

Girdling of shoots at flowering reduces shatter in grapevine cv. Malbec

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Abstract

Background and Aims: Some grapevine cultivars such as Malbec have unstable yield, due to poor fruitset or fruitlet abscission. The phenomenon is known as 'shatter' and this study aims to explore the potential of applying the shoot girdling technique at flowering, to direct the carbohydrate partitioning towards inflorescences and reduce shatter.

Methods and Results: Fruitful Malbec shoots were girdled above the apical bunch, below the basal bunch, double girdled or not girdled (Control) during 2017 and 2018. Most vegetative growth parameters were unaffected, but shoots widened and the leaf expansion improved in the fruit zone. Fruitset doubled compared to that of the Control, ovary abortion was reduced, and fruit yield increased, mainly due to a greater number of small berries in the base and double girdled treatments. In addition, base girdling increased TSS per berry, reducing the concentration of phenolic substances in the normal size berries. Girdling effects were consistent during both seasons.

Conclusions: Shoot girdling below the basal bunch proved to be effective in reducing shatter. The increase in carbohydrate supply available to the inflorescences during flowering was able to augment their weak sink strength relative to the perennial organs.

Significance of the Study: The study contributes to an understanding of the importance of the photoassimilates flow towards inflorescences in reducing shatter and increasing yield in Malbec vines.

Keywords: fruitset, girdling, shatter, *Vitis vinifera* L., yield

Introduction

In *Vitis vinifera* L. the berries in a bunch are determined by the number of flowers per inflorescence, but also by fruitset (the proportion of flowers in an inflorescence that turn into berries) and berry abscission. Certain grape cultivars have relatively few berries per bunch due to a genetic predisposition to produce inflorescences with a small number of flowers, for example Tempranillo, Sauvignon Blanc, Pinot Noir and Chardonnay (Dry et al. 2010). Other cultivars can exhibit poor fruitset, which limits yield and vineyard productivity (May 2004). This is a common problem, colloquially known as 'shatter' and can occur in cultivars, such as Merlot, Grenache, Gewürztraminer and especially Malbec, which can be unstable in its productive potential. For example, in Mendoza, the main wine region in Argentina, with 86% of the total Malbec cultivated area (34 672 ha), based on 20 years of record (1996 to 2016) the average fruit yield ranged from a minimum of 3.4 to a maximum of 10 T/ha with a variation coefficient of 20% (Instituto Nacional de Vitivinicultura 2017).

The number of flowers in grapevines is determined over two growing seasons at different stages; that is during the initiation of the inflorescences before the end of the growing season and during budburst in the following season (Dunn and Martin 2007). Vascular plants commonly initiate more reproductive meristems than those that develop and mature in a given season (Keller et al. 2001). Accordingly, in grapevine a proportion of flowers abscise from the inflorescence before turning into berries (Collins and Dry 2009), possibly as a mechanism for adjusting reproductive output to the level of available

resources. Fruitset is considered 'normal' when higher than 50% and 'poor' when lower than 30% (Bessis 1993).

The causes of shatter are perhaps multiple, concomitant and, until now, relatively unclear, being usually associated to plant material (different cultivars, clones or rootstocks/scions), excessive vigour (May 2004), insufficient supply of carbohydrates (Candolfi-Vasconcelos and Koblet 1990), nitrogen deficiency (Ewart and Kliewer 1977) and also to unfavourable meteorological events during flowering such as low temperature or rainfall (Keller and Koblet 1994). Root and shoot apices are the major sinks during vegetative growth, while fruits become the dominant sinks during reproductive development. Often during flowering, however, inflorescences represent weak sinks for carbohydrates in relation to the rapidly extending shoot tips (Coombe 1959) and hence limitations in photoassimilates supply may cause abortion of flowers (Ruan et al. 2012). In addition, if resources are scarce fruitset decreases (Caspari et al. 1998, Lebon et al. 2008) and a major proportion of fruitlets abscise from bunches (Bessis et al. 2000).

A range of management strategies has been proposed to reduce the incidence of shatter in established vineyards, mainly by increasing the availability of carbohydrates to the inflorescences by using practices such as girdling (Caspari et al. 1998). Other cultural practices to improve fruitset include shoot tipping (Vasconcelos and Castagnoli 2000) and the application of growth regulators (Collins and Dry 2009). These approaches, however, have not yet been sufficiently evaluated under agroecological conditions of Malbec

vineyards planted in Mendoza. A significant proportion of the Malbec planted in the area suffers severe shatter problems.

Sucrose, amino acids, phytohormones and some inorganic ions are transported in phloem flow from sources to sinks of metabolism and storage, following anatomic patterns and physiological mechanisms (Murcia et al. 2016). Trunk or cane girdling consists of the removal of a ring of bark (phloem) to restrict the sap flow without impairing xylem function (water status) and can be used as a strategy to redirect photo-assimilates between plant organs (Glad et al. 1992). The interruption of phloem is temporary and most girdled vines formed a callus across the girdle within five weeks (Williams et al. 2000) so restoring the vessels functionality (Lang and Thorpe 1989). Girdling has proven to affect fruitset, bunch architecture and berry sugar accumulation, increasing fruit yield in Sauvignon Blanc, Pinot Noir and the tablegrape Italia (Coombe 1959, Brown et al. 1988, Caspari et al. 1998, Ferrara et al. 2014). Nonetheless, it is difficult to generalise about shatter since the reports differ in the cultivars studied, the girdling location and the phenological timing of the treatment. Hence the present study was undertaken to examine the effect of girdling shoots at the beginning of flowering in *V. vinifera* L. cv. Malbec on the reduction of shatter. The hypothesis is that the increase in the availability of carbohydrates for inflorescences at flowering improves fruitset, reduces berry abscission and increases bunch size and yield.

Materials and methods

Plant material and experimental design

The trial was undertaken in a commercial vineyard of *V. vinifera* L. cv. Malbec, located in Valle de Uco, in the piedmont region of Mendoza, Argentina (33°26'S, 69°13'W and 1205 m asl) during the 2016/17 and 2017/18 seasons. Vines were planted in 2001, grafted onto 1103 Paulsen rootstock, trained in a bilateral cordon (1.3 m between vines N-S orientated rows and 2.5 m between rows), protected with anti-hail nets (black polyethylene) and drip irrigated to soil field capacity. A zone of homogeneous vine size was selected

within a parcel of 8 ha based on a NDVI image and trunk diameter measurements (data not shown). Then, 23 vines in 2017 and 14 vines in 2018 were selected, choosing each one as representative from five consecutive vines in a row and based on trunk diameter measurement (different plants were used between seasons). Vines were spur-pruned during winter dormancy to retain eight nodes per arm (16 nodes per plant) and shoot-thinned to 16 fruitful shoots. At the beginning of flowering (first flower caps loosening) in mid-November [modified Eichhorn and Lorenz stage 19 (Coombe 1995)], four girdling treatments were randomly applied to four shoots per plant arm (experimental unit $n = 46$ for 2017 and $n = 28$ for 2018): girdling the internode above the apical bunches (Top G), girdling the basal shoot internode below bunches (Base G), girdling at the base and above bunches (Double G) and without girdling (Control). All girdles were made at the mid-point of the internode (Figure 1). Shoots were girdled with a tool with two blades separated 3 mm apart to completely sever the phloem down to the xylem, around the whole circumference of the shoot internodes. In 2017, every 20 days and until veraison, any callus formed was removed with tweezers, while in 2018 no callus was removed after girdling at flowering.

Fruitset, vegetative growth, fruit yield and measurement of fruit composition

The number of flower caps and aborted ovaries was assessed as described by Keller et al. (2010), placing gauze bags over all the basal inflorescences from the beginning of flowering up to 1 month after fruitset and counting the collected caps and ovaries. The percentage of fruitset (proportion of flowers that set a berry) was calculated by counting the final number of berries at harvest.

Shoot length, number of leaves, leaf area (LA) and total shoot LA were determined in all the treated shoots at the onset of veraison (berries begin to colour and soften), in mid-January [modified Eichhorn and Lorenz stage 35 (Coombe 1995)]. Leaf area was non-destructively estimated by measuring the leaf central vein length in the treated shoots and then using a linear regression model between LA and

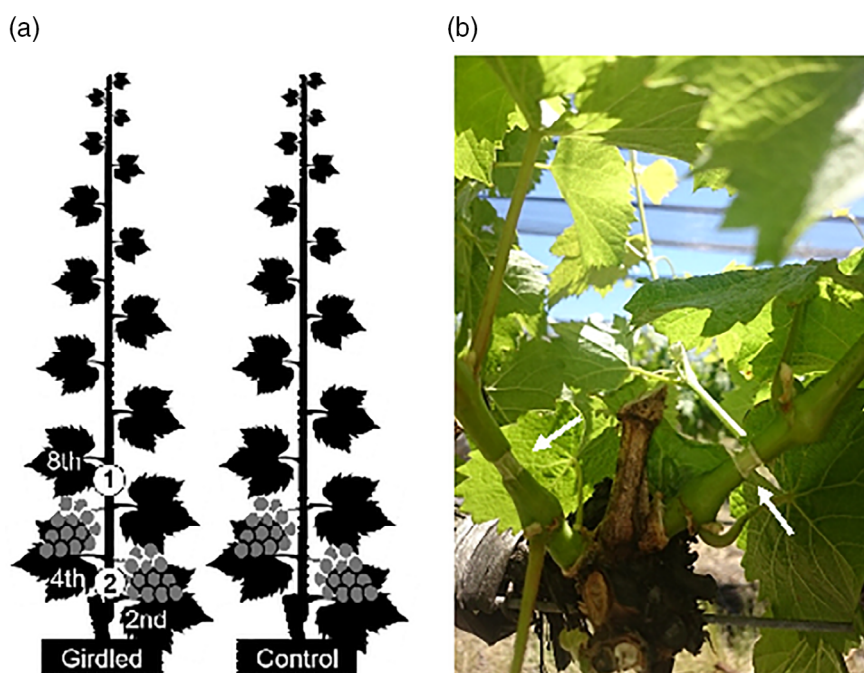


Figure 1. (a) Schematic representation of (1) Top G, girdling the internode above the apical bunches; (2) Base G, girdling the basal shoot internode below bunches; (1,2) Double G, girdling at the base and above bunches; and Control shoots, with the sampled basal leaf position; and (b) representative Base G treated shoot at the beginning of flowering.

vein length, generated with all the leaves of shoots from adjacent plants, as described by Berli et al. (2013). Relative chlorophyll content (CHL) was assessed in the second, fourth and eighth leaf from the base of the shoot (represented in Figure 1a), with a SPAD 502 chlorophyll metre (Konica Minolta, Osaka, Japan). During winter dormancy, the diameter of shoot nodes at the widest point in basal, middle and apical sections, matching the second, fourth and eighth leaf, was determined with a digital calliper.

Fruit was harvested when the TSS of Control berries reached 22°Brix as measured with a Pocket PAL-1 digital hand-held refractometer (Atago, Tokyo, Japan), shortly before the commercial harvest date, in early March for both seasons. Bunches were weighed fresh (FM) and the berries within a bunch were removed from the rachis and sieved. Berries were classified as normal (≥ 12 mm diameter) and small (< 12 mm diameter). For each group, total berries FM, number of berries and berry FM were recorded and the seeds per berry, seed dry mass (DM) (dried at 80°C to constant mass) and seedless berries (parthenocarpic) were recorded in subsamples containing three randomly selected berries per experimental unit. In addition, subsamples of 15 normal berries and 15 small berries were used to evaluate TSS and the concentration (per skin DM) of phenolic substances (including anthocyanin), as described in Berli et al. (2008).

Meteorological data

Temperature, RH and rainfall were recorded during the 2016/17 and 2017/18 growing seasons, between September

and May (from grapevine budburst to leaf fall), with an automatic weather station located in Valle de Uco (33°20'S, 69°9'W and 1074 m asl), 12 km away from the experimental site (www.contingencias.mendoza.gov.ar). Data were collected every 60 min for 24 h a day and the daily mean temperature and RH were calculated. The maximum and minimum temperature and rainfall were also recorded. The RH and the temperature of the air in the fruit zone were measured every 60 min (from 14 October 2017 to 25 January 2018) with hygrochron loggers (iButton DS1923, Maxim Integrated Products, San Jose, CA, USA), located and shielded as described by King et al. (2014).

Statistical analysis

InfoStat-Statistical Software, version 2017 (Universidad Nacional de Córdoba, Córdoba, Argentina) was used for statistical analysis. The effects of girdling treatments, growing season, berry size and their interaction were determined by multifactorial ANOVA and Fisher's LSD ($P \leq 0.05$). The effect of each girdling treatment was evaluated against the Control, by one-way ANOVA and Dunnett's multiple comparisons test.

Results

The number of flowers ranged between 110 and 130 flowers per inflorescence and was unaffected by girdling (Figure 2a), although the proportion of flowers that set fruit was significantly increased in the Base G and Double G treatments, more than twofold that of the Control (Figure 2b). In addition, Base G and Double G markedly reduced the number of aborted ovaries (Figure 2c) and increased bunch FM (67 and 43%, respectively) as compared to that of the Control (Figure 2d). Top G was not significantly different to the Control in any of these variables (Figure 2b–d).

The number of berries per bunch, both normal and those of small size, did not differ between the two seasons, while the FM of normal berries was higher in 2018 (Table 1). The seed number per berry and seed DM were higher in 2017 for small berries, with a lower proportion of parthenocarpic. Normal berries did not show differences for these parameters between 2017 and 2018. Yield characteristics were markedly affected by girdling treatments, that is bunches from Base G and Double G had 127 and 109% more berries at harvest, respectively, than those from Control, especially of small berries. Base G and Double G increased the number of small berries in bunches about eightfold as compared to Control (37 small berries vs 4.5 small berries); reduced the number of seeds in small berries and increased the proportion of parthenocarpic (significant interaction effect of treatments and berry size). The berry FM was 69% lower in small berries than in normal berries, while the number of seeds per berry and seed DM varied by about 50%. The number and FM of normal berries were not affected by girdling; the number of seeds per berry was higher in berries from Double G as compared to Control, whereas the seed DM was lower.

The diameter of the shoots was affected only by the Base G treatment which increased the diameter of the shoot middle section (Figure 3). Most of the vegetative growth variables and CHL measured at the onset of veraison tended to be higher in 2018 than in 2017, with significant differences for shoot length, total shoot LA and LA of the second and fourth leaves (Table 2). The shoot length, number of leaves per shoot and total shoot LA were not affected by girdling,

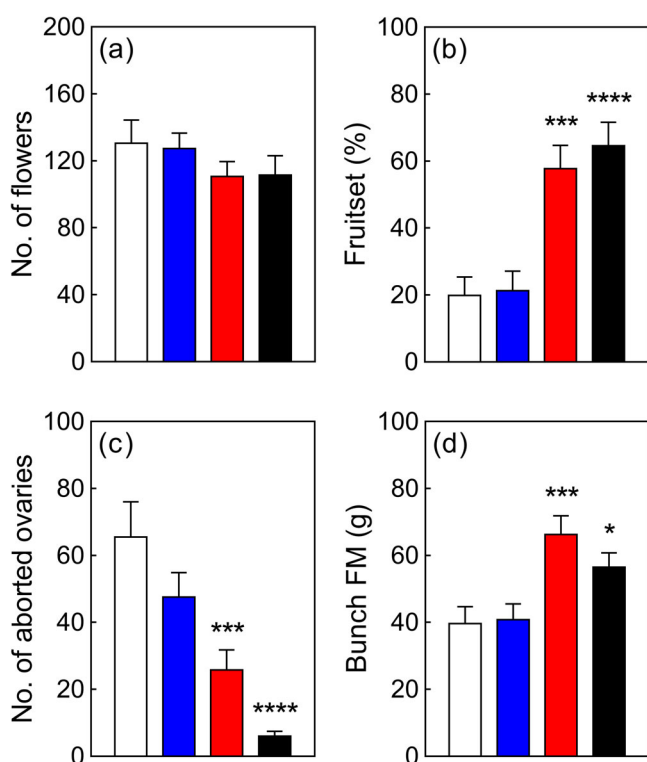


Figure 2. Effect of girdling in 2018 on the (a) number of flowers per bunch, (b) proportion of fruitset, (c) aborted ovaries and (d) bunch fresh mass (FM). Values are means \pm SEM and each treatment was compared with the Control using Dunnett's multiple comparisons test where *, $P \leq 0.0332$, ***, $P \leq 0.0002$ and ****, $P \leq 0.0001$. Control, without girdling (□); Top G, girdling the internode above the apical bunch (■); Base G, girdling the basal shoot internode below bunches (■); Double G, girdling at the base and above bunches (■).

Table 1. Effect of girdling treatments on the yield components of Malbec at harvest during the 2016/17 and 2017/18 seasons.

	No. berries/ bunch	Berry FM (g)	No. seeds/ berry	Seed DM (mg)	Proportion of parthenocarp (%)
Season					
Normal berries					
2016/17	23.78a	1.44b	1.29a	37.79a	0.67c
2017/18	19.52a	1.87a	1.17a	33.60a	3.21cb
Small berries†					
2016/17	19.27a	0.48c	0.67b	22.33b	36.54b
2017/18	22.26a	0.53c	0.48c	14.48c	51.58a
Girdling treatment					
Normal berries					
Control	21.90b	1.68a	1.17b	40.80a	0.64d
Top G	23.28b	1.67a	1.14b	38.94ab	3.85d
Base G	22.97b	1.66a	1.30ab	34.59bc	2.08d
Double G	18.47b	1.61a	1.33a	28.45c	1.19d
Small berries					
Control	4.58c	0.54ab	0.74c	20.40d	22.63c
Top G	4.54c	0.54ab	0.64cd	18.69d	37.25b
Base G	37.03a	0.48ab	0.42e	18.44d	53.85a
Double G	36.92a	0.45b	0.50de	16.09d	62.50a
ANOVA					
$P_{(Season)}$	0.6935	0.7745	0.0023	0.0007	0.0063
$P_{(Treatment)}$	<0.0001	0.0001	0.5987	0.0052	<0.0001
$P_{(Berry\ size)}$	0.5840	<0.0001	<0.0001	<0.0001	<0.0001
$P_{(Treatment \times Berry\ size)}$	<0.0001	<0.0001	0.0020	0.3013	0.0001

Values are means and different letters within each factor and column indicate a statistically significant difference (Fisher's LSD, $P \leq 0.05$). †Berries less than 12 mm diameter. Base G, girdling the basal shoot internode below bunches; Control, without girdling; DM, dry mass; Double G, girdling at the base and above bunches; FM, Fresh mass; Top G, girdling the internode above the apical bunches.

but LA was increased in the sampled leaves (second, fourth and eighth) with a major effect for Base G. For example, LA in the fourth leaf was 28% higher in Base G than in Control. Chlorophyll was not affected, however, by the girdling, except for the eighth leaf of Double G shoots, which was higher than in Control (Table 2).

While there was no difference in TSS per berry between the growing seasons, TSS was greater in 2017 (Table 3) and phenolic substances increased in 2018. Most of the compositional differences were berry size-dependent with significant interaction effects. In normal berries, Base G reduced phenolic substances and increased TSS per berry, without significantly affecting TSS. In small berries, all the treatments increased TSS, without affecting TSS per berry. The

concentration of anthocyanin, however, was reduced by Base G (Table 3).

Based on the meteorological data, the 2016/17 growing season was characterised by a spring with lower temperature and a wetter summer as compared to 2017/18 (Table S1). In addition, with more detailed data of temperature and RH in the fruit zone for 2017/18, no adverse weather conditions were recorded, especially during the period of flowering to fruitset (Figure S1).

Discussion

An improved understanding of the factors that influence berry development may enable enhanced management of vineyards in terms of yield and fruit composition. The

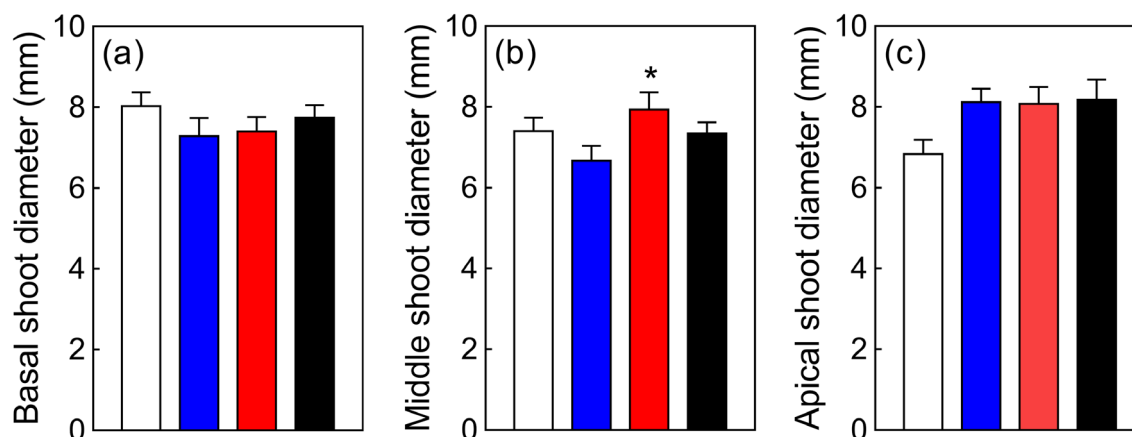


Figure 3. Effect of girdling on the (a) shoot node diameter at mid-point in basal, (b) middle and (c) apical sections at harvest in 2018. Values are means \pm SEM and statistically significant differences were analysed with Dunnett's multiple comparisons test, comparing each treatment with the Control (*, $P \leq 0.0332$). Control, without girdling (\square); Top G, girdling the internode above the apical bunch (\blacksquare); Base G, girdling the basal shoot internode below bunches (\blacksquare); Double G, girdling at the base and above bunches (\blacksquare).

Table 2. Effect of girdling treatments on the vegetative growth variables of Malbec shoots at the onset of veraison, during the 2016/17 and 2017/18 seasons.

	Shoot length (cm)	No. leaves/shoot	Total shoot leaf area (cm ²)	Leaf area (cm ²)			Relative chlorophyll concentration (%)		
				Second†	Fourth	Eighth	Second	Fourth	Eighth
Season									
2016/17	147.39b	28.56a	2360.48b	21.43b	86.96b	134.98a	28.16a	29.86a	29.32a
2017/18	190.25a	29.67a	2734.84a	45.71a	133.56a	133.42a	29.69a	30.22a	30.95a
Girdling treatment									
Control	164.90a	28.96a	2549.93a	22.65b	94.31b	124.15b	29.77a	28.71a	28.25b
Top G	159.00a	29.83a	2512.88a	34.81ab	114.70a	131.96ab	30.00a	31.14a	30.66ab
Base G	168.93a	28.67a	2469.63a	39.33a	121.07a	142.01a	27.07a	31.23a	29.08b
Double G	182.44a	29.00a	2658.19a	37.50a	110.94ab	138.68a	28.80a	29.06a	32.55a
ANOVA									
<i>P</i> _(Season)	0.0045	0.5530	0.0378	0.0001	<0.0001	0.7662	0.6454	0.8056	0.1782
<i>P</i> _(Treatment)	0.7164	0.9754	0.8904	0.1497	0.1094	0.0742	0.7992	0.4675	0.0700

Values are means and different letters within each factor and column indicate a statistically significant difference (Fisher's LSD, $P \leq 0.05$). †Basal leaf position. Base G, girdling the basal shoot internode below bunches; Control, without girdling; Double G, girdling at the base and above bunches; Top G, girdling the internode above the apical bunches.

different girdling treatments were used to modify the supply of carbohydrates to the developing inflorescence. Photoassimilates from basal leaves are generally transported basipetally, while those from mid leaves flow both acropetally and basipetally (Hunter and Visser 1988) and these directions depend on the relative sink strength (Hale and Weaver 1962). The Base G treatment was intended to direct the photoassimilates towards the inflorescence by excluding trunk, cordons and roots as sinks, while the Top G and Double G aimed to reduce photoassimilates supply by eliminating the upper leaves as a source. The double G treatment evaluated the contribution of a limited supply of photoassimilates from two to three leaves to developing fruits.

The Base G treatment increased the cross section of the shoot above the girdles and possibly the distribution of the

grapevine photoassimilates in the fruit zone. Initial grapevine shoot development in the spring depends on the carbohydrate stored in roots, trunk and canes (Zapata et al. 2004). Then, the leaves start to export assimilates; initially directed towards the shoot tip, but later to the shoot base and the permanent structure of the vine (Koblet 1969). Girdling at flowering did not affect most of the growth variables measured at the onset of veraison, but increased LA of the mature basal leaves, mainly in Base G treatment. Reducing sinks improved expansion of the basal leaves without affecting the number of leaves per shoot and total shoot LA (the latter depends on leaf size and number of leaves per shoot). The lack of effect on the elongation of the shoot and development of new leaves may be explained by the girdling treatments conducted at flowering, when a greater

Table 3. Effect of girdling treatments on TSS and skin phenolic substances at harvest in Malbec berries during the 2016/17 and 2017/18 seasons.

	TSS (°Brix)	TSS (mg/berry)	Anthocyanin (A ₅₂₀ /g skin DM)	Phenolic substances (A ₂₈₀ /g skin DM)
Season				
Normal berries				
2016/17	21.85a	0.38a	17.70c	12.92c
2017/18	20.12ab	0.37a	21.68b	31.96b
Small berries†				
2016/17	20.76b	0.09b	39.56a	28.07b
2017/18	19.73c	0.11b	40.08a	61.73a
Girdling treatment				
Normal berries				
Control	21.23a	0.35b	25.91c	27.34c
Top G	21.14a	0.37ab	21.22cd	23.11cd
Base G	20.32ab	0.39a	13.41e	17.88d
Double G	20.93a	0.37ab	18.22ed	21.43cd
Small berries				
Control	19.08b	0.10c	45.11a	48.63a
Top G	20.68a	0.11c	38.59ab	40.02b
Base G	20.80a	0.11c	37.46b	44.34ab
Double G	20.10a	0.10c	41.24ab	47.32a
ANOVA				
<i>P</i> _(Season)	0.0001	0.7745	0.1311	<0.0001
<i>P</i> _(Treatment)	0.2553	0.0001	0.0003	0.0542
<i>P</i> _(Berry size)	0.0347	<0.0001	<0.0001	<0.0001
<i>P</i> _(Treatment×Berry size)	0.0835	<0.0001	0.0848	0.5780

Values are means and different letters within each factor and column indicate a statistically significant difference (Fisher's LSD, $P \leq 0.05$). †Berries smaller than 12 mm diameter. A, absorbance; Base G, girdling the basal shoot internode below bunches; Control, without girdling; DM, dry mass; Double G, girdling at the base and above bunches; Top G, girdling the internode above the apical bunch girdling.

proportion of the vegetative growth had already occurred. Unlike extension growth, dry matter of leaves and shoots and hence leaf and internode mass, continues to increase during the growing season (Poni et al. 1994, Cartechini and Palliotti 1995). The improvement of basal LA by girdling treatments at full flowering was unexpected and may suggest that the leaves were not fully developed when this treatment was imposed. Caspari et al. (1998) also found that basal stem-girdles (treatment that impairs phloem function) applied at flowering did not affect shoot length and total shoot DM in Sauvignon Blanc.

Budburst in the grapevine is followed by a period of rapid shoot growth with the appearance of a new leaf every few days. As the season progresses, growth rate of the vegetative tissues decreases from flowering, once the main shoot has about 18–20 leaves (Borghazan et al. 2012), when cambium cell division ceases and shoots begin to form brown periderm and store reserves [reviewed by Keller (2015)]. Chlorophyll concentration in mature leaves was not influenced by the girdling treatments indicating that no premature senescence occurred (Petrie et al. 2000). Murakami et al. (2008) found that stem girdling in sugar maple increased sucrose, glucose and fructose concentration and enhanced anthocyanin concentration in leaves. In addition, Soar et al. (2004) observed that grapevine leaves accumulated abscisic acid (ABA) above the girdle within 48 h, suggesting that ABA, normally exported from the apical tissues, was blocked and ABA biosynthesis below the girdling was induced. Thereby, increased ABA was expected to promote leaf senescence as has been previously observed in young *Pinus canariensis* seedlings (López et al. 2015), which did not occur in this study.

In other studies, phloem disruption below the inflorescences increased berry set and prevented premature berry abortion, possibly by enhancing the relative sink strength of the flowers/fruits as the other sinks had been removed, through an increase of the availability of carbohydrates (Coombe 1959) and/or by changes in the hormone balance (Kriedemann and Lenz 1972). The concentration of grapevine carbohydrates and stem diameter increased above the girdle (Kriedemann and Lenz 1972) and an increase in the concentration of sugar in the bunches was detected (Coombe 1959). In the present study the fruit yield components were markedly increased in base and double girdled treatments, with bunches characterised by a large number of small berries (both seeded and parthenocarpic), that ripened normally. We have not distinguished between seeded berries and seedless berries to calculate fruitset, but we excluded the 'live green ovaries' (LGOs) to avoid an overestimation of berry number. The increased fruitset and lower mean berry size in girdled shoots is consistent with the literature, as girdling promotes the retention of small-size parthenocarpic berries and also small berries with a reduced number of seeds, which usually would abscise (Coombe 1959, Brown et al. 1988).

The commercial Malbec vineyard where the research was conducted is prone to severe shatter and low yields. In the two consecutive seasons, however, when the shoots were girdled at the base, the proportion of fruitset changed from 20% in the Control to almost 60% in treated shoots. While shoot girdling below the basal bunch increased fruitset and decreased the number of aborted ovaries, girdling above the apical bunch did not affect them. Candolfi-Vasconcelos et al. (1994) showed that under a restricted

photosynthetic supply such as defoliation, vines responded by altering the natural translocation pattern and redirecting carbon stored in the lower parts to the fruit.

Berry abscission normally begins at flowering and can occur for up to 4 weeks after anthesis depending on the weather conditions (Kassemeyer and Staudt 1983); it is during this period that fruitset is determined. Other authors also found an increase in grapevine yield components due to shoot girdling (Reynolds and de Savigny 2004, Williams and Ayars 2005), which was explained by changes in translocation and distribution of photoassimilates (Harrell and Williams 1987, Roper and Williams 1989, Zabadal 1992, Williams et al. 2000). Therefore, fruit yield may provide an averaged and integrated record of carbohydrate availability over a large period as proposed by Caspari et al. (1998). The results obtained in the double girdled treatment show that with only a few leaves between the girdles, the effect on fruitset and berry abortion is similar to that of the base girdle treatment. This suggests that the demands of the perennial structures as sinks are the major cause of shatter. Double girdling also increased the bunch FM as compared to that of the Control, but to a lesser extent than when the shoot was girdled only at the base. The limited repercussions of the double girdled treatment on fruit yield, that is reducing the number of leaves as sinks, may be related to the capacity of the grapevine to compensate for the loss of leaf area by increasing the leaf efficiency in terms of carbon fixation (Iacono et al. 1995).

Girdling may affect various physicochemical parameters related to fruit composition, such as accumulation of sugars, organic acids and phenolic substances, depending on the phenological timing of the treatment (Basile et al. 2018). Zabadal (1992) found that cane girdling of Himrod grapevines when the berries were 'pepper size' increased the bunch mass and the number of berries per bunch by over 100%, the berry mass by 17% and the yield by 66%, but it consistently reduced the TSS in the fruit. Ferrara et al. (2014) cane girdled the tablegrape cv. Italia at berry set, in a 2-year study and found that it increased yield per vine significantly, but affected berry skin colour negatively. They also showed that the treatment effect on the fruit composition was dependent on the growing season, due to different climatic conditions (temperature and rainfall). Basile et al. (2018) trunk girdled Sugerthirteen tablegrape at berry set and veraison and found that the concentration of phenolic substances and anthocyanin in the berry skin increased only when the treatment was applied at veraison. In addition, the effects are cultivar-dependent as showed by Isci et al. (2015), who found different phenolic responses when girdling three red tablegrape cultivars at veraison.

Our results show that TSS in normal berries was not affected by the girdling treatments, but that it increased in small berries. Base girdling increased sugar per berry in normal berries, but reduced the concentration of phenolic substances including anthocyanin. Treatments negatively affected skin colour, but this aspect could be considered by delaying the harvest date (Kingston and Van Epenhuijsen 1989). In addition, the impact on wine composition is not clear and should be evaluated with a different experimental approach, considering the whole bunches (i.e. maintaining the real berry sizes proportion and skin/pulp ratios).

The reported causes of shatter are multiple, including genetic (plant material) and environmental factors, which may be important for floral development (Carbonneau et al. 2007). In the present work, meteorological differences

between the two growing seasons were recorded (Table S1) producing significant seasonal effects, especially for vegetative growth. The response to girdling, however, was consistent. The results presented show that it was possible to reduce shatter in Malbec by girdling shoots below bunches at flowering. The increase in carbohydrate supply available to the inflorescences during flowering was able to augment their weak sink strength relative to the perennial organs.

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Supporting information

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Table S1. Environmental RH, mean temperature, absolute maximum temperature, absolute minimal temperature and rainfall, during the 2016/17 (2017) and 2017/18 (2018) growing seasons for September to May, Valle de Uco, Argentina.

Figure S1. Daily mean air RH (—) and daily mean (—), maximum (—) and minimum (—) temperature (measured at the fruit zone from budburst to the onset of veraison during the 2017/18 season).