, medium and late start of anthesis. Daily maximum and minimum temperatures are plotted also. Data from 12 Chardonnay inflorescences of the Hills vineyard in spring 1988 (top) and 1990 (bottom) and 9 inflorescences in 1989 (centre).

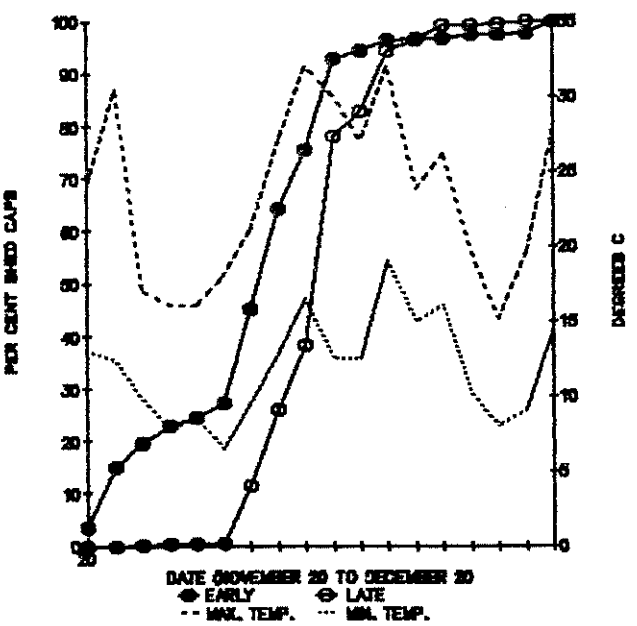

NUMBER OF SHED CAPS
DEGREES C
DATE (NOVEMBER 14 TO DECEMBER 16)
EARLY
MEDIUM
LATE
MAX. TEMP
MIN. TEMP.


PER CENT SHED CAPS
DEGREES C
DATE (NOVEMBER 14 TO DECEMBER 16)
EARLY
MEDIUM
LATE
MAX.TEMP.
MIN.TEMP.


NUMBER OF SHED CAPS
DEGREES C
DATE (NOVEMBER 30 TO DECEMBER 16)
EARLY
LATE
MAX. TEMP.
MIN. TEMP.


PER CENT SHED CAPS
DEGREES C
DATE (NOVEMBER 30 TO DECEMBER 16)
EARLY
LATE
MAX. TEMP.
MIN. TEMP.


NUMBER OF SHED CAPS
DEGREES C
DAT (NOVEMBER 20 TO DECEMBER 20)
EARLY
LATE
MAX. TEMP.
MIN. TEMP.


PER CENT SHED CAPS
DEGREES C
DATE (NOVEMBER 20 TO DECEMBER 20
EARLY
LATE
MAX. TEMP.
MIN. TEMP.

Embryo sac development

Grape ovaries are stimulated into growth by the process of fertilization and the post-fertilization development of their ovule(s). However, the continuing development of the embryo is not essential for set, as proven by the productivity of "seedless" cultivars. The microscopic studies of the developing ovary and ovule were designed to compare ovule and pollen development in Chardonnay samples from the two sites to check whether irregularities in their development may contribute to poor set.

In 1989/90, the developments of ovaries, ovules and anthers were found to be normal on both sites. In these samples, pollen grains could be found in the anthers before the embryo sac became visible within the nucellus of the ovules. Embryo sacs were found in P-samples on October 31 while cap fall commenced between November 12 and 15. On site H, pollen grains were found by November 20, embryo sacs by November 27 and shedding of caps by December 2.

In 1990/91, a number of ovules lacked the embryo sac (Table 4). Overall, the percentage of such ovules was 15.6% for P-vines and 24.7% for H-vines. This is of the same order as found by Kassemeyer and Staudt (1982) for Gewürztraminer and Pinot Blanc where about 30% and 10% respectively of ovules lacked embryo sacs and were thus sterile. The factors of the environment reigning during the developmental phase which are responsible for this type of ovule degeneration are not known. Does the proportion of such ovules ever become sufficiently large to affect per cent set ? Could it be that high temperatures in late October 1990 were responsible for its occurrence?

TABLE 4: Number of ovules with embryo sacs (ES) per number of ovules examined (Ov) from centrally and laterally positioned ovaries of inflorescence branches 3, sampled in the Hills and Plain vineyards, November 1990.

	HILLS			PLAIN	
Date	Central	Lateral	Date	Central	Lateral
17 Nov	3/4	2/3	3 Nov	4/4	4/4
	4/4	2/2		4/4	4/4
19 Nov	3/4	3/4	5 Nov	2/4	2/3
	3/4	1/4		2/2	3/4
21 Nov	2/4	2/4	7 Nov	2/3	
	4/4	2/4			
23 Nov	2/4	3/4			
	3/4	4/4			
	4/4	3/4			
27 Nov	2/3	2/4			
% ES/Ov	76.7	73.7	% ES/Ov	82.3	86.7

Degenerative conditions were also found in the male reproductive apparatus: at anthesis, some anthers were without pollen grains and were white instead of yellow. However any effect on per cent set is unlikely

considering that pollen is dispersed in numbers largely in excess of those required for satisfactory pollination.

Pollination

Table 5 gives values for estimates (1989) or counts (1990) of the numbers of pollen grains deposited on the stigma and of the pollen tubes found in three places of the ovary, i.e. the style, the lower part of the pistil and entering the ovule.

TABLE 5: Scores (1989, score 1 equals very few, score 5 equals very many) or numbers (1990) of pollen grains on the stigma and of pollen tubes in three positions within the ovary of Chardonnay flowers from Hills vines (H) and Plain vines (P).

	1989		1990	
	H	P	H	P
Stigma	1.23	2.15	35.7	19.4
Style	1.17	2.17	10.1	9.4
Pistil	0.54	1.54	5.4	7.1
Ovule	0.00	1.00	0.2	0.5

In 1989, pollination and pollen tube growth were less frequent in flowers from H than from P where a number of penetrations of the ovule was observed. The pistils of flowers from H contained a number of branched and twisting pollen tubes. They occur under conditions unfavourable to their growth. No such abnormalities were observed in 1990 when pollination numbers on the two sites were almost equal. The differences between the two sites in the respective pollination data for the two seasons are somewhat surprising. To some extent, they correspond to the differences in seed number per berry, which were greater in 1989/90 than in 1990/91. Perhaps the very high temperatures at the beginning of the flowering period affected pollination at P while the flowers at H opened after the hot period had passed. Pollination did not occur before the cap was shed.

Pollen germination

Attempts were made to test the germination *in vitro* of pollen obtained from H- and P-vines. Difficulties were encountered in obtaining reliable results for reasons which are not clear. Germination percentages given in Table 6 are values for pollen grains which budded. Pollen from H did not develop past the stage of forming a "bubble" outside the wall of the pollen grain while some pollen from P continued development to form pollen tubes.

The relatively high germination percentage of pollen from P at 10° is surprising, because temperatures below 15° are considered to be inhibitory. However the results of the tests in the two seasons were too variable to allow definite conclusions on the effect of temperature on the germination of pollen grains or on possible differences in this respect between H and P.

TABLE 6: Per cent germination *in vitro* of pollen collected in the Hills and Plain vineyards and incubated at varying temperatures for varying periods. November 1989.

	HILLS					PLAIN				
Hours	2	6	24	48	72	2	6	24	48	72
℃										
5			0	2	3			2	5	7
10			9	7	12			7	18	25
15	3	2	11			3	18	33		
20	3	7	32			1	23	57		
25	2	13	27			19	24	50		

The viability of pollen, tested by its fluorescence after incubation in the presence of fluorescein diacetate, was of the order of 40% or less. Similar values have been reported in the literature.

As lack of pollination could be a major cause of poor set, the method of testing pollen viability and germination under various conditions is important and needs to be refined further. Such tests should include pollen derived from cultivars more resistant to unfavourable weather conditions, such as Riesling, which may serve as pollinators (either in open pollination or by applying pollen sprays) for the more susceptible cultivars.

Applications of boron

The boron applications in season 1989/90 had no effect on fruit set. As it seems unlikely that response to this minor element interacts with weather conditions the experiment was not repeated. The result agrees with episodic evidence by growers in the Adelaide Hills who have applied boron to their vines. However, boron deficiency has been reported in other crops in that region and more systematic studies may be advisable, considering that boron deficiency has serious effects on set. This may also apply to applications of zinc.

Application of proline

Proline has been reported to protect pollen of *Lilium* germinating *in vitro* from shock due to low or high temperatures (Zhang Hong-Qi, 1983). No reports are available to show whether this amino acid has any effect when applied to whole plants. Such applications to inflorescences in the Hills vineyard at the beginning of anthesis in 1989/90 and 1990/91 were without effect. This is not surprising considering the favourable weather conditions prior to and during the flowering period in these seasons.

The morphology of Chardonnay inflorescences and bunches

Very little information is available on the distribution of flowers and berries over the framework of inflorescences and bunches. Thus it is not known whether and how the differences in the size and composition of berries of the same bunch are related to their position. Any systematic positional differences should be known not only for studies on berry set but also for research on the growth and composition of berries and for

sampling to determine the maturation of the crop. The present project gave the opportunity to investigate on two sites differing considerably in plant vigour and climate such problems as the distribution of flowers and berries over the framework of the inflorescence or bunch, the relationship between flower position and timing of anthesis, the size distribution of developing and mature berries within the bunch, the relationship between seed number and berry size.

Determining berry set

Determining per cent berry set from counts of all flowers before anthesis and of all berries after set on sufficient numbers of inflorescences requires a prohibitively large amount of work and the destruction of many inflorescences. The method used here of bagging inflorescences from before anthesis until after set is reliable despite the creation of a modified environment and increased exposure to insect damage (here overcome by using insecticides within the bags), but it also is suitable only for detailed studies of fruit set.

Data from Pinot Noir vines in Burgundy showed a close relationship between the total flower number of inflorescences and that of their branches 3 and 4 (May, 1987). Counts of 19 Chardonnay inflorescences from the Hills vineyard in 1989 gave a similar highly significant regression ($Y = 34.01 + 6.847\ X$) with a coefficient of determination (R^2) of 92%. In 1990, a regression based on 19 uniformly sized inflorescences selected for other observations in the same vineyard was also highly significant, but its R^2 was only 48%. Adding only four additional but much larger inflorescences from the Plain-vineyard increased the R^2 to 77%. This confirms the suspicion expressed by Roy (1991) that the lack of relationship in similar determinations made by him was due to the uniformity of the selected inflorescences. On the other hand, some of 49 inflorescences collected in a Waikerie vineyard in 1989 were comparatively short but had many flowers on their proximal branches; they had the somewhat low R^2 of only 52%. Other measurements such as the length of the bunch stem or the number of its branches proved to be unsuitable indicators of the number of flowers per inflorescence.

To make flower number on branches 3 and 4 a useful predictor of total flower number, to be used for evaluating berry set, further determinations will be needed. In particular, it has to be established whether the relationship varies for any given cultivar depending on season, growing region and perhaps even clone.

Observations on Early Bunch Stem Necrosis

Casual observations in spring 1988 showed a high incidence of Early Bunch Stem Necrosis (Jackson and Coombe, 1988). Detailed checks in 1989 on a considerable number of vines used annually by the management of the Hills vineyard for monitoring vineyard performance failed to find a single

inflorescence with this disorder. However, many cases were found by casual observation in spring 1988 in the H-vineyard and in 1990 in P also. Clearly, seasonal factors play a role in determining the frequency of occurrence of this phenomenon which needs to be studied further both as to its cause and its effect on vine performance. In particular the respective timing of the death of peduncle portions and of their flowers need to be established.

CONCLUSIONS

This study has not led to practical solutions of the problem of poor set in "cool"-climate vineyards. It has led however to the conclusion that this condition is due to malfunction of the sexual apparatus under unfavourable environmental conditions, in particular at low temperature. Poor set in Chardonnay in the situations tested is not the consequence of shedding ovaries some days after they have commenced growth which is the usual characteristics of "coulure". Remedies for failed fertilization are not available at present. Altering the foliage canopy of the vine had no effect: set was as good or better on vines with dense canopies but growing under warm conditions than on vines with sparse canopies but exposed to lower temperatures. Ovule abnormalities were more frequent and pollen germination, pollen tube growth and seed number per berry tended to be less under cooler conditions even in seasons when set was normal. It was not possible to expand the project to include tests of such treatments as girdling, applications of cycocel, clonal differences, the effect of applying foreign pollen and applying zinc. Detailed studies on the effect of growth substances may also prove fruitful as much time has passed since the early studies on the effects of auxins, kinins and gibberellins were done. Experiments based on knowledge acquired since may help to make ovaries "stick" by replacing the effect of the missing fertilized ovules. Tests under controlled conditions will be necessary to overcome the difficulties inherent in the regular occurrence of poor set.

Until ways are found to overcome the problem, the sporadic losses due to poor set which occur in cool-climate vineyards may have to be accepted as part of the production risk in these usually high-value enterprises. This is obviously somewhat difficult, particularly where grapes are purchased from grapegrowers without wine-making facilities.

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APPENDIX

INVESTIGATIONS ON FRUIT SET

Investigations on fruit set in grapevines are not plentiful compared with studies of other factors influencing vine performance. Those which were made dealt mainly with cultivars with genetic defects of the reproductive apparatus. The apparent lack of interest in this problem is surprising considering that low fruit set is not rare in cultivars without such defects, causing partial or complete crop failure particularly in regions which produce highly praised and prized wines. Clearly the lack of relevant research is due to the experimental difficulties inherent in such investigations and the sporadic occurrence of the phenomenon. Here, some of these difficulties are discussed and suggestions are made for future research.

A. DETERMINING FRUIT SET

It is general in the plant kingdom that the success rate is small for flowers to develop into fruits. For instance, the inflorescences of such tropical plants as avocado and mango produce mostly only one fruit from one thousand or more flowers. Thus the grapevine must be considered as being efficient in this respect under normal conditions, with fruit set varying from less than 10% (Muscat of Alexandria) to over 50% (many small-fruited cultivars). Excessive fruit set can be as damaging as insufficient set because very tight bunches caused by excessive set are liable to suffer serious mould damage. Optimal set depends on the relative dimensions of the rachis and its berries.

As fruit set is defined as the ratio of number of fruits over number of flowers, the latter has to be determined by the time of anthesis. This is not easy and hardly ever done in field experiments where fruit set is not the main problem to be investigated. Instead, the number of berries per bunch is determined. Such a substitution would only be valid if

similar proportions of flowers were transformed into berries on all bunches. This is not the case; for instance per cent set varied between 7.1 and 64.8 among 65 bunches of Chardonnay sampled randomly in one row of vines.

The determination of the number of flowers per inflorescence by counts done prior to flowering is very time consuming and almost impossible without detaching the inflorescence. Therefore alternate methods are needed. An estimate of flower and berry number per bunch (Fa, Ba) can be obtained with a reasonable degree of accuracy by determining flower and berry numbers on inflorescence branches 3 and 4 (Fb, Bb) only, after standard regression lines of Fa on Fb and Ba on Bb have been established. This reduces the numbers to be counted by more than 4/5th. Present information suggests that this method gives sufficiently accurate estimates for large-scale investigations of Pinot Noir and Chardonnay in some situations, but further tests in a wider range of growing conditions and with other cultivars are still required. For detailed experiments on set, bagging of sample inflorescences at the time of anthesis for later counts of set berries and non-set ovaries appears to be the only practical solution, despite some obvious problems associated with it. Care has to be taken to include counts of the many ovaries which do not develop but remain attached to the bunch stem instead of lodging in the bag.

B. THE REASONS FOR POOR FRUIT SET

The normal sequence leading to berry set is fertilization of at least one of the usually four ovules of the ovary by a penetrating pollen tube, followed by rapid development of the embryo and endosperm and stimulation of cell division in the pericarp of the ovary. The failure of the flower to become a berry can be due (a) to faulty development of all ovules, (b) to failed fertilization leading to early abscission, (c) to early embryo or endosperm abortion resulting in either abscission of the slightly expanded ovary or in a seedless berry, or (d) to nutritional or hormonal insufficiency, causing early cessation of the growth of the ovary (millerandage) or to ovary abscission some days after anthesis (coulure). Many abnormalities of the reproductive apparatus have been described. Some cultivars, e.g. Ohanez, have female flowers (reviewed by Levadoux, 1946) and require cross pollination; others, such as Sultana and Currant, have ovules deficient in some respect and produce "seedless" berries, i.e. berries without hard seeds (reviewed by Ledbetter and Ramming, 1989); others still, e.g. Picolit, have sterile pollen grains because their outer coat (exine) lacks the pores through which the pollen tube normally emerges (Lombardo *et al.*, 1976). Berry set in cultivars with infertile or no pollen can be induced by cross-pollination. Seedless cultivars do not suffer from poor berry set despite the lack of viable ovules. The growth of the ovary pericarp is stimulated by the process of pollination. This points towards the possibility that further research in seeded cultivars on replacing the ovule-induced stimulus by chemical means may be fruitful.

Some cultivars appear to have a "normal" reproductive apparatus but low fruit set occurs often (Muscat Ottonel, Poulsard) for reasons which are not understood. The survival of such cultivars, often valuable for local production of speciality wines, may depend on finding means to ensure reliable fruit set. Some virus diseases as diverse as Fanleaf and Leafroll also cause poor fruit set; here also, the mechanism responsible for the damage is unknown. It is also possible that clones may differ genetically in the frequency of berry set under varying environmental conditions. Finally, the abscission of whole branches or branchlets of the inflorescence prior to anthesis (Early Bunch Stem Necrosis; Jackson and Coombe, 1988) is a recently described factor of poor cropping. It is not known yet whether failure in the development of the flowers contributes to this condition.

Most commercially grown cultivars set adequate numbers of berries when the weather is favorable but suffer when conditions during flowering are unfavorable. Information on the effect of such conditions have been gathered by Staudt (1982), Kassemeyer and Staudt (1981, 1982, 1983) and Koblet (1966), but more knowledge is needed. In particular it is not known how weather conditions affect the pre-anthesis development of the floral organs. Ideally, it should be possible to predict "flower strength" (May, 1970) and the commencement of anthesis from weather records. The mechanisms are unknown which cause cultivars or even clones of cultivars to differ in their susceptibility to unfavorable weather conditions.

C. FUTURE RESEARCH ON FRUIT SET

The reasons are many why the research effort in Australia and elsewhere on fruit set and millerandage is inadequate, when compared with other perhaps less important viticultural problems. In addition to those already listed, the location of Australia's research centres is important. Almost all were and still are situated in regions which are seldom affected by poor set. This occurs mainly in the newer, smaller viticultural regions which are becoming increasingly important for high-quality production but where research facilities are limited. Furthermore, funding bodies are reluctant to support studies of fundamental research which by necessity are long-term and which, if successful, give results which still need to be translated into practical application. However, in the case of fruit set, the present knowledge of flower formation, flowering and post-flowering development is clearly insufficient to plan remedial action against poor fruit set in the field.

The following studies, done with plant material raised under controlled environmental conditions, are required for the main cultivars (and perhaps also for some of their important clones):

(1) the development of the floral organs, pistil with ovules and anthers with pollen, under varying environmental conditions;
(2) the frequency of faulty development of embryo sac and pollen grains and its effect on success or failure of fertilization;
(3) the process of pollination, pollen tube growth and ovule fertilization under varying environmental conditions;
(4) the effect of early ovule development on the growth of the ovary pericarp;
(5) the process of abscission of ovaries and its possible prevention;
(6) the possibility of producing persistent "parthenocarpic" berries in normally seeded cultivars.

Without the knowledge obtained from such studies, any field experiments designed to improve set will be "educated guesswork", extended necessarily over many years to include variations in seasonal conditions.

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