Vitis **28**, 13—19 (1989)

Nuriootpa Research and Advisory Centre, Nuriootpa, South Australia

# Virus thermotherapy effects on the performance of a Muscadelle selection

by

M. G. McCarthy, R. M. Cirami and R. J. Van Velsen

## Einfluß der Virusthermotherapie auf die Leistungsfähigkeit eines Muscadelle-Klons

Zusammenfassung: Bei einem ertragreichen, aber Blattroll-infizierten Muscadelle-Klon waren nach Wärmebehandlung zur Virustherapie Traubenertrag und vegetatives Wachstum signifikant erhöht. Die wärmebehandelten Reben wiesen jedes Jahr mehr Beeren je Traube und schwerere Trauben sowie in 3 der 4 Untersuchungsjahre ein höheres Schnittholzgewicht auf. Bei den untersuchten Merkmalen der Beerenreife war kein Einfluß der Behandlung zu erkennen.

Key words: clone, selection, leaf roll, thermotherapy, bunch, shoot, yield, must quality, Australia.

## Introduction

Clonal selection has been widely adopted for the improvement of *Vitis vinifera* cultivars in most grape growing countries. Freedom from debilitating yield reducing virus diseases, in conjunction with high yield of satisfactory quality, have been the main selection criteria. High yield does not, however, infer virus freedom just as virus freedom does not necessarily result in high yield. Woodham *et al.* (1984 a) reported that high yielding clones of Sultana (syn. Thompson Seedless) used in Australia were infected with a combination of leaf-roll and yellow speckle viruses which when graft inoculated into Cabernet franc reduced its yield. Similarly the high yielding Cabernet Sauvignon selection A.S.70.2351 (Ikin 1983), widely used in Australia, shows a positive reaction to mild leaf-roll with Cabernet franc, LN33, BACO 22A and Mission indicators. In contrast, some of the lowest yielding clones in a Shiraz clonal evaluation trial were leaf-roll free (McCarthy 1986).

There are few reports in the scientific literature of thermotherapy effects on the performance of clones selected for yield, due probably to a divergence of attitudes between European and New World viticulturists over the need for thermotherapy in clonal selection programmes, and a concern over the effects of thermotherapy on the performance of high yielding clones. This paper reports the effect of thermotherapy for virus elimination on the performance of a high yielding leaf-roll infected Muscadelle selection.

## Materials and methods

### Preliminary

During 1968, 37 candidate clones were selected from Muscadelle vineyards in the Barossa and Clare Valleys of South Australia. Clones were selected for trueness to type, high yield and absence of virus induced autumn leaf colours.

Propagation material of each of the candidate clones was rooted and grown in glasshouse/shadehouse conditions and planted in a replicated trial in winter 1969. The trial was designed as a randomised block of 5 vine plots with 6 replicates.

Individual vine fruit weights were recorded for harvest years 1972—1977. Although there were significant differences in fruit weight between clones in each year, there were no consistent clones with significantly higher yield. After the 1977 harvest, aggregate data for years 1972—1977 was analysed (Table 1). There was no significant difference in yield between the top 20 clones. The highest yielding clon (32) had about 1 kg more fruit per vine than the lowest yielder.

Clone 32 was chosen for further evaluation. Virus indexing for 3 successive growing seasons indicated the presence of leaf-roll virus, using the leaf-roll virus indicator test plant Cabernet franc and Mission Seedling. The method of Goheen *et al.* (1965) was used to obtain plants free of leaf-roll virus.

3 well grown potted vines of clone 32 were placed in a thermotherapy cabinet heated to 37.8 °C. 92 d after being placed in the cabinet, 11 shoot apices were removed from a single vine and propagated under mist. On day 102, a single tip was taken and on day 119 two tips were taken from a single vine and propagated as before. 14 explants were grown on and multiplied to give sufficient plants of each for virus indexing.

Table 1

Average yield of Muscadelle selections evaluated for harvest years 1972—1977

Durchschnittlicher Traubenertrag der Muscadelle-Klone (Jahrgänge 1972—1977)

| Clone | Fruit weight (kg/vine) | Clone | Fruit weight (kg/vine) |
|---|---|---|---|
| 32 | 2.31 | 51 | 1.69 |
| 13 | 2.22 | 57 | 1.68 |
| 30 | 2.17 | 54 | 1.67 |
| 3 | 2.16 | 29 | 1.67 |
| 6 | 2.15 | 27 | 1.66 |
| 7 | 2.09 | 21 | 1.64 |
| 48 | 2.05 | 5 | 1.61 |
| 56 | 2.04 | 45 | 1.60 |
| 31 | 1.98 | 55 | 1.57 |
| 15 | 1.96 | 50 | 1.54 |
| 26 | 1.96 | 46 | 1.48 |
| 38 | 1.93 | 49 | 1.47 |
| 22 | 1.92 | 4 | 1.44 |
| 28 | 1.86 | 23 | 1.42 |
| 58 | 1.84 | 25 | 1.42 |
| 42 | 1.83 | 8 | 1.40 |
| 17 | 1.79 | 1 | 1.38 |
| 16 | 1.76 | 2 | 1.30 |
| 18 | 1.73 | | |
| | LSD (5 %) | | 0.61 |

Buds of the indicators Mataro, Cabernet franc, Mission Seedling, Baco 22 A, LN-33 and St. George were each green grafted to separate vines of the 14 explants, the candidate explant acting as the rootstock. 2 vines of each indicator/explant were grafted. Successfully grafted vines were planted in the field, trellised and drip irrigated to achieve rapid growth. Plants were classified positive or negative for the presence of yellow speckle, mild leaf-roll, leaf-roll, corky bark and fan-leaf complex for 3 consecutive growing seasons. Only 1 of the original 14 explants showed no reaction for the presence of mild leaf-roll or leaf-roll (Table 2).

### Thermotherapy comparison trial

Single internode cuttings of explant 102/1 were taken and rooted using mist propagation and grown on in a glasshouse before autumn hardening off in a shadehouse. Green tips of the original clone 32 were concurrently propagated. In November 1982, a randomised block design of 60 replicates of heat treated clone 32 (HT32) and the original 32 was planted on the Barossa Viticultural Research Centre. Drip irrigation at approximately 0.2 of weekly Class A Pan evaporation during the growing season and the use of nitrogen weekly in the irrigation water ensured early vine establishment and satisfactory growth. Vines were trained on a single wire 1 m high and spur pruned.

Table 2

Scores of virus presence or absence for the 14 explants of Muscadelle clone 32 tested on 6 different indicators

Ergebnisse der Virustests für die 14 Explantate des Muscadelle-Klons 32 mit 6 verschiedenen Indikatorreben

| Explant | Virus indicator | | | | | | |
|---|---|---|---|---|---|---|---|
| | Mataro (Yellow speckle) | Cabernet f. (Mild leaf-roll) | Mission (Leaf-roll) | Seedling (Yellow speckle) | Baco 22A (Corky bark) | LN-33 (Leaf-roll) | St. George (Fan-leaf complex) |
| 92/1 | 0/2[1] | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 92/2 | 0/2 | 1/1 | 0/1 | 0/1 | 0/2 | 0/2 | 0/2 |
| 92/3 | 0/2 | 2/2 | 1/1 | 0/1 | 0/2 | 0/2 | 0/2 |
| 92/4 | 0/2 | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 92/5 | 0/2 | 2/2 | 1/1 | 0/1 | 0/2 | 0/2 | 0/2 |
| 92/6 | 0/2 | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 92/7 | 0/2 | 2/2 | 1/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 92/8 | 0/2 | 2/2 | 2/2 | 0/2 | — [2] | 0/2 | 0/2 |
| 92/9 | 0/2 | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 92/10 | 0/2 | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 92/11 | 0/2 | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/1 |
| 102/1 | 0/2 | 0/2 | 0/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 119/1 | 0/2 | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 119/2 | 0/2 | 2/2 | 2/2 | 0/2 | 0/2 | 0/2 | 0/2 |

[1] No symptoms on either two of the test plants. For some combinations there was only one vine. A positive numerator indicates virus presence.

[2] No successful graft combinations.

At harvest 1985, the total weight of fruit and the number of bunches per vine were recorded. Prior to harvest in 1986, 1987 and 1988, a 50-berry sample, taken from 10 bunches chosen at random on each vine was collected for the determination of berry weight, pH, titratable acid and °Brix. In 1987, a 1 in 50 juice/water sample was prepared and immediately frozen for subsequent determination of malic and tartaric acid concentration using ion exchange chromatography. Sodium concentration was determined using flame photometry.

Approximately 1 month before the 1987 and 1988 harvest, the number of shoots per vines were counted. During winter in each year, all vines were pruned to the same bud number (as 2-bud spurs) and the weight of prunings recorded. Bud number was increased by about 6 buds/vine on all vines in each of the years reported.

Where appropriate, data was subjected to analysis of variance.

Table 3

Yield per vine and its components of Muscadelle 32 and heat-treated clone 32 for the 1985—1988 harvests

Ertrag je Rebe und Ertragskomponenten des unbehandelten und des wärmebehandelten Muscadelle-Klons 32 (Jahrgänge 1985—1988)

| Year | Selection | Fruit wt (kg) | No. of bunches | Bunch wt (g) | Berry wt (g) | Berries/ bunch | Pruning wt (kg) | Shoots |
|---|---|---|---|---|---|---|---|---|
| 1985 | 32 | 5.7 | 43 | 133.4 | ND | ND | 0.6 | ND |
| | HT32 | 8.0 | 48 | 167.2 | ND | ND | 0.8 | ND |
| | LSD (5 %) | 1.2 | NS | 10.7 | | | 0.1 | |
| 1986 | 32 | 9.6 | 57 | 168.9 | 1.4 | 119 | 0.8 | ND |
| | HT32 | 13.3 | 64 | 208.3 | 1.3 | 157 | 0.9 | ND |
| | LSD (5 %) | 1.3 | 5 | 15.5 | 0.1 | 12 | 0.1 | |
| 1987 | 32 | 9.9 | 67 | 146.9 | 1.9 | 76 | 1.0 | 47 |
| | HT32 | 12.0 | 69 | 173.1 | 1.9 | 94 | 1.3 | 50 |
| | LSD (5 %) | 1.5 | NS | 10.6 | NS | 7 | 0.2 | NS |
| 1988 | 32 | 16.5 | 145 | 113.8 | 1.2 | 95 | 1.1 | 55 |
| | HT32 | 20.2 | 152 | 133.5 | 1.2 | 111 | 1.1 | 58 |
| | LSD (5 %) | 1.6 | NS | 13.8 | NS | 10 | NS | NS |
| 4-year average | 32 | 10.4 | 78 | 133.6 | | | | |
| | HT32 | 13.4 | 83 | 161.1 | | | | |
| | LSD (5 %) | 1.2 | 6[1]) | 10.5 | | | | |

[1]) Significance not apparent due to rounding.
ND = Not determined.
NS = Treatments did not differ significantly at 5 %.

## Results

At harvest 1985, HT32 had significantly heavier bunches resulting in a significantly greater fruit weight per vine (Table 3). The weight of prunings removed from HT32 was significantly greater than from the original clone 32.

Although pruned to the same bud number, at harvest 1986 HT32 had more bunches which were heavier and resulted in a significantly greater fruit weight per vine (Table 3). The heavier bunch weight of HT32 was due to more berries per bunch; berry weight was lower. HT32 was lower in pH, titratable acid and °Brix (Table 4) but these differences were not significant and were probably yield related. The weight of prunings removed from HT32 was significantly greater compared with 32.

At harvest 1987, there was no significant difference in the number of shoots per vine, the number of bunches per vine or berry weight (Table 3). The significantly greater fruit weight on HT32 vines was due to more berries per bunch. There was no significant difference in malic acid concentration (Table 4). The tartaric acid concentration of 32 was significantly higher than that of HT32 although the difference was small. The juice of HT32 contained about 26 mg/l less sodium than 32. As for 1985 and 1986, HT32 had significantly heavier weight of prunings compared with 32 (Table 3).

At harvest 1988 HT32 vines yielded 3.7 kg/vine more than 32 (Table 3). This was a result of heavier bunches caused by more berries per bunch; there was no significant difference in the number of bunches or berry weight between selections. There was also no significant difference in the weight of prunings removed in 1988. Although there was no significant difference in °Brix (Table 4) between selections, HT32 was significantly lower in titratable acid and higher in pH compared with 32. The juice of HT32 was lower in sodium but not significantly. When averaged over 4 years HT32 yielded about 3 kg/vine more than 32 as a result of more and heavier bunches.

Table 4

Maturity indices of Muscadelle 32 and heat-treated clone 32 for the 1986—1988 harvests

Reifemerkmale des unbehandelten und des wärmebehandelten Muscadelle-Klons 32 (Jahrgänge 1986—1988)

| Year | Selection | °Brix | pH | Titratable acid (g/l) | Malic acid (g/l) | Tartaric acid (g/l) | Na (mg/l) |
|---|---|---|---|---|---|---|---|
| 1986 | 32 | 20.0 | 3.58 | 5.3 | | | |
| | HT32 | 19.5 | 3.39 | 5.1 | | | |
| | LSD (5 %) | NS | NS | NS | | | |
| 1987 | 32 | 21.0 | 3.42 | 6.3 | 4.4 | 6.3 | 86.3 |
| | HT32 | 21.0 | 3.41 | 6.1 | 4.3 | 6.0 | 60.1 |
| | LSD (5 %) | NS | NS | NS | NS | 0.1 | 10.5 |
| 1988 | 32 | 21.4 | 3.42 | 6.0 | | | 74.7 |
| | HT32 | 21.6 | 3.47 | 5.2 | | | 59.0 |
| | LSD (5 %) | NS | 0.02 | 0.2 | | | NS[1]) |

[1]) Significant at 6 %.

## Discussion

That all selections of Muscadelle chosen were leaf-roll infected contrasts with selections of other cultivars (McCarthy 1986) in the Barossa Valley and other grape-growing districts in South Australia, which in the majority have been free of the virus diseases listed in Table 2. This suggests that the original import of Muscadelle from a now unknown source into South Australia sometime after settlement 1836 was leaf-roll infected and all subsequent propagation material similarly infected. Virus transmission from rootstocks is unlikely because then, as now, South Australia was phylloxera free and all vineyards were planted with own-rooted vinifera.

Woodham *et al.* (1984 b) reported no significant improvement in the yield of H5 Sultana after the elimination of an attenuated strain of leaf-roll virus nor difference in the weight of 1-year-old wood. Differences in the sugar concentration in some seasons could not be consistently related to any treatment. In contrast, Woodham *et al.* (1984 a) found that mild symptoms of graft transmitted leaf-roll virus was associated with a yield reduction of 14 %, and severe symptoms with a decrease of 35 %. The lower yield was a response to fewer buds being retained to balance the reduced annual growth associated with both mild and severe leaf-roll inocula. In the experiment reported here, elimination of an attenuated leaf-roll strain resulted in a significant increase in yield caused by more and heavier bunches, a significant increase in the number of inflorescences per vine and the number of berries per inflorescence in each of the years recorded and an increase in the weight of prunings removed in all years except 1988. It was not determined whether the increase in the number of berries per bunch was due to an increase in the number of florets per inflorescence or an increase in percent florets set. Although the number of shoots per vine and hence percentage budburst were similar in each of the 2 years recorded, there remains the possibility that the pruning method of leaving a similar bud number on both treatments may have masked the true production potential of HT32. This is under investigation by comparing the effect of minimal pruning (Clingeleffer 1983) on both treatments as is the effect of thermotherapy and virus elimination on wine quality. The absence of consistent significant differences, excluding the small significant difference in tartaric acid content in 1987, in any of the maturity indices measured suggests that wine quality differences are unlikely.

Although only significant in 1 year, the lower concentration of sodium in the juice of HT32 was probably caused by a dilution effect of the increased crop on HT32, suggesting root uptake was limiting plant sodium concentration.

The trend for lower cluster number on heat treated H5 Sultana (Woodham *et al.* 1984 b) may have been due to the long heat treatment times ranging from 196 to 338 d. In the experiment reported here, minimum heat treatment times were used (102 d). Bovey (1980) cited several instances of heat treatment having positive or negative effects on the performance of propagules. Until recently thermotherapy has been the only reliable method of virus elimination; the method of Barlass *et al.* (1982) for virus elimination during tissue culture may be a more satisfactory technique for obtaining virus free explants with reliable yield performance without the need for time consuming thermotherapy and a 3-year delay for re-indexing as was necessary in the experiment reported here.

## Summary

Virus thermotherapy of a clonally selected, high yielding but leaf-roll infected Muscadelle selection resulted in significantly greater yield and vegetative growth. There were more berries per bunch and heavier bunches in each year and a greater weight of annual prunings of heat treated propagules in 3 of the 4 years reported. No consistent differences in selected maturity components were observed.



## Acknowledgements

We thank the staff of Northfield Horticultural Unit and the Nuriootpa Research and Advisory Centre for virus indexing and thermotherapy and for maintenance of the field experiments respectively.


## Literature cited


BARLASS, M.; SKENE, K. G. M.; WOODHAM, R. C.; KRAKE, L. R.; 1982: Regeneration of virus-free grapevines using *in vitro* apical culture. Ann. Appl. Biol. **101**, 291—295.

BOVEY, R.; 1980: Control of virus and virus-like diseases of grapevine: Sanitary selection and certification, heat therapy, soil fumigation and performance of virus-tested material. Proc. 7th Meeting Intern. Counc. Study of Virus and Virus-like Diseases of the Grapevine, Niagara Falls, 299—309.

CLINGELEFFER, P. R.; 1983: Minimal pruning — its role in canopy management and implications of its use for the wine industry. Proc. 5th Austral. Wine Ind. Tech. Conf., Perth, 133—140.

GOHEEN, A. C.; LUHN, C. F.; HEWITT, W. B.; 1965: Inactivation of grapevine viruses *in vivo*. Proc. Intern. Conf. Virus and Vector on Perennial Hosts, Davis, Ca. 255—265.

IKIN, R.; 1983: Accession List of Virus Tested Fruit Varieties in Australia. Commonwealth Department of Health, Canberra.

MCCARTHY, M. G.; 1986: Vine clonal selection trials 1958—1985. Nuriootpa Research and Advisory Centre, Department of Agriculture, South Australia, Tech. Rept. 100.

WOODHAM, R. C.; ANTCLIFF, A. J.; KRAKE, L R.; TAYLOR, R. H.; 1984 a: Yield differences between Sultana clones related to virus status and genetic factors. Vitis **23**, 73—83.

— — ; EMMENT, R. W.; FLETCHER, G. C.; 1984 b: Effects of thermotherapy and virus status on yield, annual growth and grape composition of Sultana. Vitis **23**, 268—273.


*Eingegangen am 14. 9. 1988*


M. G. MCCARTHY
Nuriootpa Research and Advisory Centre
South Australia Department of
Agriculture
P.O.B. 245
Nuriootpa S.A. 5355
Australia