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COMPENSATION AND STRESS RECOVERING RELATED TO LEAF REMOVAL IN *VITIS VINIFERA.*

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To my mother and
to my husband.

I. General Introduction

Pests, diseases and unfavorable weather conditions can strongly reduce the functional leaf area of the grapevines. Mechanical defoliation applied to promote a better microclimate of dense canopies also contribute to reduce the leaf surface. However, the repercussions of defoliation on quantity and quality of the fruit do not follow a linear pattern because grapevines have a strong capacity of compensation for the loss of leaf area by increasing the lateral shoots production (KLIEWER, 1970; KLIEWER and FULLER, 1973; WOLF *et al.*, 1986; HUNTER & VISSER, 1988; REYNOLDS and WARDLE, 1989), and also by increasing the leaf efficiency in terms of carbon fixation (BUTTROSE, 1966; KLIEWER, 1970; KLIEWER *et al.*, 1973; HOFÄCKER, 1978; HUNTER and VISSER, 1988; REYNOLDS and WARDLE, 1989). In the context of integrated pest management, this compensation ability must be taken in consideration to allow a more rational use of pesticides.

Here we present the results of our own investigations about the compensation capacity for stress induced by defoliation, its mechanisms and limitations. We investigated when defoliation can be performed without negative consequences for fruit yield, quality and carbohydrate reserves in the wood. We analyzed which phenomenons make compensation possible and which mechanisms are involved.

In a first step, the roles of main leaves and lateral leaves during the season were compared. The possibility of the remaining leaves assuming the missing leaves' functions to assure a normal crop, was investigated. The sensitive period when the plant can loose potential yield due to flower or berry abscission or to reduced bud fertility was examined. The accumulation of carbohydrate reserves in the wood after defoliation stress was also investigated. Furthermore, we examined the possibilities of recovery after a long period of stress.

In a second step, we investigated the influence of removing main or lateral leaves on gas exchange parameters and chlorophyll content in order to find the mechanisms that contribute to the increment of the physiological efficiency of the remaining leaves and therefore allow compensation.

II. Summary

In the present study, the compensation capacity for stress induced by defoliation in grapevines was examined.

In a first group of experiments, the effect of removing either main leaves or lateral shoots on final leaf area, yield components, fruit quality and starch reserves in the wood, was studied on mature field grown Pinot noir grapevines. The roles of main and lateral leaves were compared, and the sensitive period to berry drop was also examined. The aim of this study was to determine the mechanisms and limitations of compensation related to defoliation and to find out if the plants can recover after a prolonged defoliation stress.

Plants deprived of main leaves (L) produced more lateral shoots with a greater number of leaves. At vintage-time L plants had approximately the same leaf surface as the control plants. This was not the case during the second defoliation season: L plants still produced more leaves but of smaller size which caused a reduced total final leaf area. Plants bearing only main leaves (M) compensated for the absence of laterals by delaying leaf senescence and abscission. During the second defoliation season this plant group also produced leaves of smaller size.

Fruit yield was little affected by defoliation the first year but was 50% lower than the control in the second consecutive defoliation season for L plants. M plants showed no reduction on fruit production in both seasons. Must soluble solids and fruit coloration were slightly higher for L plants after the first but were not affected after the second defoliation season.

Final crop yield proved to be dependent on the existing leaf surface during bloom and 2 - 3 weeks after. The accumulation of sugar in the fruit seems to depend on the available active leaf surface during the period between veraison and fruit harvest.

The level of starch reserves in the wood was greatly reduced after 2 seasons of defoliation. Significant but low correlations were found between sugar content of the must and starch content of the wood.

Defoliation during early stages of berry development causes not only berry drop but also reduces bud fertility in the following season. This critical period is yet limited to 2 - 3 weeks after bloom.

Prolonged defoliation stress cannot be readily recovered after one season with normal cultural practices. This is due to the fact that flower initiation occurs when the defoliation stress is still being applied to the plant. Sugar accumulation

in the fruit and replacement of starch reserves proceeds normally already in the season following the stress. Complete recovery occurs, therefore, in the second season after the stress is removed.

In a second group of trials, gas exchange response to defoliation as well as chlorophyll content were investigated on field grown Pinot noir grapevines in order to study the compensation mechanisms related to leaf removal. Mature 16 years old bearing plants and 2 years old fruitless potted plants were compared. Defoliation treatments were performed one week after full bloom. Besides topping, 3 levels of main leaf removal (3, 6, or all 12 main leaves retained) were combined with 2 levels of laterals (all retained or all removed). The single leaf measurements (on the 11th main leaf from the base) were carried out from treatment time to fruit maturity.

Young potted plants and mature field grown plants showed very similar responses to defoliation treatments.

Plants with fewer main leaves showed higher photosynthetic rates and chlorophyll content than the control plants but only during the pre-veraison period. However, compensation was only partial because the increments registered on the gas exchange performance were insufficient to overcome the shortage of leaf area. Removal of lateral leaves resulted in the maintenance of higher assimilation rates of the remaining main leaves during fruit maturation. Plants without lateral leaves showed an increment on the water use efficiency. Chlorophyll content was always higher for defoliated plants.

Increase of the photosynthetic activity as response to defoliation was achieved mainly by enhancing the mesophyll conductance, but also by an increase of the stomatal conductance. Another compensation mechanism observed was a delay in leaf senescence and abscission.

III. Zusammenfassung

In dieser Arbeit wurde die Kompensationsfähigkeit der Rebe bei Entblätterungsstress untersucht.

In einer ersten Serie von Experimenten wurde an Blauburgunder Ertragsreben der Einfluss des Entfernens von Hauptblättern oder Geiztrieben auf Gesamtblattfläche, Traubenertrag und -qualität sowie Stärkegehalt des Holzes studiert. Die Bedeutung von Haupt- und Geizblättern sowie des Zeitpunkt der Entblätterung auf das Verrieseln wurde untersucht. Ziel der Studie war es, Kompensationsmechanismen und -limiten der Rebe unter Stressbedingungen kennenzulernen, wie sie durch eine Entblätterung verursacht werden. Ferner sollte die Erholung der Pflanze nach längerer Stresseinwirkung erforscht werden.

Wurden die Hauptblätter entfernt (L= nur Geizblätter), so bildeten die Pflanzen mehr Geiztriebe mit einer grösseren Anzahl Blätter. Dies führte nach dem ersten Stressjahr zu einer ungefähr gleichen Gesamtblattfläche wie bei den Kontrollpflanzen. Nach einem weiteren Stressjahr jedoch hatten die L-Pflanzen zwar weiterhin mehr Geizblätter, allerdings von geringerer Grösse. Hieraus resultierte eine im Vergleich zur Kontrolle verringerte Gesamtblattfläche. Pflanzen nur mit Hauptblättern (M) kompensierten das Fehlen der Geiztriebe mit verzögerter Blattalterung und späterem Blattfall. Auch hier ergab sich nach dem zweiten Stressjahr eine geringere Blattgrösse.

Der Traubenertrag der L-Pflanzen wurde im ersten Jahr durch die Blattentfernung kaum negativ beeinflusst, aber im zweiten Jahr war er 50% niedriger als in der Kontrolle. Bei den M-Pflanzen ergab sich in beiden Jahren kein verringerter Ertrag. Der Zuckergehalt der Trauben war in den L-Pflanzen im ersten Stressjahr leicht erhöht, nicht aber im zweiten Jahr. Die Beeren hatten während beider Jahre einen höheren Anthocyangehalt.

Die Blattfläche vom Zeitpunkt der Blüte bis 2-3 Wochen danach ist für den Traubenertrag entscheidend. Eine Entblätterung zu diesem Zeitpunkt verursachte nicht nur ein Verrieseln, sondern im folgenden Jahr zusätzlich eine reduzierte Knospenfruchtbarkeit. Die Zuckereinlagerung in den Trauben hängt von der assimilierenden Blattfläche während der Reifeperiode ab. Der Stärkegehalt im Holz war nach zwei Stressjahren erheblich reduziert. Es ergaben sich schwach positive Korrelationen zwischen Zuckergehalt des Mostes und Stärkegehalt des Holzes.

Die Zuckereinlagerung in der Traube und das Auffüllen der Stärkereserven im Holz ging bereits in der auf eine Stressbehandlung folgenden Saison normal vonstatten. Ein normaler Ertrag war ein Jahr nach einem längeren Entblätterungsstress jedoch noch nicht möglich, da die Bildung der Infloreszenzen bekanntlich während dieser Zeit (in unserem Fall der Stressperiode) einsetzt. Erst im zweiten Jahr kam es zu einer vollständigen Erholung der Pflanze.

Um die Blattkompensationsmechanismen zu studieren, wurde in einer zweiten Serie von Experimenten der Einfluss einer Entblätterung auf den Gaswechsel und den Chlorophyllgehalt an Blauburgunder Freilandreben untersucht. Sechzehnjährige, traubentragende Reben und zweijährige, nicht tragende Topfreben wurden verglichen. Die Entblätterung geschah eine Woche nach der Vollblüte. Drei unterschiedliche Varianten des Entfernens der Hauptblätter (Stehenlassen von 3, 6 bzw. 12 Hauptblättern) wurden mit zwei Varianten des Entfernens der Geizblätter (mit bzw. ohne Geizblätter) kombiniert. Gaswechsel und Chlorophyllgehalt wurden vom Zeitpunkt der Entblätterung bis zur Ernte am elften Hauptblatt durchgeführt.

Der Einfluss einer Entblätterung auf den Gaswechsel war sowohl an jungen Freiland-Topfreben wie an Ertragsreben sehr ähnlich.

Die Pflanzen mit weniger Hauptblättern hatten eine höhere photosynthetische Leistung und einen höheren Chlorophyllgehalt als die Kontrollpflanzen, jedoch nur bis zum Reifebeginn. Diese Leistungssteigerung von teilweise entblätterten Reben konnte jedoch den Blattflächenverlust nur zum Teil kompensieren. Das Entfernen von Geizblättern resultierte in einer Erhöhung der photosynthetischen Leistung der Hauptblätter während der Reifeperiode. Die Pflanzen ohne Geizblätter hatten einen höheren Wasserausnützungskoeffizienten. Der Chlorophyllgehalt war bei entblätterten Reben generell höher.

Die Erhöhung der photosynthetischen Leistung als Folge einer teilweisen Entblätterung war hauptsächlich das Resultat einer erhöhten Mesophyll-, aber auch einer verbesserten stomatären Leitfähigkeit. Ein weiterer Kompensationsmechanismus bestand in der Verzögerung der Seneszenz der Blätter und des Blattfalles.

IV. YIELD, FRUIT QUALITY, BUD FERTILITY AND STARCH RESERVES OF THE WOOD AS A FUNCTION OF LEAF REMOVAL IN *VITIS VINIFERA*. EVIDENCE OF COMPENSATION AND STRESS RECOVERING.

1. Abstract

The effect of removing either main leaves or lateral shoots on final leaf area, yield components, fruit quality and starch reserves in the wood was studied on mature field grown grapevines. The roles of main and lateral leaves were compared, and the sensitive period to berry drop was also examined. The aim of this study was to determine the mechanisms and limitations of compensation for stress induced by defoliation and to find out if the plants can recover after a prolonged defoliation stress.

Plants deprived of main leaves (L) produced more lateral shoots with a greater number of leaves. At vintage-time L plants had approximately the same leaf surface as the control plants. This was not the case during the second defoliation season: L plants still produced more leaves but of smaller size which caused a reduced total final leaf area. Plants bearing only main leaves (M) compensated for the absence of laterals by delaying leaf senescence and abscission. During the second defoliation season this plant group also produced leaves of smaller size.

Fruit yield was little affected by defoliation the first year but was 50% lower than the control in the second consecutive defoliation season for L plants. M plants showed no reduction on fruit production in both seasons. Must soluble solids and fruit coloration were slightly higher for L plants after the first but were not affected after the second defoliation season.

Final crop yield proved to be dependent on the existing leaf surface during bloom and 2 - 3 weeks after. The accumulation of sugar in the fruit seems to depend on the available active leaf surface during the period between veraison and fruit harvest.

The level of starch reserves in the wood was greatly reduced after 2 seasons of defoliation. Significant but low correlations were found between sugar content of the must and starch content of the wood.

Defoliation during early stages of berry development causes not only berry drop but also reduces bud fertility in the following season. This critical period is yet limited to 2 - 3 weeks after bloom.

Prolonged defoliation stress cannot be readily recovered after one season with normal cultural practices. This is due to the fact that flower initiation occurs when the defoliation stress is still being applied to the plant. Sugar accumulation in the fruit and replacement of starch reserves proceeds normally already in the season following the stress. Complete recovery occurs, therefore, in the second season after the stress is removed.

2. Introduction

Pests, diseases and unfavorable weather conditions can strongly reduce the functional leaf area of the grapevines. Mechanical defoliation applied to promote a better microclimate of dense canopies also contribute to reduce the leaf surface. However, the repercussions of defoliation on quantity and quality of the fruit do not follow a linear pattern because grapevines have a strong capacity of compensation for the loss of leaf area by increasing the lateral shoots production (KLIEWER, 1970; KLIEWER and FULLER, 1973; WOLF *et al.*, 1986; HUNTER & VISSER, 1988; REYNOLDS and WARDLE, 1989), and also by increasing the leaf efficiency in terms of carbon fixation (BUTTROSE, 1966; KLIEWER, 1970; KLIEWER *et al.*, 1973; HOFÄCKER, 1978; HUNTER and VISSER, 1988; REYNOLDS and WARDLE, 1989). Here we present the results of our own investigations about the compensation capacity for stress induced by defoliation, its mechanisms and limitations. In a first step, the roles of main leaves and lateral leaves during the season are compared. Then, the possibility of the lateral shoots to assume the missing main leaves' functions in assuring a normal crop, is investigated. The level of carbohydrate reserves after defoliation stress is also studied.

Incidence and severity of *Botrytis* bunch rot are reduced significantly when the leaves around grape clusters are removed (BONIFACE and DUMARTIN, 1977; WOLF *et al.,* 1986; KOBLET, 1988; ENGLISH *et al.,*1989). This management practice is more efficient if carried out early in the season (KOBLET, 1969) but it can reduce the fruit yield. On the other hand, if leaf removal is accomplished later there are no consequences for the final yield. Between bloom and a short time after the grapevines are susceptible to flower or berry abscission. If the supply of organic nutrients is not sufficient, berry drop due to a reduced assimilating surface can account for considerable crop loss. The sensitive period for berry shedding is examined in this study.

Another aim of this investigation is to verify if the plants stressed over a long period by defoliation, can completely recover after the stress is released.

3. Material and methods

Defoliation trials were carried out from 1985 to 1987 in 2 vineyards at the Swiss Federal Research Station for Fruit-Growing,Viticulture and Horticulture in Wädenswil, Switzerland.

3.1. Experiment 1: Influence of removing main leaves or lateral shoots on yield components, fruit quality and starch reserves in the wood. Evidence of compensation capacity.

3.1.1. Experimental design and plant material

In 1985, mature grapevines of *Vitis vinifera* L., cv. Pinot noir, clone M1/17 on 5C rootstock were used in this investigation. The plants were trained to double Guyot (cane pruning), with a spacing of 2.2 X 1.2 m. The experiment included 4 defoliation treatments replicated 5 times, each replicate being a single vine. All the non-fruiting shoots were removed at the end of June. Defoliation was accomplished on August 8, at about 6 weeks after full bloom. The 4 treatments were:

- C - Control: shoot tip, all leaves and lateral shoots retained
- CT - Control topped: topped to 16 nodes per shoot, all leaves and laterals retained
- L - Lateral leaves: topped to 16 nodes, all main leaves removed
- M - Main leaves: topped to 16 nodes, all lateral shoots removed at weekly intervals from this date onward.

Mature grapevines of Pinot noir, clone 2/45 on G-1 rootstock, cane pruned, were used in 1986. Defoliation treatments CT, L, M, each replicated 12 times, were carried out on July 8, one week after full bloom.

In 1987, half of the plants from each treatment group of the previous year experiment, was defoliated one week after full bloom (15.7.87). The other half was defoliated six weeks after full bloom (10.8.87). The treatments were applied to the same plants as in the previous year.

3.1.2. Harvest and data collected

The crop was harvested on October 23, 17 and 28 in 1985,1986 and 1987, respectively. Two days before fruit harvesting, the leaves of all vines under treatment were picked and the fresh weight, leaf color, leaf area and dry weight of all leaf laminae was recorded. The main leaves and lateral leaves from each vine were kept separately in plastic bags with suitable identification and were stored in a cold room at 1°C until measurement. During leaf harvesting, the number of main leaves, lateral leaves, and also the number of lateral shoots arising from each main shoot was recorded. Leaf color was scored using a 5 point scale as follows: 0= completely yellow; 1= 0-25% green; 2= 25-50% green; 3= 50-75% green; 4= 75-100% green. Leaf area was measured with an area-meter, (model LI-3100 from Li-cor, Inc., Lincoln, Nebraska, USA). Immediately after, the leaves were dried at 65°C in an oven and dry weight was noted. Just prior to fruit harvest, 100 berries from each vine were choosen randomly to determine mean berry weight. Afterwards they were used for color determination. Number of clusters per plant was registered. Number of berries per plant was calculated dividing crop weight by mean berry weight. Each vine was harvested individually, and after weighing the crop was crushed to determine soluble solids and acidity. For starch analysis, slices of wood were taken during pruning in the first week of February, 1988. The fifth internode from 4 mature canes was sampled from each plant. Using a sharp curve chisel and a hammer, a portion of trunk wood was equally sampled, leaving a small wound of no consequence for the plant. The samples were oven-dried at 65°C and then frozen until analysis.

3.1.3. Analytical procedures

3.1.3.1. Must quality

Total soluble solids were evaluated with a density meter (model DMA 46 provided by Anton Paar KG., Graz, Austria), total acidity was determined using an automatic end point titration unit (Dosimat 665, Impulsomat 614, Digital pH-meter 632, from Methrom AG, Herisau, Switzerland) on samples collected from each vine.

3.1.3.2. Fruit coloration

For the anthocyanin analysis, skins from 100g samples of berries from each plant were extracted with methanol acidified with 1% Hydrochloric acid. The berries were manually crushed, the skins were placed in 150ml flasks with 70ml of acidified methanol and shaken during 4 hours at ambient temperature. The extraction was repeated , first with 40 ml methanol during 3 hours and then with 30ml methanol during 3 hours. The extracts were mixed together and the absorbency was measured at 530nm, using a spectrophotometer, after appropriate dilution (1:50). Skin coloration results are given in percentage of the highest value of optical density observed.

3.1.3.3. Carbohydrate analysis

Wood samples were pulverized and 200mg dust were used for the extraction. Soluble sugars were extracted twice with 8 ml of 70% ethanol at 60°C for 30 minutes each. After evaporation and suitable dilution the sugar content was measured by the anthrone method as described by SCOTT and MELVIN (1953). Starch was then extracted twice with 8 ml of 1M Perchloric acid, one hour each time at 60°C and was measured after dilution by the same method. Absorbency readings were made at 620 nm with a spectrophotometer. Glucose was used as standard for both soluble sugars and starch. This method had previously been tested to ascertain that no structural carbohydrates would be extracted and to determine which of the solutions (0.5M NaOH and 1M Perchloric acid) would be more adequate for starch extraction. After the ethanol extraction, samples of ground wood and cotton wool (98% cellulose), were extracted both with 0.5 M NaOH and 1M perchloric acid. There were no carbohydrates extracted from the cotton wool samples neither with the NaOH nor with the perchloric acid solution. Starch extraction from the wood samples by the acid solution proved to be much more efficient than by the alkaline solution. For this reason, perchloric acid was used on the routine analyses.

3.2. EXPERIMENT 2: INFLUENCE OF TIME OF DEFOLIATION ON BERRY DROP, YIELD, FRUIT QUALITY AND BUD FERTILITY.

3.2.1. Plant material and experimental design

Mature grapevines of Pinot noir, clone 2/45 on G-1 rootstock, trained and pruned as in the previous experiments, were used in this trial. At full bloom in 1988, 4 marked inflorescences from 25 plants were enclosed in gauze bags. On the same day (June 21) all the plants were topped to 12 nodes per shoot. They were divided into 5 treatment groups replicated 5 times each as follows:

C - control
T1 - all main leaves removed at full bloom
T2 - all main leaves removed 2 weeks after full bloom
T3 - all main leaves removed 4 weeks after full bloom
T4 - all main leaves removed 6 weeks after full bloom

3.2.2. Harvest, data collected and analytical procedures

The gauze bags were emptied at weekly intervals until August, 24. The number of fruit caps, flowers and fruitlets were then counted. Plants were harvested on October 18. Cluster number, yield per plant, berry number, berries per cluster, fruit coloration, soluble solids and acidity of the juice were determined and recorded using the same methods as described in experiment I.

3.2.3. Bud fertility

The following winter one shoot per plant was used to test bud fruitfulness. During pruning on the first week of February, the shoots were cut into single node portions and placed into water. For this purpose a metal box (45 X 25 X 10cm) was filled with water and charcoal activated was added to prevent water deterioration. The nodes were held in place by a hardware screen of 11.5mm mesh size placed on top of the box. Incubation was carried out at a temperature

of 25°C. When the inflorescences were sufficiently visible, the number of clusters per node and number of sprouted buds were recorded.

3.3. EXPERIMENT 3: EVIDENCE OF RECOVERING CAPACITY AFTER DEFOLIATION STRESS.

3.3.1. Plant material and experimental design

The plants used in the experiment 1 in 1986 and 1987 were followed in the next 2 seasons to test if they would recover completely after 2 years of defoliation stress. They were all treated as the control plants (CT), i.e., besides topping, no other defoliation treatment was performed.

3.3.2. Harvest, data collected and analytical procedures

At harvesting time in October 1988 and 1989, cluster number, yield per plant, berry number, berries per cluster, fruit coloration, soluble solids and acidity of the juice were determined and registered using the corresponding methods already described in experiment I. During pruning in the first week of February 1989 pruning weight was recorded and samples from the trunk and from the 5th internode of one and 2 years old canes were taken from each vine for starch analysis.

3.4. Statistical analysis

Statistical analysis of data was performed utilizing the WIDAS statistical package (Wissenschftliches Integriertes Daten-Auswertungs-System, copyright Data General Corporation). Results were subjected to a factorial one way (treatment) or two way (treatment X time of treatment) analysis of variance with previous data transformation (square root transformation for counts or arc sine transformation for proportions) whenever required. Duncan's multiple range test was used to compare means. Linear regression followed by analysis of variance and F-test, was used to test relationships between some of the measured variables.

4. Results and discussion

4.1. Experiment 1: Influence of removing main leaves or lateral shoots on yield components, fruit quality and starch reserves in the wood. Evidence of compensation capacity.

4.1.1. Leaf area

In 1985 treatment L produced a 3 times larger lateral leaf area than the control topped plants which resulted in larger total leaf area (Table 1). This was achieved by a stronger production of lateral shoots with a greater number of leaves. In 1986 the same tendencies were observed but the differences were not as remarkable as in the previous year. This ability to increase lateral leaf area with increasing defoliation had also been observed by Weaver (1963); Kliewer (1970) and Reynolds and Wardle (1989). After 2 stressing seasons the L plants still produced more lateral leaves but they were smaller in average size (Table 2). Therefore, the lateral leaf area was inferior to that of the control plants. The same constraint on the leaf growth was observed in 1987 for M plants: they produced leaves of smaller average size than the control plants (Table 2). This could be due to an insufficient accumulation of reserves required for the initial growth as a consequence of the previous year defoliation.

On plants bearing only main leaves, all the developing lateral shoots were periodically removed and, unable to increase the leaf area, these plants had to adopt another strategy to compensate for the absence of lateral leaves: they delayed leaf senescence and abscission. This phenomenon is particularly evident in 1986 (Fig. 1). Canopies from M plants remained green until vintage time in contrast to CT plants which were not only yellowish but had already lost part of their leaves. It is apparent that the process of leaf senescence was somehow restrained in M plants. This overcharged leaves managed to remain physiologically younger and probably more actively assimilating. Therefore, it is evident that defoliation causes an increase of leaf efficiency of the remaining leaves to compensate the stress of reducing the source to sink ratio. Buttrose (1966), May *et al.* (1969), Kliewer (1970), Kliewer and Fuller (1973), Hofäcker (1978), Reynolds and Wardle, (1989) arrived at the same conclusion.

Table 1: Influence of removing main leaves or lateral shoots on number, size, area, and specific weight of main and lateral leaves at vintage time of the 1st stressing season. C: control; CT: control topped; L: only lateral leaves left; M: only main leaves left.

	C		CT		L		M		SE[1]
1985									
Total leaf area per vine (m^2)	5.54	a[2]	3.98	ab	4.00	ab	2.51	b	0.66
Main leaves area per shoot (m^2)	0.34	a	0.20	b	-		0.22	b	0.02
No. of main leaves per shoot	26	a	15	b	-		15	b	1.0
Main leaves size (cm^2)	132.3	a	137.3	a	-		154.2	a	7.7
Lateral leaf area per main shoot	0.23	ab	0.15	a	0.45	b	-		0.07
No. of leaves per lateral shoot	4	a	4	a	8	b	-		0.5
No. of laterals per main shoot	12	a	7	b	11	a	-		1.3
Lateral leaves size (cm^2)	47.9	a	44.9	a	50.8	a	-		3.9
1986									
Total leaf area per vine (m^2)	-		5.77	a	4.80	a	2.98	b	0.34
Main leaves area per shoot (m^2)	-		0.14	a	-		0.22	b	0.01
No. of main leaves per shoot	-		8	a	-		13	b	0.4
Main leaves size (cm^2)	-		172.5	a	-		170.0	a	5.4
Main leaves S.L.wt[3] ($mg.cm^2$)	-		4.9	a	-		5.6	b	0.1
Lateral leaf area per main shoot	-		0.30	a	0.36	a	-		0.03
No. of leaves per lateral shoot	-		7	a	7	a	-		0.3
No. of laterals per main shoot	-		7	a	9	b	-		0.3
Lateral leaves size (cm^2)	-		61.1	a	60.1	a	-		2.5
Lateral leaves S.L.wt[3] ($mg.cm^2$)	-		4.2	a	3.6	b	-		0.1

[1] Standard error of the mean

[2] Mean separation by Duncan's multiple range test. Means followed by the same letter within rows, do not differ significantly at 5% level.

[3] Specific leaf dry weight

Table 2: *Influence of removing main leaves or lateral shoots at 2 different times on number, size, area and specific weight of main and lateral leaves at vintage time of plants stressed over 2 seasons (1987). CT: control topped; L: only lateral leaves left; M: only main leaves left; T1 treated one week after bloom; T2: treated 6 weeks after bloom.*

1987	Defoliation treatment								Time of treatment						SE	Inter-action
	CT		L		M		SE[1]		T1		T2					
Total leaf area per vine (m^2)	6.51	a[2]	3.20	b	2.12	c	0.32		4.12	a	3.78	a			0.26	*[3]
Main leaves area per shoot (m^2)	0.24	a	-		0.19	b	0.01		0.22	a	0.21	a			0.01	ns
No. of main leaves per shoot	14	a	-		14	a	0.3		15	a	14	a			0.3	ns
Main leaves size (cm^2)	167.8	a	-		134.3	b	7.1		151.4	a	150.7	a			7.1	ns
Main leaves S.L.wt[4] ($mg.cm^{-2}$)	5.4	a	-		5.8	b	0.1		5.6	a	5.6	a			0.1	ns
Lateral leaf area per main shoot	0.38	a	0.29	a	-		0.04		0.35	a	0.31	a			0.04	ns
No. of leaves per lateral shoot	9	a	8	a	-		0.4		9	a	9	a			0.6	ns
No. of laterals per main shoot	7	a	8	a	-		0.4		8	a	7	a			0.4	ns
Lateral leaves size (cm^2)	52.4	a	43.1	b	-		2.4		49.3	a	46.2	a			2.4	*
Lateral leaves S.L.wt[4] ($mg.cm^{-2}$)	4.2	a	4.2	a	-		0.2		4.4	a	4.1	a			0.2	ns

[1] Standard error of the mean

[2] Mean separation by Duncan's multiple range test at 5%level. Means followed by the same letter within row sections do not differ significantly.

[3] ns,*, Nonsignificant or significant at 5% level, respectively.

[4] Specific leaf dry weight

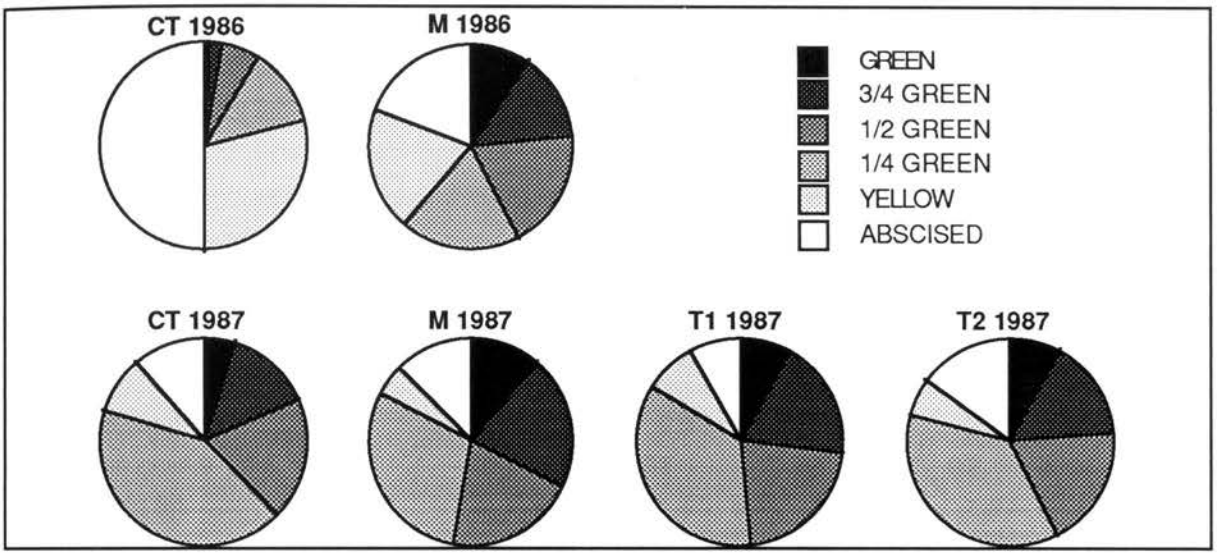

Figure 1: Influence of removing lateral shoots and of treatment time on leaf coloration and abscission at vintage time. CT: control topped; M: only main leaves left; T1: plants treated one week after bloom; T2 plants treated 6 weeks after bloom.

Main leaves from defoliated plants had a higher specific leaf weight (Tables 1 and 2). This is difficult to explain because leaf carbohydrate content was not measured but the visual impression was that the main leaves from M plants were thicker and greener. The higher specific weight should not be interpreted as accumulation of surplus carbohydrates in the leaves but as a consequence of a different physiological age: most of the main leaves of the control plants were already senescent and so the translocation of proteins out of the leaves associated with senescence (DALE, 1982) might explain this phenomenon.

4.1.2. Yield and yield components

In 1985 there were practically no significant differences in the yield components of the control and defoliated plants (Table 3). This was probably due to the time of treatment (6 weeks after full bloom) which was later in the season than in 1986 and also to the very high variations of the plants inside the treatments. Berry weight was the only yield component strongly affected by defoliation in 1985. An interesting result in this experiment was that the control plants which were not topped (C), did behave like the plants with all the main leaves removed (L). Both L and C plants showed throughout the season an intense production of new leaves (Table 1) which most probably affected the fruit growth. Control topped plants had an advantage over C plants because they did not have the actively growing shoot tip. Probably for this reason they achieved the best yield performance. Topping improves the fruit set

Table 3: Influence of removing main leaves or lateral shoots on fruit yield and quality during the first stressing year (1985 and 1986). C: control; CT: control topped; L: only lateral leaves left; M: only main leaves left.

	C		CT		L		M		SE[1]
1985									
Yield									
Fruit Yield (kg.m^{-2})	0.60	a[2]	0.91	a	0.57	a	0.76	a	0.11
Berries per cluster	55	a	58	a	54	a	52	a	5.6
Berry weight (g)	1.71	a	1.85	b	1.70	a	1.87	b	0.04
Fruit quality									
Must soluble solids (°Oe) (A)	84.8	a	84.5	a	86.0	a	83.1	a	1.4
Must total acidity (g.l^{-1}) (B)	13.3	a	13.2	a	12.0	b	14.0	a	0.4
Maturity Index M = (A×10): B	64	a	64	a	72	b	60	a	2.3
Fruit coloration (%ofhighestvalue)	86.4	a	73.0	b	86.7	a	71.2	b	4.2
Yield performance index = (M x yield)	103	a	152	a	107	a	117	a	16.2
1986									
Yield									
Fruit Yield (kg.m^{-2})	-		1.32	a	0.87	b	1.37	a	0.09
Berries per cluster	-		78	a	69	a	82	a	5.3
Berry weight (g)	-		1.68	a	1.28	b	1.65	a	0.03
Fruit quality									
Must soluble solids (°Oe) (A)	-		80.1	a	82.0	b	76.7	c	0.6
Must total acidity (g.l^{-1}) (B)	-		14.6	a	12.8	b	14.7	a	0.2
Maturity Index M = ((A ×10)/ B)	-		55	a	64	b	51	c	1.1
Fruit coloration (%ofhighestvalue)	-		55.3	a	93.2	b	66.2	a	2.0
Yield performance index = (M x yield)	-		174	a	134	a	169	a	12.9

[1] Standard error of the mean

[2] Mean separation by Duncan's multiple range test. Means followed by the same letter within rows do not differ significantly at 5% level.

because it eliminates a sink which would compete with the fruit for organic nutrients (COOMBE, 1962; KOBLET, 1966; VERGNES, 1981). QUINLAN and WEAVER(1970) showed that the direction of translocation of photosynthates from a newly exporting leaf during berry set stage was reversed after tipping: instead of upwards to the shoot tip the assimilates were diverted basipetally. In 1986 (as in 1985) plants bearing only lateral shoots had approximately one third lower fruit yield as compared with the CT plants, owing to lower berry number and weight (Table 3). Nevertheless, it has to be stated that treatment L represents a tremendous stress for the plant: when the main leaves were removed the lateral shoots were practically non-existent so that the plants looked completely stripped during the first weeks following the treatment. The first weeks after full bloom proved to be of great importance for the final berry number and size (experiment II). COOMBE *et al.* (1987) found that increments in dry matter per pericarp increased with initial berry size. They observed also that defoliation reduced the increments both on a per fruit and a per g fresh weight basis. In other words, if stage 1 of berry growth (described by HARRIS *et al.*,1968) is deterred, the rate of dry matter accumulation will be reduced and the final berry weight will be lighter.

An interesting feature is the fact that treatment M with only 50 to 60% (1985 and 1986) and 30% (1987) of the CT plants leaf area obtained equivalent results in crop yield and yield components. These results show that defoliation can be compensated by an increase in the physiological efficiency of the remaining leaves.

The final crop yield seems to depend on the existing assimilating surface during the first period of berry growth. During this critical period plants bearing only main leaves had the entire available leaf area consisting of fully grown actively assimilating and exporting leaves (KOBLET, 1969) and competition from a growing shoot tip or lateral shoots was excluded. That was not the case in the young growing lateral shoots from L plants whose leaves had to provide assimilates for their own growth, diverting them from the fruit.

In 1987 L plants showed a reduction of 50% in the fruit yield compared with the control (Table 4). They were weakened by 2 consecutive deprivation seasons[1] . Contrary to the predictions (MAY *et al.*,1969), there was no decline of the bud fertility (= number of clusters per shoot and number of shoots per plant). The yield reduction was mainly due to berry fall which was significantly stronger only in L plants, treated one week after bloom (L.T1). In treatment L, contrary to M and CT, the berry drop was more severe in case of the first treatment term. This fully agrees with the previous explanation: plants L.T1 were deprived of the main leaves 5 weeks before plants L.T2 and so the latter had the main leaves for a longer time, exactly during the hypothetical critical period. On the other hand, elimination of the immature growing leaves from the

[1] Plants used in 1987 were the same as in 1986, and the same treatments were made on the same plants.

Table 4: *Influence of removing main leaves or lateral shoots at 2 different times on fruit yield and quality in 1987 (2nd stressing season). CT: control topped; L: only lateral leaves left; M: only main leaves left; T1 treated one week after bloom; T2: treated 6 weeks after bloom.*

1987	Defoliation treatment								Time of treatment							Inter-
	CT		L		M		SE[1]		T1		T2		SE			action
Yield																
Crop yield (kg.m^{-2})	0.91	a[2]	0.45	b	0.70	a	0.08		0.67	a	0.70	a	0.06			ns[3]
No. of shoots per vine	11.0	a	11.3	a	11.3	a	0.5		11.0	a	11.3	a	0.4			ns
No. of clusters per shoot	1.9	a	1.8	a	1.9	a	0.1		1.9	a	1.9	a	0.1			ns
Berries per cluster	69	a	46	b	64	a	2.8		58	a	61	a	2.3			**
Berry weight (g)	1.47	a	1.10	b	1.20	b	0.04		1.27	a	1.24	a	0.04			ns
Fruit quality																
Must soluble solids (°Oe) (A)	77.6	a	73.1	b	77.0	a	0.8		77.1	a	74.7	b	0.7			ns
Must total acidity (g.l^{-1}) (B)	13.5	a	11.8	b	13.1	a	0.3		12.6	a	13.0	a	0.2			ns
Maturity Index M = ((A×10)/B)	58	a	62	a	59	a	1.5		62	a	58	a	1.2			ns
Fruit coloration (% of highest value)	52.6	a	68.1	b	55.1	a	2.5		63.4	a	53.8	a	2.0			***
Yield performance index = (M x yield)	125	a	65	b	98	a	10.1		96	a	96	a	8.2			*

[1] Standard error of the mean

[2] Mean separation within row sections by Duncan's multiple range test. Means followed by the same letter do not differ significantly at 5% level.

[3] ns,*,**,***, Nonsignificant or significant at 5%, 1% or 0.1% level respectively.

shoot tip (treatments CT, M) and lateral shoots (treatment M) which are sharing the same reserve pool with the fruit, represented a favouring circumstance for plants CT and M treated one week after bloom, as compared with the same treatments performed later.

4.1.3. Fruit quality

The variation in fruit quality is explained by the differences observed in the leaf area (Table 1). In the first defoliation season it is evident that lateral leaves were more efficient than main leaves in feeding the clusters during the ripening period and could fully compensate for the absence of main leaves. Fruit maturation was better in plants bearing only lateral shoots in 1985 and 1986 (Table 3). No differences were seen in the maturity index (sugar/acid ratio) in all treatments in 1987 (Table 4) because the lower sugar reading also coincided with a lower level of total acidity. L plants had a significantly lower degree Oechsle in the second stressing season (1987). This fact might be explained by the incapability of this plant group to reconstruct an adequate assimilating apparatus after the defoliation treatment to allow a satisfactory fruit ripening as had been accomplished in the previous season. Treatment M had the poorest sugar reading but the acid content of the juice was not different from the control plants. Fruit coloration followed more or less the same pattern of the sugar content. Treatment L had the highest color intensity on a per g basis, even in the second defoliation season because of the smaller berries with more specific area. If expressed on a per fruit basis, fruit coloration in 1987 would be 65%, 63% and 52% for treatments CT, L, M, respectively. Berry skin pigmentation and sugar content of the fruit juice were correlated both in 1985 ($r = 0.62$, $p < 1\%$) and 1986 ($r = 0.64$, $p < 0.1\%$) but no significant interdependence was seen in 1987. WEAVER (1963) reported a parallelism between the curves of sugar accumulation and change in amount of color during the ripening period. PIRIE and MULLINS (1980) state that sugar flux to grape tissues is one of the factors that govern the rate of phenolics accumulation. This relationship is easy to explain since the pigments of grapes are anthocyanidins glycosylated by glucose, forming the anthocyanins. They are synthesized from sugar, via shikimic acid and acetate provided by acetyl-co-enzime-A from the glycolitic pathway (SALISBURY and ROSS 1985). If glucose is limiting for the storage in the fruit, it is also limiting for the formation of the anthocyanins. Fruit coloration was also negatively correlated with crop level ($r= -0.76$, $p < 0.1\%$; $r= -0.79$, $p < 0.1\%$; $r=-0.61$, $p < 0.1\%$ in 1985, 1986, 1987, respectively). Similar findings are reported by PIRIE and MULLINS (1977) and SOMERS (1968).

The accumulation of sugar and color in the berries seems to depend on the available active leaf area during the period between veraison and harvest. During this period plants L have a canopy composed of relatively young leaves in opposition to

plants M which can only count on old leaves for the sugar accumulation in the berries. KOBLET and PERRET (1971) showed clearly a positive influence of lateral shoots on grape quality. STOEV *et al.*(1966), KRIEDEMANN (1968), KRIEDEMANN *et al.*, (1970) and ALLEWELDT *et al.*, (1982) agreed that photosynthetical activity is higher in recently formed leaves and that the peak of photosynthesis occurs when leaves attain full size. Then it decreases gradually with increasing age. Plants treated earlier in 1987 had a better maturation index and this was probably due to the earlier stimulation of the laterals growth.

Defoliated plants had no statistically significant reduction of the yield performance index (maturity index X yield per plant) in 1985 and 1986. However, in 1987, after two accumulated stressing seasons, plants bearing only lateral shoots revealed a 50% decrease in comparison to the control, probably due to the 50% smaller leaf area observed in this plant group.

4.1.4. Starch reserves in the wood

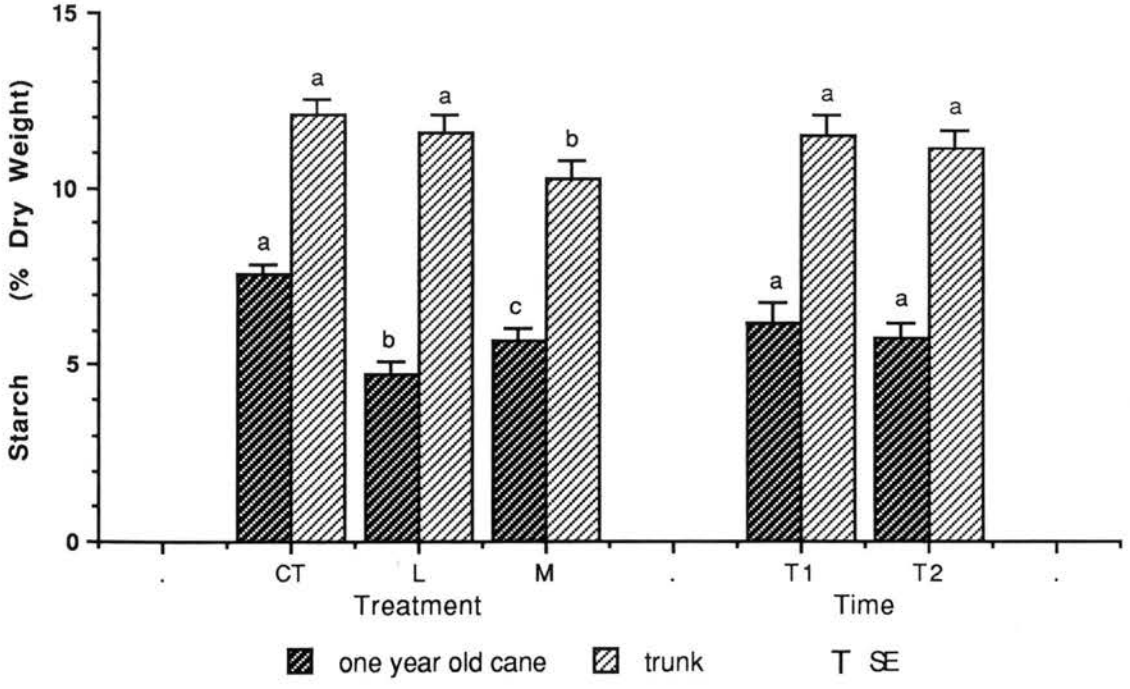

Figure 2: Influence of removing main or lateral leaves over two consecutive seasons on starch reserves in the wood. CT: control topped; L: only lateral leaves left; M: only main leaves left; T1: plants treated one week after bloom; T2: plants treated 6 weeks after bloom. Samples were taken on February,1988. Mean separation by Duncan's multiple range test at 5% level. Means of the same plant part, headed by the same letter, do not differ significantly.

After 2 stressing seasons defoliated plants had considerably less starch than the control plants (Fig. 2). Time of treatment did not influence the starch content of the

wood. The replacement of the carbohydrate reserves in the wood was most probably restrained to allow fruit maturation. It has to be noted that the plants did not have the possibility to produce carbohydrates and to refill the reserves after harvest because all the leaves had been removed at vintage time for measurement. The fruit clusters are the first sink organs to benefit of the phloem load because they have the advantage of being situated closer to the source organs and hence, their needs are satisfied before the other reserve organs in the plant. Shoot reserves (one year old cane) seem to be the most affected by the sink activity of the fruit in stressed plants since the reduction observed reached 40% for treatment L compared to the trunk reserves which were utmost 15% lower than the control. MATSUI *et al.* (1979) found that not only the translocation of photosyntates synthesized in the leaves but also the translocation of sugars converted from polisaccharides in shoots to the berries were responsible for the sugar accumulation in the fruit.

Final crop yield seems to depend on the existing assimilating area during bloom and some weeks after. According to YANG *et al.* (1980) retranslocation from the parent vine ceases by the flowering stage. During this critical time, main leaves are the only source organs. The lateral shoots are just starting their growth and act as sink organs. Removal of main leaves during this period means removal of the only available source organs and a reduction of the crop yield due to flowers and fruitlets abscission is an inevitable consequence. Main leaves are during this critical period the actively assimilating and exporting leaves and play the main role for the final fruit quantity. The correlation found between main leaf area and yield per plant in 1987 ($r = 0.87$, $p < 0.1\%$) supports this hypothesis.

The accumulation of sugar in the berries probably depends on the available active leaf area during the period between veraison and fruit harvest. During this period the lateral shoots are already source organs and provide the bunches with assimilates more efficiently than the main leaves. They represent the young and photosynthetically active part of the canopy in contrast to the main leaves which have already started the senescence process. These conclusions are based on the fact that the highest content of soluble solids was found in plants bearing only lateral leaves. Hence, the lateral leaves play the main role in fruit ripening. As result of insufficient assimilating surface during fruit maturation the carbohydrate reserves are not fully replaced in the parent vine. The fruit clusters seem to be stronger sinks than wood during the ripening period.

4.2. EXPERIMENT 2: INFLUENCE OF TIME OF DEFOLIATION ON BERRY DROP, YIELD, FRUIT QUALITY AND BUD FERTILITY.

4.2.1. Berry drop

Berry drop was particularly drastic in plants defoliated at full bloom (T1) and 2 weeks after (T2) (Fig. 3). Treatment T1 and T2 had a 50% and 25% lower berry set respectively as compared to the control. Plants defoliated later did not show increased berry drop in comparison with the control plants. In all the treatments the period of most intense berry abscission occurred between the second and third weeks after bloom and it stopped completely 6 weeks after bloom.

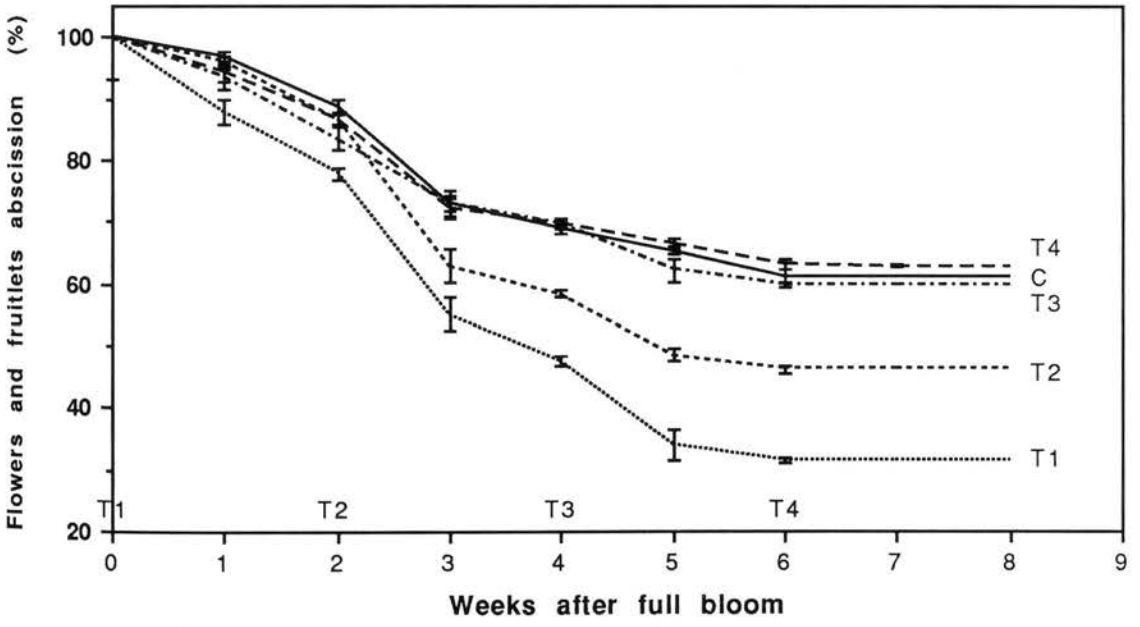

Figure 3: Influence of time of defoliation on flowers and fruitlets abscission. Vertical bars represent the standard error of the mean. C: control; T1: plants defoliated at full bloom; T2: plants defoliated 2 weeks after bloom; T3: plants defoliated 4 weeks after bloom; T4: plants defoliated 6 weeks after bloom. Defoliation consisted on removing all the main leaves.

This results show clearly that the critical period of berry drop due to an insufficient supply of organic nutrients to the inflorescence is limited to 3 weeks after bloom. This findings confirm the results obtained by KOBLET in 1966. This period seems to coincide with the period of rapid cell division which lasts according to HARRIS *et al.* (1968) 3 - 4 weeks after anthesis in cultivar Sultanina and, according to JONA and BOTTA (1988),

12 days in cultivars Barbera and Freisa. KASSEMEYER and STAUDT (1982), working with cvs. Weisser Burgunder and Gewürztraminer, found that the mitotic cycle of the zygotes requires 20 days. COOMBE (1960) states that most of the cell division in the pericarp occurs on the first 5 - 10 days after bloom, and that meristematic activity is limited to the first period of berry growth which lasts 45 days in cultivar Muscat. Beginning of cell differentiation, after cessation of cell division, could as well be one of the reasons that stops berry drop.

4.2.2. Yield and fruit quality

At vintage time mean berry weight was on all defoliated plants lower than in the control (Table 5). Furthermore, the earlier the defoliation was accomplished, the greater was the decrease in weight. KLIEWER, reported similar conclusions in 1970. Several investigators (BUTTROSE, 1966; MAY *et al.*, 1969; COOMBE *et al.*, 1987; KINGSTONE and VAN EPENHUIJSEN, 1989) showed that defoliation affects negatively berry growth and development. The earlier the reduction in assimilating surface is completed the earlier the scarcity of carbohydrates and the more drastic the consequences. Even after a possible reconstruction of the assimilating apparatus (by a stronger growth of the lateral shoots) the increments in dry matter per fruit would increase with initial berry size (COOMBE *et al.*, 1987) and so the final berry weight would be irremediably lower in the defoliated plants. A reduced number of berries together with a lower berry weight contributed to the decrease of the crop yield registered on plants defoliated during bloom and two weeks after. Treatments T3 and T4 had also a small reduction of the yield, although not statistically significant.

Soluble solids of the must were not affected by defoliation (Table 5) except for the last treatment time probably because at this time overall growth is slowed down and lateral shoot production is not efficient enough to enable a complete reconstruction of the assimilating apparatus. Acidity of the juice was lower than that of the control for all defoliated plants except the group defoliated at the last date. Fruit coloration, expressed on a per berry basis, was lower only for plants treated 6 weeks after anthesis; but if expressed on a weight basis was not influenced by defoliation.

It is clear that elimination of leaves in early stages of berry development cause a decrease of fruit yield, the critical period being limited to 2 - 3 weeks after full bloom. On the other hand, a strong defoliation stress applied later in the season can cause a decrease of fruit quality.

4.2.3. Bud fruitfulness

Bud burst and number of clusters per node on the following season were severely affected by defoliation (Table 5) in contrast to experiment 1. Flower clusters start to

develop during the beginning of bloom of the previous season (SHAULIS and PRATT, 1965). Therefore, an adequate supply of assimilates is essential for maximum flower development. The most affected plants were those treated during bloom and two weeks after. This period is particularly sensitive not only for the current year's fruit production but also for the following season's yield as well. KOBLET (1985) obtained similar results by covering the buds instead of removing the leaves.

Table 5: Influence of time of defoliation on fruit quantity and quality and on bud fruitfulness on the following season. C: control; T1: plants defoliated at full bloom; T2: plants defoliated 2 weeks after bloom; T3: plants defoliated 4 weeks after bloom; T4: plants defoliated 6 weeks after bloom. Defoliation consisted on removing all main leaves.

	Control	T1	T2	T3	T4	SE[1]
Fruit yield						
Mean berry weight (g)	1.5 a[2]	0.9 b	1.0 b	1.1 b	1.1 b	0.07
Crop yield (kg.m-2)	1.2 b	0.3 a	0.5 ac	0.9 bc	0.8 bc	0.16
Fruit quality						
Must soluble solids (°Oe)	77.7 ac	81.7 a	77.8 ab	75.2 bc	64.5 d	1.72
Must total acidity ($g.l^{-1}$)	13.4 a	11.7 b	11.6 b	12.0 b	13.5 a	0.35
Fruit coloration (%)	47.5[3] ac	82.2 b	72.0 b	58.9 a	35.4 c	4.22
	68.4[4] a	76.5 a	71.9 a	64.0 a	40.1 b	6.56
Bud fruitfulness						
Bud burst (%)	95.0 a	35.0 b	32.5 b	55.0 b	52.5 b	12.58
Number of clusters/node	1.5 a	0.5 b	0.3 b	0.8 b	0.5 b	0.20

[1] Standard error of the mean

[2] Mean separation by Duncan's multiple range test. Means followed by the same letter within rows do not differ significantly at 5% level.

[3] Percentage of highest value of optical density on a weight basis

[4] Percentage of highest value of optical density on a berry basis

4.3. EXPERIMENT 3: EVIDENCE OF RECOVERING CAPACITY AFTER DEFOLIATION STRESS

4.3.1. Yield and yield components

The effects of defoliation during the 2 previous seasons were still visible in 1988, even if the plants were allowed to keep all the leaves (Table 6). Again a 50% decrease of the crop yield was observed on the plants defoliated earlier. This decrease was mainly due to a reduced bud fertility (number of shoots and clusters per plant) and a poor fruit set which caused a lower cluster weight. In 1989, the plants that had been defoliated in 86 and 87, showed no yield reduction and even surpassed the control plants with respect to mean berry weight (Table 7). Mean berry weight seems to be the most sensitive measured item to describe the stress status of the plant. It follows that defoliation stress cannot be readily recovered during the following season. The inflorescence primordia are initiated one year before they bloom (SHAULIS and PRATT, 1965; HUGLIN, 1986) and defoliation will affect their development at the very beginning. In consequence the crop yield is affected not only in the season of defoliation but in the following one as well, even if leaf area is not limited any more.

4.3.2. Fruit quality

From 1988 (Tables 6 and 7) no differences were observed in all plants with respect to must soluble solids, sugar/acid ratio (maturity index) and fruit coloration. As soon as the leaf area is sufficient during the ripening period, sugar accumulation in the grapes proceeds without constraints.

4.3.3. Starch reserves in the wood

On February 1989 the starch reserves in the two years old canes of the defoliated plants (1986 and 1987) were significantly higher than those of the control plants (Fig. 4). No differences in the starch content of the wood could be observed on the other analyzed plant parts. Comparing these results with those obtained for the same plants in 1988 (Fig. 2), it is evident that an extra effort was undertaken to fill up the wood reserves in order to compensate for the shortage they had suffered during the 2 preceding seasons.

Table 6: Influence of main leaves or lateral shoots removal on the yield components and quality of the fruit on the 1st season following the defoliation treatment. In 1988, the plants were all treated as the control vines. They had been defoliated in 1986 and 1987. Treatments were: CT: control topped; L: only lateral leaves left; M: only main leaves left; T1: plants treated one week after bloom; T2: plants treated 6 weeks after bloom.

1988	CT		L		M		SE[1]	T1		T2		SE1	Inter-action
Yield components													
No. of shoots per vine	14.1	a[2]	12.2	b	14.0	a	0.5	13.4	a	13.5	a	0.4	ns
No. of clusters per shoot	1.9	a	1.3	b	1.4	b	0.08	1.5	a	1.5	a	0.07	ns
No. of berries per cluster	67.9	a	46.7	b	51.1	b	4.1	55.2	a	55.3	a	3.3	ns
Mean berry weight (g)	1.6	a	1.4	b	1.4	b	0.03	1.5	a	1.4	b	0.03	ns
Mean cluster weight (g)	109	a	69	b	73	b	6.4	86	a	81	a	5.2	ns
Yield (kg.m-2)	1.2	a	0.5	b	0.6	b	0.08	0.8	a	0.7	a	0.07	ns
Fruit quality													
Must soluble solids (°Oe)	75.3	a	76.7	a	76.8	a	0.6	76.6	a	75.9	a	0.5	ns
Maturity index	54.5	a	58.1	a	57.8	a	1.4	57.0	a	56.6	a	1.1	ns
Fruit coloration (%)	51.3	a	57.7	a	55.1	a	3.7	51.7	a	57.7	a	3.0	ns

[1] Standard error of the mean

[2] Mean separation by Duncan's multiple range test at 5% level. Means followed by the same letter within row sections do not differ significantly.

Table 7: Influence of main leaves or lateral shoots removal on the yield components and quality of the fruit on the 2nd season following the defoliation treatment. In 1988 and 1989, the plants were all treated as the control vines. They had been defoliated in 1986 and 1987. Treatments were: CT: control topped; L: only lateral leaves left; M: only main leaves left; T1: plants treated one week after bloom; T2: plants treated 6 weeks after bloom.

1989	CT		L		M		SE[1]	T1		T2		SE[1]	Inter-action
Yield components													
No. of shoots per vine	16.6	a[2]	16.0	a	15.9	a	0.7	16.4	a	16.0	a	0.6	ns
No. of clusters per shoot	2.0	a	1.8	a	1.8	a	0.08	1.9	a	1.9	a	0.07	ns
No. of berries per cluster	73.0	a	71.1	a	65.9	a	3.0	72.2	a	68.4	a	2.4	ns
Mean berry weight (g)	1.5	a	1.6	b	1.6	b	0.04	1.6	a	1.6	a	0.03	ns
Mean cluster weight (g)	110	a	114	a	106	a	4.6	112	a	109	a	3.8	ns
Yield (kg.m-2)	1.5	a	1.4	a	1.2	a	0.1	1.4	a	1.4	a	0.1	ns
Fruit quality													
Must soluble solids (°Oe)	78.3	a	78.5	a	79.8	a	0.9	78.6	a	78.5	a	0.8	ns
Maturity index	51.4	a	53.0	a	53.2	a	1.5	51.9	a	52.0	a	1.2	ns

[1]Standard error of the mean
[2]Mean separation by Duncan's multiple range test at 5% level. Means followed by the same letter within row sections do not differ significantly.

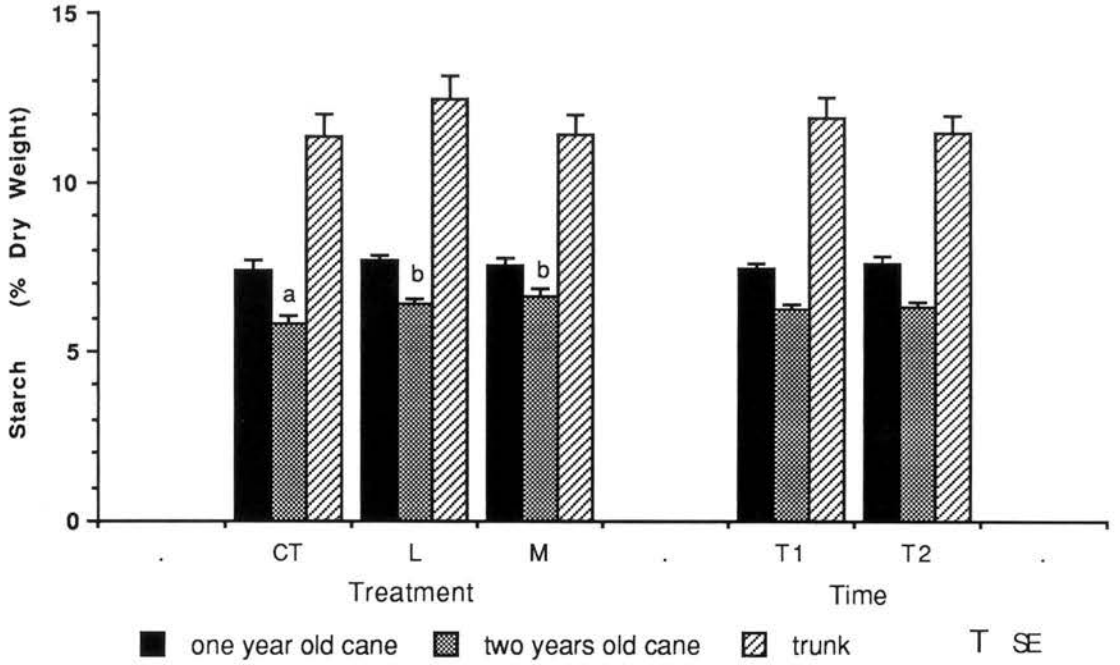

Figure 4: Starch content of the wood in February 1989. In 1988, the plants were all treated like the control vines. They had been defoliated in 1986 and 1987. Treatments were: CT: control topped; L: only lateral leaves left; M: only main leaves left; T1: plants treated one week after bloom; T2: plants treated 6 weeks after bloom. Mean separation by Duncan's multiple range test at 5% level. Means of the same plant part, headed by the same letter (or none), do not differ significantly.

These results show clearly that defoliated plants are able to fill up the reserve pool after one season without assimilating surface restrictions. Carbohydrate accumulation in the form of sugar in the fruit and storage as starch in the wood are related. In fact, significant correlations were found between must soluble solids and starch content specially in the one year old cane analyzed in the following winter. These correlations were however, rather low: $r = 0.32$, $p < 5\%$ in 1987 and $r = 0.46$, $p < 1\%$ in 1988. With increasing distance from the fruit to the reserve pool (two years old cane and trunk) this correlation was lower ($r = 0.32$, $p < 5\%$ for the two years old cane in 1988 and non significant in the trunk both in 1987 and 1988). Further experiments on this subject confirmed these assumptions and revealed a very good correlation ($r = 0.81$, $p < 0.01\%$) between must sugar content of the grapes and starch reserves of the wood (publication in prep.). Soluble solids in the fruit juice seem to be a good indicator of the carbohydrate status of the plant. The problem of carbohydrate partitioning in stressed plants needs further investigation. According to our results, both fruit and wood are storage sinks for carbohydrates during fruit maturation.

Prolonged defoliation followed by one season with a normal cultural practice is not enough for the complete recovery of the plants because flower bud initiation occurs when the assimilating potential is still being limited. It is therefore affected in its beginning, thus influencing the following season's crop yield. Carbohydrate accumulation in the fruit and in the wood on the other hand seems to depend only on the available leaf area during the ripening period. If the canopy is not restricted sugar accumulation both in the fruit and in the wood proceeds normally and allows already in the season following the defoliation stress the production of grapes with satisfactory sugar content and adequate starch reserves. Complete recovery occurs therefore in the second season after the stress is released.

For the survival of a perennial plant like the grapevine, to fill the wood reserves seems to be as important a goal as the maturation of the fruit (seeds).

5. Conclusions

Main leaves appear to play the main role for the final fruit quantity and, lateral leaves seem to be of primary importance in fruit ripening and starch accumulation in the parent vine.

Flower and fruitlets abscission occurs when the leaves are eliminated in early stages of berry development, causing a decrease of the fruit yield. However, the critical period is limited to 2 - 3 weeks after full bloom. On the other hand, a strong defoliation stress applied later in the season can cause a decrease of fruit quality.

Defoliation between bloom and two weeks after reduces bud fertility in the following season. This short period after bloom is particularly sensitive both for the current year's and following season's fruit production.

Prolonged defoliation followed by one season with a normal cultural practice is not enough for the complete recovery of the plants because flower bud initiation occurs when the assimilating potential is still being limited. Sugar accumulation in the fruit and replacement of starch reserves proceeds normally already in the season following the stress. Complete recovery occurs therefore in the second season after the stress is released.

6. Practical considerations

To obtain a good crop in quantitative and qualitative terms, the plants have to be properly supplied with leaves during 2 critical periods: fruit set and ripening period. If the main leaves are removed in the period between bloom and 3 weeks after, a reduction in the quantity of the yield of the current and following year is to be expected. In fact, berry drop is responsible for the yield reduction in the season of the stress, and a reduced bud fertility will affect the crop yield of the next year. On the other hand, during the ripening period the main leaves will already have started their senescence process and the main role in the sugar supply to the fruit and reserve organs is played by the lateral leaves. If the lateral shoots are not allowed to grow, a reduction in the sugar content of the fruit and lower starch reserves in the wood is the expected result.

In case that *Botrytis cinerea* presents a threat to the crop the leaves in the clusters area should be removed to promote a better aeration. This should however, not be done until the first critical period is finished. The lateral shoots should be left intact because they can very well compensate for the absence of the main leaves during the ripening period. Lateral shoots should never be removed above the cluster area because they supply sugars for fruit maturation and are thus directly involved in the final fruit quality.

In summary, main leaves should be present during fruit set to assure fruit quantity and bud fertility in the following season and lateral leaves should be present during fruit maturation to assure fruit quality and starch reserves in the wood.

V. INFLUENCE OF DEFOLIATION ON GAS EXCHANGE PARAMETERS AND CHLOROPHYLL CONTENT OF FIELD GROWN GRAPEVINES. MECHANISMS AND LIMITATIONS OF THE COMPENSATION CAPACITY.

1. Abstract

In order to study the compensation mechanisms related to leaf removal, gas exchange response to defoliation as well as chlorophyll content were investigated on field grown Pinot noir grapevines. Mature 16 years old bearing plants and 2-years-old fruitless potted plants were compared. Defoliation treatments were performed one week after full bloom. Besides topping, 3 levels of main leaf removal (3, 6, or all 12 main leaves retained) were combined with 2 levels of laterals (all retained or all removed). The single leaf measurements (on the 11th main leaf from the base) were carried out from treatment time to fruit maturity.

Young potted plants and mature field grown plants showed very similar responses to defoliation treatments.

Plants with fewer main leaves showed higher photosynthetic rates and chlorophyll content than the control plants but only during the pre-veraison period. However, compensation was only partial because the increments registered on the gas exchange performance were insufficient to overcome the shortage of leaf area. Removal of lateral leaves resulted in the maintenance of higher assimilation rates of the remaining main leaves during fruit maturation. Plants without lateral leaves showed an increment on the water use efficiency. Chlorophyll content was always higher for defoliated plants.

Increase of the photosynthetic activity as response to defoliation was achieved mainly by enhancing the mesophyll conductance, but also by an increase of the stomatal conductance. Another compensation mechanism observed was a delay in leaf senescence and abscission.

2. Introduction

Canopy management practices are very important to promote a suitable microclimate not only for fruit growth and maturation but also to avoid the propagation of fungal diseases. These practices reduce the assimilating leaf area. Pests, diseases and unfavorable weather conditions can also greatly reduce leaf area. In a previous paper (CANDOLFI-VASCONCELOS and KOBLET, 1990) we studied how defoliation stress affects fruit yield and quality as well as bud fertility and starch reserves in the wood. We found that grapevines have a strong capacity of compensation by increasing leaf area and we had evidence of an increment of the physiological efficiency of the leaves on defoliated plants. We also saw that plants bearing only main leaves compensated for the absence of laterals by delaying leaf senescence and abscission. HOFÄCKER (1978), working with green cuttings of Riesling X Silvaner and established potted Riesling plants under controlled environment, found that photosynthesis, stomatal conductance and chlorophyll content increased with increased level of defoliation. HUNTER and VISSER (1988 and 1989), working practically simultaneously with us in South Africa, report similar findings for mature field grown Cabernet Sauvignon.

The aim of our experiments was to study the possibilities and limitations of the compensation capacity related to leaf removal in Pinot noir grapevines. A study on compensation to leaf removal in grapevines was up till now, to our knowledge, never reported in the literature. We investigate the influence of removing main or lateral leaves on gas exchange parameters and chlorophyll in order to find possible mechanisms contributing to this increment of the physiological efficiency of the remaining leaves.

Non-destructive gas exchange studies on grapevine have with few exceptions (for instance, WILLIAMS and SMITH, 1985; DOWNTON *et al.*, 1987; HARREL and WILLIAMS, 1987; HUNTER and VISSER, 1988 GOODWIN *et al.*, 1988; SCHULTZ, 1989) been conducted with potted plants (e.g., KRIEDEMANN, 1968; KRIEDEMANN *et al.*, 1970; HOFÄCKER, 1978; LIU *et al.*, 1978; EIBACH and ALLEWELDT, 1984; DÜRING, 1984; KAPS and CHAOON, 1989) mostly in greenhouses or growth chambers.

In this work we also compare the gas exchange response of 2 plant systems to defoliation: mature 16 years old field grown plants and 2 years old potted plants grown under field conditions. The grapevine, like other woody perennials, has a long juvenile period during which the growth is only vegetative. It is very difficult to obtain young fruit bearing Pinot noir experimental plants. Working with young

potted plants is nonetheless a much simpler approach to use in physiological studies. However, in order to know if we can simulate mature plants using young potted plants we propose to compare both plant systems. We tried to find out whether these 2 plant systems react the same way when they receive the same defoliation treatments.

3. Materials and methods

3.1. Plant material

Mature plants: Field grown sixteen years old grapevines, cv. Pinot noir, were used in this investigation. The plants were trained to double Guyot (cane pruning) with a spacing of 2.2 X 1.2 m. All the weak and non-fruiting shoots were removed on May, 26 leaving an average of 14 shoots per plant.

Potted plants: Two years old grapevines, cv. Pinot noir, were planted on April 18 in 5 l pots containing soil. The pots were buried in an open field. On May 25 the plants were thinned to one shoot per plant. These young plants had no fruit.

3.2. Defoliation treatments

The experiment included 6 defoliation treatments. Each treatment consisted of 5 single-plant replications for the mature and 8 for the potted vines in a randomized complete block design. Defoliation was accomplished June 29, about 1 week after full bloom, on the mature plants and July 13, at the 16 leaf stage for the potted plants which corresponds to the same phenological stage as for the mature plants. Just before defoliation, all the plants were topped to 12 nodes per shoot. The defoliation treatments were:

12: All the main leaves were retained

6: The upper 6 main leaves were retained

3: The upper 3 main leaves were retained

In half of the plants from these 3 groups lateral shoots were removed periodically as they emerged. On the other half, laterals were allowed to grow:

LR: Laterals removed

LP: Laterals present

The resulting experimental design was a 3 by 2 factorial, with 3 levels of defoliation (3, 6, or all 12 main leaves retained) and 2 levels of laterals (all retained or all removed).

3.3. Gas exchange measurements

Net CO_2 assimilation rate (A), transpiration rate (E), stomatal conductance to CO_2 transfer (g_s), mesophyll or intracellular conductance to CO_2 transfer (g_m) and intercellular CO_2 partial pressure were measured on the 11th main leaf (from the

base) of one shoot per plant. For this purpose we used a portable LCA-2 system (Analytical Development and Co. Ltd., Hoddesdon, Herts, England). This apparatus consists of 4 units: an infra-red gas analyzer model LCA-2, a Parkinson leaf chamber (PLC-B for broad leaf), a data-logger (DL2) and an air supply unit with an incorporated mass flow meter (ASUM). The instrument operates as an open system. Air flow rate was adjusted to 200 $ml.min^{-1}$. A value of 0.3 $m^2 \cdot s \cdot mol^{-1}$ was obtained for the boundary layer resistance using PARKINSON'S method (1984). All measurements were carried out under saturated light conditions, between 10:00 and 12:00 a.m. and the air temperature ranged between 26 and 31°C. The measurement period started just before the defoliation treatments were accomplished and proceeded until fruit harvest.

Except for g_m and water use efficiency (WUE), all the calculations were performed using the built-in equations of the program version 5.1 of the data-logger DL2 (Analytical Development and Co. Ltd., Hoddesdon, Herts, England).

g_m was calculated using the calculated values supplied by the data logger for C_i and A and assuming that CO_2 partial pressure at the site of carboxilation (C_c) is zero:

$$A = \frac{C_i - C_c}{r_m}; \; g_m = \frac{1}{r_m}$$

By assuming $C_c = 0$, the carboxilation resistance is implicitly included in the estimate of r_m (JARVIS, 1971).

Water use efficiency was calculated as the quotient between the photosynthetic and transpiration rates (WUE = A / E).

3.4. Data collected and analytical procedures

The chlorophyll content was determined only for the potted vines on 4 occasions during the same period as for the gas exchange measurements using 4 other groups of plants treated exactly the same way. For these measurements 5 leaf discs (8mm in diameter) were taken from the 11th leaf of 8 plants per treatment on each date using a cork borer. Chlorophyll was extracted with the method described by HISCOX and ISRAELSTAM, 1979. Chlorophyll contents and chlorophyll a/b ratio were computed using the equations of ARNON (1949).

Leaf area was measured on the same plants with an area-meter (model LI-3100 from Li-cor, Inc., Lincoln, Nebraska, USA).

3.5. Statistical analysis

The data logged in the above mentioned DL2 data-logger was transfered via an interface to the central computer for statistic analysis. The WIDAS statistical package (Wissenschaftliches Integriertes Daten-Auswertungs-System, Data General Corporation) was used for statistical analysis of data. Results were subjected to a two-way analysis of variance (number of main leaves left X presence or absence of lateral shoots). Duncan's multiple range test was used to compare means.

4. Results

It is evident that veraison is an important physiological event having not only an influence on the fruit itself but also on leaf factors related to photosynthesis (Fig. 1: A1-A5 and 2: A1-A5). For convenience, the measuring season was divided into 2 distinct periods which reflected major changes in gas exchange parameters. For mature plants period I was the time interval between defoliation treatment (one week after full bloom) and veraison (seven weeks after treatment); period II, from veraison to fruit maturity, corresponds to the ripening period. For young potted plants period I elapses during the first 5 weeks following defoliation treatment and period II lasts from this date until "vintage time".

4.1. Influence of removing main leaves

Fig. 1 shows the main effect of removing main leaves on gas exchange parameters of mature plants bearing fruits (Fig. 1: A1-A5) and young fruitless potted plants (Fig. 1: B1-B5). It is obvious that most of the responses to main leaf removal are confined to period I or pre-veraison.

During period I photosynthetic rate (A) (Fig. 1: A.2 and B.2), mesophyll conductance (g_m) (Fig. 1: A.1 and B.1) and stomatal conductance (g_s) (Fig. 1: A.3 and B.3) were higher for either mature or young potted plants with fewer main leaves. There was no treatment effect on the intercellular CO_2 partial pressure (C_i) and water use efficiency (WUE) for the mature plants. Young potted plants during period I showed in response to defoliation, higher efficiency of carbon fixation per unit of water loss (Fig. 1: A.4 and B.4 respectively).

During period II no treatment effect could be detected on mature plants in any of the parameters studied (Fig. 1: A1-A5). Young potted plants showed higher values of A and g_s on treatments 3 and 6 as compared to treatment 12 during the same period (Fig. 1: B.2 and B.3, respectively). No treatment effect was observed on g_m (Fig. 1: B.1), C_i (Fig. 1: B.4) and WUE (Fig. 1: B.5) on young vines during period II.

Fig. 3 shows the effect of removing main leaves on total chlorophyll content, chlorophyll a/b ratio, and total leaf area on young potted plants. During period I chlorophyll increased with increasing level of leaf removal (Fig. 3: 1). Chlorophyll a/b ratio showed a peak on the 2nd week after treatment and was higher for plants with a reduced number of main leaves (Fig. 3: 2). During the second period, chlorophyll content was higher in leaves of defoliated plants and

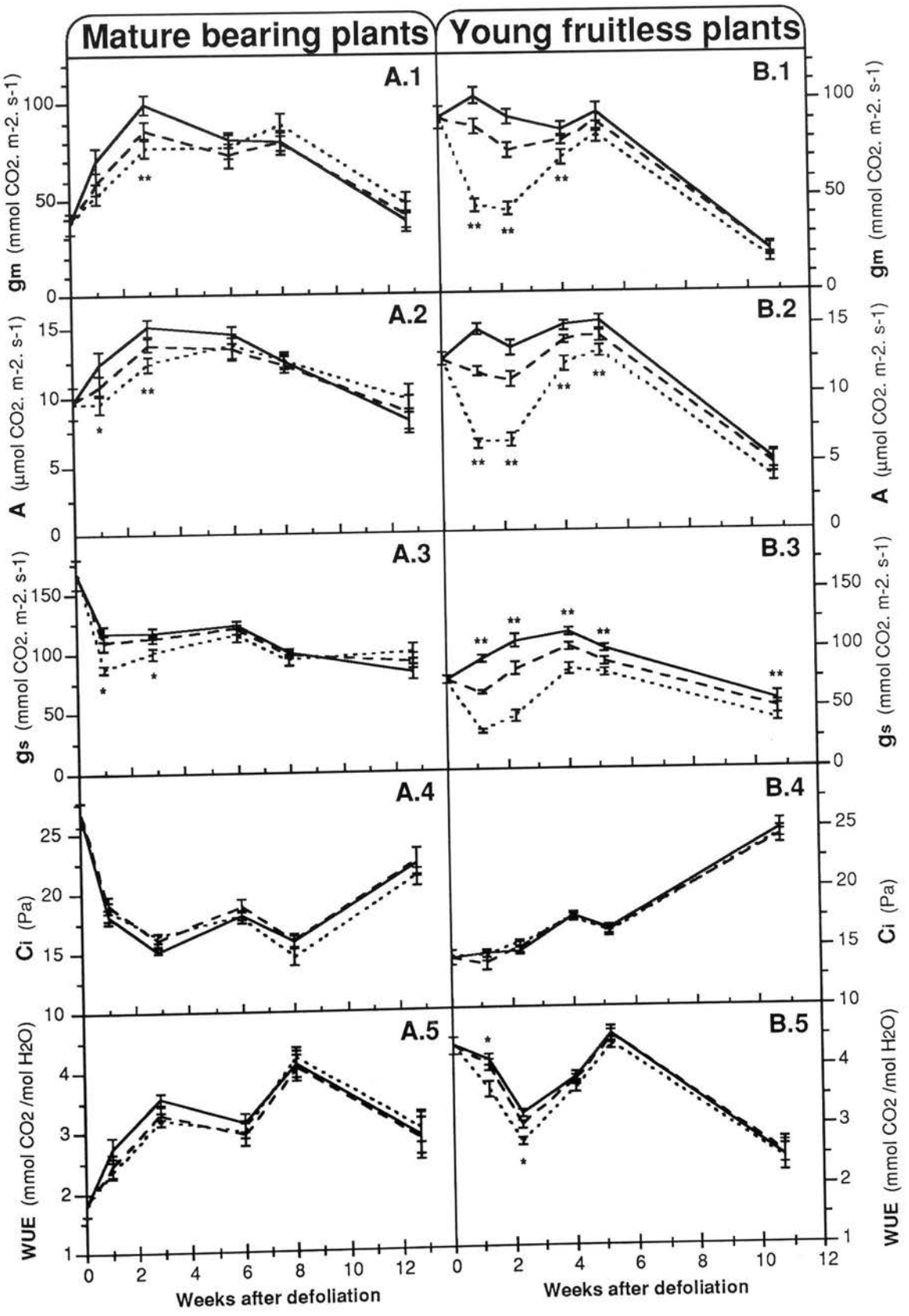

Figure 1: Effect of removing main leaves on mature, fruit bearing plants (A.1-A.5) and young fruitless potted plants (B.1-B.5) on the mesophyll conductance g_m (A.1 and B.1), net CO_2 assimilation rate A (A.2 and B.2), stomatal conductance to CO_2 transfer g_s (A.3 and B.3), intercellular CO_2 partial pressure C_i (A.4 and B.4), and water use efficiency WUE (A.5 and B.5) of the11th main leaf (from the base) of one shoot per plant. Full line: plants with 3 main leaves left; dashed line: plants with 6 main leaves left; dotted line: plants with all 12 main leaves left. Vertical bars: standard error; * and **: statistically significant at the 5% and 1% level of probability, respectively.

decreased continuously in all treatments (Fig. 3: 1). There was no treatment effect on the chlorophyll a/b ratio during this period (Fig. 3: 2).

Just after treatment, the remaining leaf area of plants with reduced main leaf number was 30% for treatment 3 and 60% for treatment 6 of that of the control plants, respectively. Leaf area increased for all treatments attaining a maximum at the end of period I (Fig. 3: 3). Total leaf area decreased during period II for all treatments but leaf abscission was hastened on plants having more main leaves (Fig. 3: 3).

4.2. Influence of removing lateral leaves

The main effect of removing lateral leaves on gas exchange processes of mature fruit bearing and young potted vines is plotted on Fig. 2. In contrast to the responses to main leaf removal, responses to lateral leaf removal were more pronounced during period II.

During period I practically no treatment effect could be detected on mature grapevines (Fig. 2: A1-A5). In contrast, young potted vines showed higher mesophyll conductance, photosynthesis, and stomatal conductance on plants with no lateral shoots (LR) (Fig. 2: B1, B2, and B3 respectively).

During period II both plant systems showed higher g_m, A, g_s, and WUE on plants whose lateral shoots were removed (Fig. 2: A1-A3, A5 and B1-B3, B5 respectively). Mature and young potted plants showed lower C_i on plants without lateral shoots (Fig. 2: A.4 and B.4, respectively).

Fig. 4 shows the response of removing lateral shoots on total chlorophyll content, chlorophyll a/b ratio of the remaining main leaves and total leaf area of young potted plants. During period I chlorophyll content increased for both plants with and without laterals (Fig. 4: 1). Chlorophyll a/b ratio was higher for LP plants but after the 3rd week post treatment the tendency was inverted (Fig. 4: 2). During the second period LR plants had higher levels of chlorophyll and also higher chlorophyll a/b ratios as compared to LP plants (Fig. 4: 1 and 2).

During the first 5 weeks post treatment, total leaf area remained constant for LR plants and increased more than 2 fold for LP plants (Fig. 4: 3). Total leaf area decreased during period II due to leaf abscission on LP plants but LR plants managed to maintain a constant leaf area by delaying leaf abscission (Fig. 4: 3).

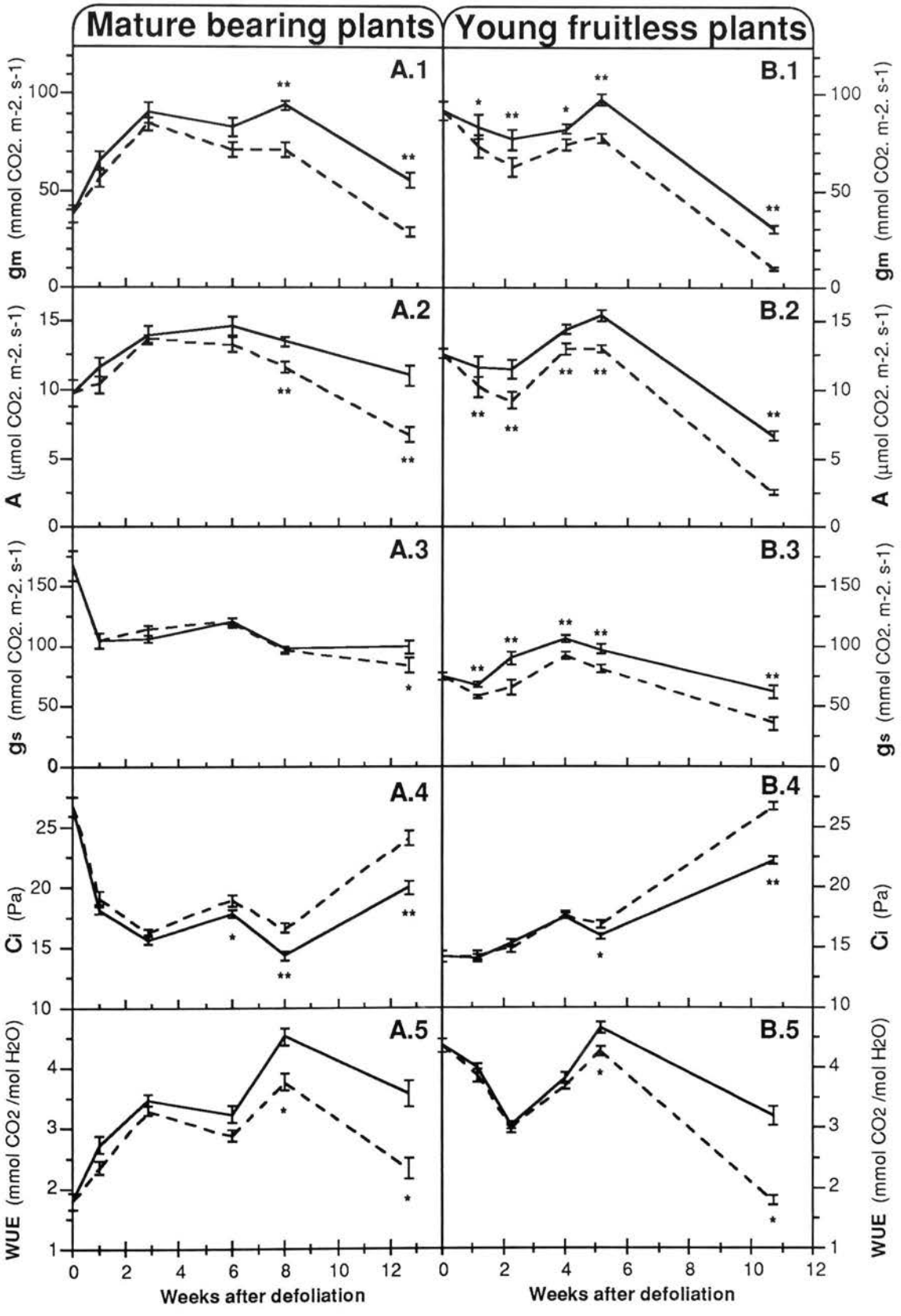

Figure 2: Effect of removing lateral leaves on mature, fruit bearing plants (A.1-A.5) and young fruitless potted plants (B.1-B.5) on the mesophyll conductance g_m (A.1 and B.1), net CO_2 assimilation rate A (A.2 and B.2), stomatal conductance to CO_2 transfer g_s (A.3 and B.3), intercellular CO_2 partial pressure C_i (A.4 and B.4), and water use efficiency WUE (A.5 and B.5) of the11th main leaf (from the base) of one shoot per plant. Full line: treatment LR, plants without lateral shoots; dashed line: treatment LP, plants with lateral shoots. Vertical bars: standard error; * and **: statistically significant at the 5% and 1% level of probability, respectively.

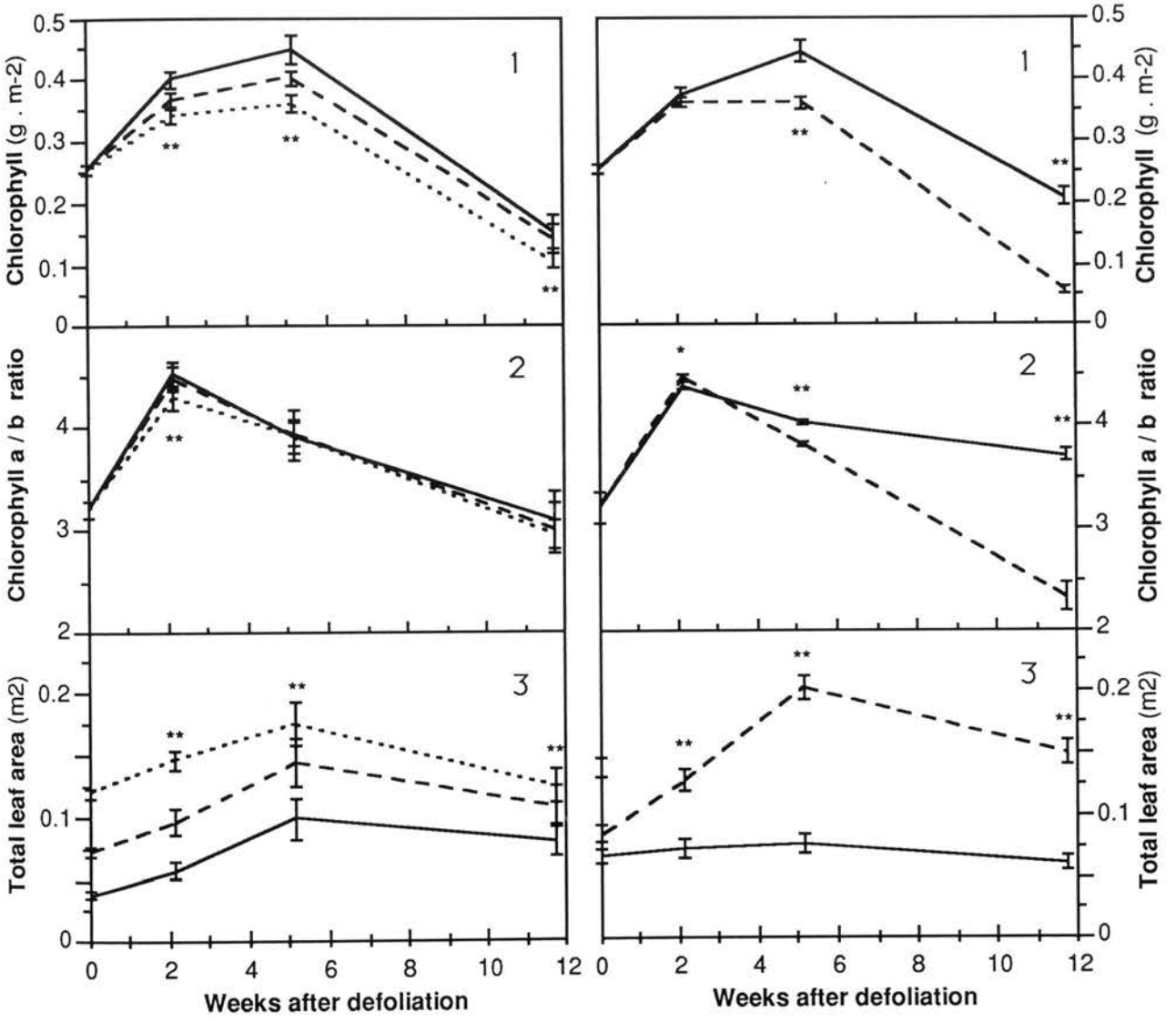

Figure 3: Effect of removing main leaves on young fruitless potted plants on the total chlorophyll content (1) and chlorophyll a/b ratio (2) of the 11th main leaf (from the base), and total leaf area of the plant (3). Full line: plants with 3 main leaves left; dashed line: plants with 6 main leaves left; dotted line: plants with all 12 main leaves left. Vertical bars: standard error; * and **: statistically significant at the 5% and 1% level of probability, respectively.

Figure 4: Effect of removing lateral leaves on young fruitless potted plants on the total chlorophyll content (1) and chlorophyll a/b ratio (2) of the 11th main leaf (from the base), and total leaf area of the plant (3). Full line: treatment LR, plants without lateral shoots; dashed line: treatment LP, plants with lateral shoots. Vertical bars: standard error; * and **: statistically significant at the 5% and 1% level of probability, respectively.

5. Discussion

Source/sink relationship was greatly reduced in defoliated plants, specially in the mature bearing plants that possessed an extra sink: the fruit. There are numerous reports that demand for assimilates by sinks can determine photosynthetic supply (NEALES and INCOLL, 1968; KRIEDEMANN *et al.*, 1976; KRIEDEMANN, 1977; HOFÄCKER, 1978; GIFFORD and EVANS, 1981; KAPS and CAHOON, 1989). During period I the increment of the photosynthetic rate was as more marked as the level of main leaf removal increased suggesting a compensatory response to defoliation (Fig 1: A.2 and B.2).

The leaf used for measurements throughout the season was one of the youngest main leaves of the canopy by the time of the defoliation treatment, but had already reached 85% of its final size. When the defoliation treatments were accomplished these upper leaves, being close to the apex, were exporting assimilates to the growing shoot tip (HALE and WEAVER, 1962; KOBLET, 1969; QUINLAN and WEAVER, 1970). The removal of the shoot tip represented the elimination of an important sink. This explains the different assimilation patterns observed immediately after the treatment on plants with reduced number of main leaves and control plants. For the former, it meant a lesser decrease of the source to sink relationship since there was not only elimination of mature leaves (source) but also the growing leaves (sink). In consequence there was an increment of A. For the latter, it represented an increment on the source/sink ratio and lead to a slight reduction of A. The effect of topping on the young potted fruitless grapevines (Fig. 1: B.2) was much more pronounced. It caused a 50% decrease of the assimilating rate of treatment 12 and a decrease of approximately 10% of treatment 6. This shows the comparatively greater importance of the vegetative shoot tip as a sink in the carbohydrate budget of plants bearing no fruit. This is in agreement with the data obtained by CHAVES (1984). It was also surprising that the removal of 50% of the source tissue in treatment 6 did not give rise to increased A as had happened with the mature fruit bearing plants. There was apparently no decrease on the source/sink ratio since the removal of the 6 basal leaves was counterbalanced by the removal of the shoot-tip sink. This highlights again the strength of the shoot tip as a sink in these vegetatively growing plants.

The general increase of the CO_2 uptake rate registered during the first 3 to 5 weeks after defoliation might be the response to increased sink size: during this period there was a rapid increment in dry weight of roots, trunk, main and lateral

shoots (results not shown). This rapid growth slowed down after the 5th week and simultaneously the photosynthetic rate decreased.

The decrease in the CO_2 uptake rate of the measured main leaves during the period post-veraison indicates that these leaves were already undergoing senescence. Younger leaves must then replace these older leaves to assure fruit maturation and replenishment of the parent vine reserves. During fruit ripening, lateral leaves are most probably the largest contributors to canopy photosynthesis. SCHULTZ (1989) showed that lateral leaves have higher rates of photosynthesis than main leaves during the period from veraison to fruit maturity which gives support to our hypothesis. CANDOLFI-VASCONCELOS and KOBLET (1990) arrived to the conclusion that lateral leaves play the main role during the ripening phase. Moreover, KRIEDEMANN (1968 and 1977), KRIEDEMANN *et al.* (1970), ALLEWELDT *et al.* (1982) agreed that recently formed leaves are photosynthetically more active than older leaves in the vine. Lateral leaves are definitely the youngest of the canopy during fruit maturation.

With the beginning of the lateral shoots growth new vegetative sinks appear and shift the assimilating rates to higher values (Figs. 1 and 2). Young potted plants and mature bearing plants showed an increment of the lateral shoot production as the main leaf area decreased (data not shown). In previous studies we observed the same behavior on completely defoliated plants (CANDOLFI-VASCONCELOS and KOBLET, 1990). An interesting feature of defoliated plants was that they had a higher rate of organogenesis. In fact, on the plants where lateral shoots were removed periodically, plants with fewer main leaves always yielded higher dry masses of lateral shoots between two consecutive removals (data not shown). Plants whose lateral shoots were periodically removed were always in a less favorable situation: they were repeatedly investing nutrients in the production of lateral shoots that would never contribute to the canopy's photosynthesis. According to HALE and WEAVER (1962), lateral shoots are no longer sinks as soon as they have 2 mature leaves. KOBLET (1969) states that lateral leaves become exporters of assimilates when they reach 75% of their final size. They export assimilates not only to their own apex in support of their own growth but also to the main shoot. Plants whose lateral shoots were allowed to grow, are therefore correctly investing assimilates to their own benefit. From veraison to fruit ripening, plants without lateral leaves maintained higher rates of photosynthesis because they could not count on the laterals' contribution.

Defoliated plants showed compensatory responses to defoliation stress. Observed compensatory mechanisms were the increase of chlorophyll content,

the increase of stomatal and mesophyll conductance which allowed an increase of the photosynthetic activity.

The CO_2 influx to the reaction sites inside the chloroplasts is controlled by conductances to CO_2 transfer in the gaseous and liquid phase (SESTAK, 1981). Stomatal movements allow the entry of the CO_2 needed for the photosynthesis into the intercellular spaces. Defoliated plants showed higher stomatal conductance which confirms the results obtained by HOFÄCKER (1978) and HUNTER and VISSER (1988). The dependence of the CO_2 assimilation on the stomatal control was found to be much more important in young potted plants than in mature plants. In fact, young plants showed a strong parallelism between the photosynthesis and stomatal conductance curves (Figs. 1 and 2). However, it has to be stated that stomatal control was not dominant for either of the two plant systems. This can be concluded from the fact that the intercellular CO_2 partial pressure varied in opposite direction as did photosynthetic rate. FARQUHAR and SHARKEY (1982) state that a change in C_i in the same direction as a change in A is a necessary condition to establish primacy of the stomatal response. If the changes are opposite, the most important change must have been in the mesophyll cells.

Mesophyll conductance has two components (see material and methods): conductance to CO_2 in the liquid phase from the intracellular spaces to the carboxylating enzyme (Ribulose bisphosphat carboxylase) and the activity of this enzyme and subsequent chemical processes (RAVEN and GLIDWELL, 1981). The first component is negligible (BJÖRKMAN, 1981, FARQUHAR and VON CAEMMERER, 1982) at least in the temperature range we registered during our measurements (MÄCHLER *et al.*, 1990). Much evidence indicates that mesophyll conductance is mainly limited by the biochemical activity of the enzymes (RAVEN and GLIDWELL, 1981; FARQUHAR and VON CAEMMERER, 1982). The main component of the mesophyll conductance is therefore the carboxylating efficiency. Both mature fruiting plants and young fruitless plants showed a high degree of mesophyll control over the photosynthesis, as judged by the similarity of the curves of these parameters. Increase of the photosynthetic activity as response to defoliation was achieved by enhancing the carboxylating efficiency and increasing stomatal conductance. These results are consistent with those presented by KRIEDEMANN in 1977.

Stomata serve to balance the need for the leaf to allow the entry of CO_2 for photosynthesis whilst limiting the transpiratory loss of water vapor (COWAN, 1982). A measure of the carbon gain in relation to the water loss is the water use

efficiency. Increase on the photosynthetic rate as response to defoliation could only be achieved with an increment of the transpiration rate. Mature plants, except for the first 3 weeks following treatment, showed no response either to main leaves' or lateral shoots' removal. In contrast, young potted plants either with no laterals or with reduced number of main leaves, showed always higher rates of transpiration than the LP plants after treatment. Nevertheless, increase of the rate of carbon gain was almost always higher than on that of water loss, resulting in increased water use efficiency. This was particularly evident on mature and young plants without lateral shoots during period II (Fig.1: B.5 and Fig. 2: B.5). Young potted plants with reduced main leaf area showed also a higher WUE during period I (Fig. 1: B.5). HUNTER and VISSER (1988) also reported increased transpiration and water use efficiency with increased level of defoliation.

The use of potted plants as simpler approach to test gas exchange response to defoliation stress cannot be considered an unrealistic design. In fact, we obtained very similar treatment responses in both mature bearing grapevines and young potted plants. The differences between these two plant systems were associated with a higher sensitivity of the leaf stomata of potted plants. This could be due to the restricted soil volume available to root growth which could limit water absorption and affect water relations.

Total chlorophyll content increased with increasing level of defoliation which was another compensatory mechanism to defoliation. Similar results are reported by HOFÄCKER (1978), and HUNTER and VISSER (1989). Chlorophyll a and b contents were higher in plants with fewer main leaves and in plants without lateral shoots. Chlorophyll a/b ratio increased during the first 2 weeks after defoliation because chlorophyll a showed the steepest increment during this initial period and chlorophyll b exhibited a delayed response, showing the highest increment rate between the 2nd and 5th week post defoliation. Plants bearing no lateral shoots showed higher chlorophyll a/b ratios, particularly after midseason, indicating that even if they possessed more chlorophyll a and b than LP plants they were not utilizing their light harvesting pigments as efficiently as plants with lateral shoots. Lower chlorophyll a/b reflects, according to BJÖRKMAN (1981) increased proportion of the light-harvesting Chlorophyll-ab-protein (LHchl) complex to the total chlorophyll complement of the chloroplast. The LHchl complex contains all the chlorophyll b and is primarily associated with photosystem II (PS II) and therefore, lower chlorophyll a/b also reflects higher PS II / PS I ratio. Curiously, the increments observed in chlorophyll content led to increased chlorophyll a/b ratio indicating that higher efficiency of light capturing is needed when the chlorophyll

level is low. Plants with reduced leaf area showed a slower rate of chlorophyll degradation and a delay in the rate of leaf abscission during period II, indicating a delay of leaf senescence which confirms the results obtained in our previous experiments (CANDOLFI-VASCONCELOS and KOBLET, 1990).

Compensation to a decrease of the main leaf area was only observed during the period pre-veraison. However, the largest differences in the assimilation rates observed in the mature fruiting plants between defoliated and control plants did not exceed 30% for treatment 3 and 14% for treatment 6, respectively. Young potted plants having 3 and 6 main leaves left, attained photosynthetic rates 66 and 42% higher than the control, respectively. However, these increments were not high enough to enable full compensation because the assimilating leaf area was reduced up to 70%.

Compensation to removal of lateral leaves became more pronounced during fruit ripening. During period II, fruit-bearing and fruitless plants with no laterals had 20 to 45% higher photosynthetic rates (Fig. 2: A.2 and B.2) and managed to keep up to 260% higher levels of chlorophyll (Fig. 4: 1) as compared to the control. The fact that these differences are more pronounced during period II, when the photosynthetic rate and chlorophyll content already started to decline, indicates that main leaves are able to delay senescence if they do not have the support of the lateral leaves, whose photosynthetic performance is reported to be higher than that of main leaves during the ripening phase (SCHULTZ, 1989).

6. Conclusions

1. Compensation by increasing the photosynthetic performance as a response to main leaves removal was only partial and confined to the pre-veraison period.

2. In contrast, removal of lateral shoots, resulted in the maintenance of higher assimilation rates on the remaining main leaves towards the end of the season.

3. Main leaves' photosynthesis had a limited importance during fruit maturation. Most likely the lateral leaves assumed the primary role.

4. The observed compensation mechanisms related to leaf removal were:

- Increase of the photosynthetic rate
    - Increase of the mesophyll conductance
    - Increase of the stomatal conductance
- Increase of the water use efficiency
- Increase of the chlorophyll content
- Delay of the leaf senescence and abscission

5. Increase of the photosynthetic activity as response to defoliation was achieved mainly by enhancing the mesophyll conductance.

6. Young potted plants and mature field grown plants showed very similar responses to defoliation treatments.

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1979-1984	Student at the Faculty of Agriculture at the Technical University of Lisbon.
1984-1985	Practical training in Viticulture, Enology and Plant-protection at the Swiss Federal Research Station for Fruit-growing, Viticulture and Horticulture in Wädenswil, Switzerland.
1985	Marriage to Marco Candolfi (Swiss citizen).
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