

## Research Article

# Assessment of the Crop Forcing Technique and Irrigation Strategy on the Ripening of Tempranillo Grapes in a Semiarid Climate

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*Background and Aims.* High temperatures during grape ripening have a negative effect on the winemaking characteristics of musts. The crop forcing technique delays ripening to a period when temperatures are lower. The objective of this study is to provide information to winemakers and grape growers on how the delay in ripening caused by crop forcing can affect berry performance. *Methods and Results.* This study of 3 growing seasons (2017–2019) analyzes the effect of this technique in a vineyard of the Vitis vinifera L. cv. Tempranillo in Extremadura, together with two irrigation strategies. The grapevines were forced 4 and 22 days after anthesis (F1 and F2, respectively), compared to a treatment without crop forcing techniques (NF). Each treatment was subjected to two irrigation strategies: to cover the water needs of the plants (C) and deficit irrigation during preveraison (RI). Crop forcing delayed the harvest between 32 and 56 days on average in relation to NF. Crop forcing and irrigation strategy modified berry composition at harvest: C-F1 and C-F2 had higher total polyphenol and anthocyanin concentrations, total acidity, malic acid content, and lower pH relative to C-NF; RI-NF increased total anthocyanin concentration and pH and decreased titratable acidity value. *Conclusions*. Crop forcing is able to delay grape ripening to lower temperature periods. This is a promising technique for restoring the coupling between phenolic and technological ripeness. The combination of both crop forcing and deficit irrigation strategy maintains the berry quality while improving water use efficiency. *Significance of the Study*. The present work shows how the "crop-forcing" technique is effective in modifying the relationship between different parameters that determine the characteristics of berries throughout ripening as a raw material for winemaking, compared to vines with traditional winter pruning, under different irrigation strategies.

To the memory of Jordi Marsal

## 1. Introduction

Climate plays a fundamental role in the development of the vineyard. The temperature regime, the availability of water, and the intensity of radiation determine plant growth, production, and harvest quality [1, 2]. Each grapevine variety has its optimum development in a specific range of temperatures. Small temperature variations can modify productivity, physical-chemical composition, and ultimately the validity and adaptability of a given variety in a region [3]. In the Tempranillo variety, the average temperature range in

the vegetative period (from April to October for the Northern Hemisphere) should be between 15 and  $19^{\circ}C$  [4]. Climate change (CC) is causing a progressive rise in temperatures, as well as increasingly frequent and prolonged episodes of heat stress and increased incident radiation, in addition to modifications in the seasonal pattern of rainfall, causing greater uncertainty in the availability of water resources [5].

Over the past 50 years, major wine regions have recorded temperature increases of more than 1°C and are expected to rise by a further 2°C by 2050 [6]. Climate change is being identified as responsible for changes in grapevine phenology and physiology, such as longer lengthening of the growth cycle, faster phenological advancement in successive stages, and earlier harvest dates. As a consequence, the composition of grapes and wines is modified and their quality is altered [7, 8]. Grape quality at harvest depends on the content of primary metabolites (sugars, organic acids, and nitrogen compounds) and secondary metabolites (phenolic and aromatic compounds). As temperatures rise, the accumulation of sugars is accelerated by changing the synchrony between fruit development and the evolution of other metabolites such as acids, polyphenols, and aromatic substances [9]. While tartaric acid (TAR) is moderately stable against temperature changes, malic acid (MAL) is strongly affected by temperature and ripening stage [2, 10]. On the other hand, temperatures above 30°C can reduce anthocyanin synthesis and even inhibit it above 37°C. Sadras and Moran [11] observed a decoupling of anthocyanin and sugar contents in red varieties subjected to environmental stress conditions. Results vary depending on plant material and conditions but harvests with excessively high sugar concentrations, excessively low acidity and polyphenols, and an aromatic expression dominated by stewed fruit aromas, and changes in wine style are cited [11-18]. The increase in potential alcohol contrasts with current consumer preferences, which are moving toward wines with moderate alcohol content [19]. Reducing sugar content is interesting to minimize the cost of dealcoholization of wines, which is limited to 2% vol [20].

Among the strategies proposed to reduce the effect of high temperatures and drought on grape production and quality are: the selection of better adapted plant material (rootstock and variety) and agronomic techniques such as irrigation, modifications in plant architecture, and different pruning strategies. [5, 8]. In 2012, Gu et al. [21] published the results obtained from a green pruning technique they called "crop forcing," aimed at prompting the grapevine growth cycle to restart after flowering. These authors managed to shift the ripening period of Cabernet Sauvignon in California from July-August to October-November, modifying grape production and characteristics at harvest. In recent years, this technique has been studied in different parts of Spain in cv. Tempranillo in Valencia [22] and the cv. Tempranillo and cv. Maturana Tinta in La Rioja [23] and recently in Japan, in the Yamanashi region, and in cv. Merlot, with similar results in all cases [24]. This technique decreased the yield but increased berry acidity and polyphenolic content. An alternative proposal to compensate for the crop loss caused by crop forcing has been to keep the original clusters on the forced grapevines, which results in a double harvest [25, 26].

The grapevine is considered a plant adapted to drought conditions [27, 28]. Most of the vineyards are located in regions with strong seasonal droughts. Under these conditions, the plant depends on the water storage capacity of the soil to cover the strong evaporative demand that accompanies high temperatures and radiation levels and scarcity of rain during a good part of the growth cycle and usually leads to low yields and deterioration in quality. Although the highest yields are achieved in vineyards that do not suffer from water limitations, moderate stress can improve berry characteristics and sensory properties in wines [29-32]. Water deficit usually reduces the acidity (TA) of the wines but increases the concentration of total polyphenols (TPP) and total anthocyanins (TAN) [33], and tasting panels report that quality attributes improve with the application of a controlled water deficit [30]. A number of deficit irrigation strategies have been proposed for vineyards that increase production compared to rainfed but enable the concentration of compounds of high enological value. These include Regulated Deficit Irrigation (RDI) which consists of maintaining good water availability in the plant during production-sensitive periods and reducing irrigation water to cause controlled stress in periods that are not critical for production and/or important to stimulate or conserve compounds important for winemaking [34-36]. Deficit irrigation during preveraison favours anthocyanin accumulation as a consequence of reduced berry size and increased expression of genes responsible for flavonoid synthesis [33, 34, 37], bringing grape ripening forward [38], and has been used to minimize decoupling of phenolic and technological ripeness caused by high temperatures [11]. On the contrary, water stress-applied postveraison increases the proportion of seeds and skin in grapes without significantly affecting secondary metabolism [39]. In general, deficit irrigation increases total soluble solids (TSS) [40-42] so that the wines obtained continue to show high alcoholic strength. The results of this technique may be different depending on the variety, timing, duration, and intensity of the deficit periods, as well as the weather conditions and the previous history of the vineyard, so that interannual differences are observed in response to the same strategy [36, 43]. Previous studies carried out in the region of Extremadura [43], with average annual rainfall around 450 mm, recommended applying a sustained deficit irrigation strategy of 25% of ETc, with which they achieved an average yield increase of 26% relative to rainfed with similar concentrations of TPP and TAN [43]. Under the same conditions, regulated deficit irrigation with water deficit in the preveraison period increased TA by improving malic acid concentration compared to rainfed [44].

Spain's grape-growing surface area is in excess of one million hectares, of which more than 80% is devoted to wines with Protected Designation of Origin (PDO) (Ministry of Agriculture, Fisheries, and Food). Most of this area is located in semiarid climate zones, which include the main vineyard areas of Extremadura. Figure 1 shows the average temperatures (Tmed) during the vegetative period of the grapevine from 1997 to 2021 in the trial plot, located in Vegas Bajas, Ribera del Guadiana PDO, southwestern Spain (Extremadura) from Budburst to harvest in 1997-2021. In all cases in this area, the average mean air temperature was above 20°C, outside or close to the recommended limit for the varieties grown in the region [4]. Berry ripening coincides with the highest temperatures of the year, with averages of up to 35°C for the months of June, July, and August (Figure 2), which then drop off in the fall. V. vinifera L. cv. Tempranillo is the main variety of half of the Spanish designations of origin, widely cultivated in the north, centre, and south of the country [45].



FIGURE 1: Average mean temperatures from April 1 to September 30 in Extremadura, Spain, from 1997 to 2019.



FIGURE 2: Distribution of maximum (black), mean (grey dashed line), and minimum (grey solid line) temperatures throughout 2017 in Extremadura, Spain, and average duration of the period from Budburst to harvest of cv. Tempranillo in the same area.

This paper evaluates the effect of the crop forcing technique on berry ripening process and final composition. The effects of this technique are compared to the application of a deficit irrigation strategy recommended for the Tempranillo variety under Extremadura conditions in nonforced grapevines, as well as the combined effect of both techniques. The objective of this study is to provide winemakers with the most suitable raw ingredients for the desired winemaking processes and to provide grape growers with tools to obtain higher value harvests that make their vineyards profitable and increase the resilience of vineyards in warm areas in the face of climate change.

## 2. Materials and Methods

2.1. Plant Material and Vineyard Site. The study was carried out in an experimental vineyard located at Badajoz, Extremadura, Spain (38°51'N; 6°40'W; 198 m), planted with Tempranillo (*Vitis vinifera* L.) grafted on Richter 110 rootstock, and trained as bilateral cordons in a vertical trellis system with a drip irrigation system of 8 L/h per grapevine. All the grapevines were winter pruned to six spurs and two buds per spur. The rows are laid out E-W, and row and grapevine spacing is 2.5 m and 1.2 m, respectively.

2.2. Treatments and Experimental Design. The experimental design was split-plot with four replications (Table 1). The main factor consisted of two treatments with crop forcing techniques on two different dates: F1, crop forcing applied 4 days after anthesis (May 18, 2017; May 29, 2018; May 20, 2019), and F2, 22 days after anthesis, both treatments were compared with a treatment without crop forcing techniques (NF) with grapevines grown following conventional practices (just winter pruning) [46]. Crop forcing consisted of hedging the growing shoots to seven nodes and removing all the summer laterals, leaves, and clusters with scissors to force the bursting of the primary buds developed in the current season. Two irrigation treatments were set up as a secondary factor. The irrigation treatments were: an irrigated Control (C) to maintain the midday stem water potential (SWP) around -0.6 MPa and a Regulated deficit Irrigation (RI) to which water was supplied in preveraison to reach -1.1 MPa and maintained above -0.8 MPa at postveraison. These treatments were maintained in the same vine during the three consecutive years [46].

The grapevine water requirements were calculated based on the crop evapotranspiration (ETc) using the crop coefficient (Kc) recommended by the FAO for these latitudes for the NF treatments. For the F1 and F2 treatments, ETc was calculated directly on a weighing lysimeter [47] with two crop forcing grapevines, integrated in the study plot. Irrigation started when a threshold value of  $\Psi$ smd of -0.6 MPa was reached. Irrigation was applied five to six times per week, measuring the amount of water applied to each subplot through volumetric water meters and maintaining irrigation until early and mid-October. Meteorological data come from a weather station belonging to the Extremadura irrigation advisory network (REDAREX) located at a distance of 100 m from the plot. The experimental unit consists of 6 rows of 18 grapevines. Ten central grapevines of the four

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	No crop forcing (NF)	Early crop forcing (F1)	Late crop forcing (F2)
Fully irrigated (C)	C-NF	C-F1	C-F2
Deficit irrigation (RI)	RI-NF	RI-F1	RI- <i>F</i> 2

TABLE 1: Summary of the treatments applied in the study [46].

central rows are for sampling and harvest. The total amount of irrigation applied and the ETo data for each growing season are summarised in Table 2.

2.3. *Measurements*. The phenological observations were carried out in 10 grapevines in each experimental plot, every 7 days from Budburst to veraison. For data analysis, only the two main phenological stages (Budburst and veraison) were used. Veraison was determined by colour change in 50% of berries.

Three hundred berries per experimental unit were randomly collected, from different positions within clusters and plants, weekly from pea size to harvest. 100 berries were weighed fresh.

From veraison to harvest, 200 grapes were frozen in the laboratory until analyzed. The rest were destemmed, crushed, and homogenized for 1 min at speed setting 3 (Mycook blender Taurus, Oliana, Spain). An aliquot of the resulting mash (pulp, juice, skins, and seeds) was filtered and used to determine technological parameters. Total Soluble Solids (TSS) (°Brix) was determined by refractometry (RE40D, Mettler Toledo, Greifensee, Switzerland), pH with a pH-meter (Basic 20, Crison, Alella, Barcelona), and Titratable Acidity (TA, g tartaric acid/L) with a titrator (T50, Mettler Toledo, Greifensee, Switzerland) according to ECC formal methods [48]. Malic (MAL) and tartaric (TAR) acid content (g/L) was determined according to ECC formal methods using an autoanalyzer (Y15, Biosystems, Barcelona, Spain).

Extraction of phenolic substances from grapes was carried out employing a methodology based on previous works [49, 50]. 100 g samples of frozen berries (-80°C) were crushed and homogenized for 30 seconds in a Freshboost blender (LM180110, Moulinex, Aleçon, France). 10 mL of the hydroalcoholic solution (methanol/water/formic acid 50:48.5:1.5, v/v/v) were added to an aliquot of the obtained mash (1.0 g) and macerated for 30 minutes at 4°C in an ultrasonic bath (USC-TH, VWR, Radnor, USA) and centrifuged at 4°C for 10 min (5810 R, Eppendorf, Hamburg, Germany). The supernatant was separated, and the resulting pellet was extracted up to three times. The supernatants (phenolic extracts) were then combined, and the final volume was annotated. Total Polyphenol Content (TPP) from the extracts was determined according to Singlenton and Rossi [51]; and Total Anthocyanin (TAN) content was quantified employing the pH differential method [52]. All determinations were carried out using an autoanalyzer (Y15, Biosystems, Barcelona, Spain). Two extractions were performed for each sample of a given plot and sampling date.

All treatments were harvested manually at 23-24°Brix, a common criterion for picking red grape varieties in this area. The average TSS of the berries from the four elementary plots was considered for each treatment. All the clusters of 10 grapevines per experimental plot were weighed (40 grapevines per treatment). The numbers of clusters per shoot were counted and weighed on a total of ten vines per experimental plot.

Total pruning dry weight was determined in the different interventions carried out: green pruning, forcing pruning, and winter pruning, of the same ten grapevines per experimental plot.

2.4. Statistical Data Analysis. Normality and homogeneity of variances were tested using Shapiro-Wilk's and Barlett's test, respectively. When the normality and homogeneity of variances were verified, data were subjected to analysis of variance (MANOVA) to study the effect of "crop forcing," irrigation," and their interaction on each parameter evaluated selecting  $p \le 0.001$ ,  $p \le 0.01$ , and  $p \le 0.05$  for significance of comparisons. The interaction between effects was evaluated by calculating the least-squares means (LS means) selecting  $p \le 0.001$ ,  $p \le 0.01$ , and  $p \le 0.05$  for significance of comparisons and the Tukey test as post hoc tests for parametric samples. When the normality and homogeneity of variances were not verified, nonparametric tests were carried out employing the Kruskal-Wallis test (alternative to one-way ANOVA) and multiple comparison p values (alternative to posthoc pairwise comparisons). Differences between means were considered statistically significant when p < 0.05. The relationships between TSS values and the rest of investigated parameters were assessed by regression analysis, and the comparison of slopes was tested by performing the corresponding analysis of variance (ANOVA). SPSS software package 12.0 for Windows (SPSS Inc., Chicago, IL) was used for processing the data. Also, principal component analysis (PCA) was performed to discriminate among treatments based on the values of acid and polyphenolic parameters, and the relationships between TSS values and the rest of parameters under study were assessed by regression analysis. This last statistical test was performed with XLSTAT-Pro 201610 (Addinsoft, 2009, Paris, France).

## 3. Results

3.1. Grapevine Timeline and Growing Conditions. The irrigation strategies did not modify the phenological cycle of the grapevines (results not shown). However, the application of crop forcing treatments displaced it, both in F1 and F2. Figure 3 reflects the growth cycle in the different treatments: from natural Budburst to harvest in C-NF and RI-NF and the first (C-F1 and RI-F1) and second postforcing (C-F2 and RI-F2) Budburst to respective dates of harvest. Figure 3 also shows the maximum temperatures during the ripening period of the different treatments. Since the forcing

TABLE 2: Irrigation applied and evapotranspiration (ETo), from budbreak to harvest on nonforced vines (NF) and from crop forcing application date to harvest date in F1 and F2 during 2017, 2018, and 2019 years.

Tuestasent	2017		2018		2019	
freatment	Irrigation (mm)	ETo (mm)	Irrigation (mm)	ETo (mm)	Irrigation (mm)	ETo (mm)
C-NF	286	881	285	763	347	909
C-F1	334	782	516	715	482	824
C-F2	405	788	524	671	499	781
RI-NF	56	848	155	763	220	876
RI- <i>F</i> 1	228	782	256	715	373	824
RI-F2	212	788	261	671	373	781



FIGURE 3: Maximum daily temperatures from July 29 through November 16 for the three years under study. Horizontal bars represent the days between veraison and harvest for NF, F1, and F2.

treatments moved phenological stages to later dates in the year, they occurred with lower temperature than in NF treatments.

With respect to NF, the average decrease in maximum temperature for this period was  $1.5^{\circ}$ C in 2017,  $3^{\circ}$ C in 2018, and  $2.7^{\circ}$ C in 2019 in F1 and 5.7, 8, and  $4.6^{\circ}$ C for F2. Annual rainfall was 282, 479, and 293 mm in 2017, 2018, and 2019, respectively. 2018 was the wettest year in both spring and autumn. The length of the F1 and F2 ripening cycle was shortened in 2017 and 2019 relative to NF, while it was lengthened in 2018.

As shown in Table 3, harvest dates varied according to year and crop forcing treatment. With respect to NF, the crop forcing technique shifted the harvest date, causing delays in the harvest, which were longer in *F*1 than in *F*2. In *F*1, grapes were harvested between September 12 and October 8, while in *F*2 the grapes were picked between October 15 and 29. In addition, when analyzing the different years, it is observed that, with respect to NF, the number of *F*1 delay days was 23, in 2017, 41 in 2018, and 34 in 2019. In *F*2, delays varied less depending on the year considered and were 58 days, 61 days, and 43 days, respectively, in the same years.

3.2. Berry Weight Evolution, Yield Components, and Vine Vigor. Figure 4 shows the evolution of berry unit weight throughout the growth cycle (fruit set to harvest) and the veraison dates of the different treatments corresponding to the three years under study. In all cases, the berry growth pattern follows a double sigmoid curve but, in the three years under study, the irrigation treatment and, particularly, the pruning technique caused differences in the evolution of this parameter. On the one hand, in the crop forcing treatments (C-F1, C-F2, RI-F1, and RI-F2), the onset of the curve was delayed. On the other hand, in the first two years, NF and F2 berry growth is evenly distributed between preveraison and

postveraison, but in F1, berries gain more weight after veraison (Figures 4(a) and 4(b)). In 2019, weight gain is higher during preveraison in all treatments (Figure 4(c)). Also, it is noteworthy that a few days after fruit set, differences between C-NF and RI-NF begin to be identified, increasing as the cycle progresses, while in F1 and F2, there are hardly any differences in the evolution of the weight of the two irrigation treatments for the same forcing date in any year of the study.

The grapevines to which the forcing treatments were applied were less productive. In the three seasons analyzed, the mean value of the F2 treatments was higher than F1 although in 2018 the differences were not significant. In addition, 2019 should be highlighted as the most productive year and also the year when there was the greatest difference in C-NF with the crop forcing treatments. For the same pruning treatment, yield was higher in C than in RI. RI-NF had higher yield than any of the crop forcing treatments, with significant differences only in 2019. Irrigation according to needs was more productive than RI although the differences were significant only in 2018. When comparing the two irrigation treatments with the same pruning, the control was always more productive, although differences were only significant in 2019.

All vines were pruned in winter leaving twelve buds per vine, from which the corresponding shoots developed. In the forced treatments, each shoot was forced in the current season leaving six buds per shoot. Although the number of clusters per shoot varied across the study years, the forced treatments had a higher number of clusters per shoot during the study. The nonforced treatments (C-NF and RI-NF) had 1.3 average bunches per shoot, while F1 and F2 reached 1.6 and 1.8 bunches per shoot, respectively (Table 4). The effect of irrigation was significant only in 2017 where RI had lower number of bunches per shoot. This parameter showed a high irrigation-forcing interaction, but these interactions did not

TABLE 3: Budburst and harvest dates and duration of the cycle.

Treatment		Budburst date			Harvest date		Days	(from Budb harvest)	urst to
	2017	2018	2019	2017	2018	2019	2017	2018	2019
NF	3-Apr	3-Apr	26-Mar	22-Aug	27-Aug	27-Aug	141	148	154
F1	18-May	29-May	20-May	12-Sep	08-Oct	30-Sep	117	132	133
F2	06-Jun	17-Jun	03-Jun	17-Oct	29-Oct	15-Oct	133	134	138

In F1 and F2, the budburst date is taken as the date of (forced) summer pruning.



FIGURE 4: Evolution of berry fresh weight in (a) 2017; (b) 2018; (c) 2019. Each point represents the mean of 8 samples (4 blocks, 2 replicates). The bars represent the standard error of the mean. In the upper part, the time of veraison for the different forcing treatments is shown. DOY: days of year.

show a clear trend during the study, as they varied depending on the year. The effect of treatments on the total dry weight of the different pruning interventions (green pruning, forcing pruning, and winter pruning) was different in each year (Table 4). In 2017, the pruning was higher in F2 than NF. In 2018, C-NF and C-F1 had the highest weight and RI-NF the lowest. In these two years, the total dry weight of pruning was lower in the RI-NF treatment compared to C-NF, while in 2019 there was no difference.

#### 3.3. Grape Ripening

3.3.1. Technological Ripeness (Total Soluble Solids and Acids). As reflected in Figure 5, crop forcing treatments delayed the grape ripening period but did not affect the metabolite increase and decrease patterns in the berries. Typical decreases in MAL and TA values and increases in berry Brix and pH were observed in these treatments. The delay in ripening dates did not prevent reaching the Brix value set for harvesting in F1 and F2. Of note is the evolution of these parameters in the F2 treatments in 2018 and 2019. Figure 5 shows that, in F2, the drops in MAL and AT were less pronounced and a stabilization of the values of these parameters was observed, and also the pH values were lower than in the rest of the treatments during the whole period analyzed. In 2017, there was a greater chronological separation between the three prunings, while in 2018 and 2019, F1 was further away from C and closer to F2 (Figure 5).

It is noteworthy that, for the same pruning treatment, the ripening processes took place on identical dates under both irrigation strategies. With regard to C, the effect of RI was higher in NF berries than in F1 and F2 berries. In the three years, the evolution of TSS values was practically identical in the C and RI treatments except for the 2019 NF treatments. The MAL evolution curves show lower values in RI-NF than in C-NF throughout the process, while no differences were observed in F1 and F2 curves, except for F1 in 2018 when RI-F1 falls below C-F1. In TAR, RI maintains higher values than C in NF, with these differences being clearer in 2017. In the forced grapevines, the values are similar in the two irrigation treatments, except for 2018, when C has higher RI values throughout ripening in F1 and F2. As a result, in all three seasons, the TA values of the C-NF treatments remained below those of RI-NF treatments, while in F1 and F2, they were very similar throughout the cycle. Finally, pH values of RI-NF berries remained above those of C-NF in all samples of the 2018 and 2019 vintages, while C-F1 and RI-F1 were very similar. In F2, pH stabilized at lower values in C in the first two harvests under study and no differences were observed in the third harvest.

3.3.2. Phenolic Ripeness. Figure 6 shows the evolution of TAN and TPP values. The treatments subjected to forcing had a similar evolution dynamic to the corresponding NF, increase and tendency to stabilization in TPP and decrease and tendency to stabilization in TAN. However, the initial values of F1 and F2 are higher, and at the time of harvest,

TABLE 4: Yield, number of cluster	s per vine,	and total pru	ining weig	ht (green p	runing, fo	rcing prun	ning, and w	rinter prur	ing) in p	runing (N	F, <i>F</i> 1, and	F2) and ir.	rigation	(C and F	tI) treatr	nents.
Downortow	Voor	Statistical		С			RI		$L \sim I$		Forcing		ΕĽ	Irriga	tion	11
r al allicle1	1 CAI	analysis	NF	F1	F2	NF	F1	F2	LVI	NF	F1	F2	1.1	U	RI	
	2017	d	3.89	1.94	2.77	2.26	1.29	1.31	n.s.	3.07 a	1.61 c	2.04 b	* * *	2.86	1.62	* *
Yield (kg/vine)	2018	d	3.73	1.65	1.84	3.05	1.54	1.78	n.s.	3.39 a	1.60 b	1.81 b	* * *	2.41	2.12	n.s.
	2019	$\overline{p}$	7.12 a	2.25 d	3.00 c	4.41 b	2.01 d	2.88 c	* *	5.77	2.13	2.94	* *	4.12	3.10	* *
	2017	n.p.	1.1 bc	1.4 b	2.1 a	0.9 c	0.9 c	1.0 bc	* * *	1.0	1.2	1.6	* * *	1.6	0.9	* * *
Clusters per shoot	2018	n.p.	1.3 b	2.4 a	1.6 b	1.5 b	2.3 a	1.8 ab	* *	1.4	2.3	1.7	* * *	1.8	1.8	n.s.
1	2019	n.p.	1.5 b	1.3 bc	2.2 a	1.4 bc	1.1 c	2.3 a	* * *	1.5	1.2	2.2	* * *	1.7	1.6	n.s.
	2017	n.p.	0.9 b	1.1 ab	1.2 a	0.6 c	1.0 ab	0.9 b	* * *	0.7	1.0	1.0	* * *	1.0	0.8	* * *
Total pruning weight (kg/vine)	2018	n.p.	1.2 a	1.2 a	1.0 ab	0.8 b	1.0 ab	1.0 ab	*	1.0	1.1	1.0	n.s.	1.1	0.9	* *
	2019	n.p.	1.1	1.0	0.9	1.0	1.0	0.9	n.s.	1.0	1.0	0.9	n.s.	1.0	1.0	n.s.
Statistical analysis: MANOVA and T irrigation factor. Different letters indi (***), significant at 0.1 percent level.	ukey test as cate the exi	s a parametric t stence of statist	test and Kru tically signif	ıskal–Wallis îcant differe	s test as a n inces betwee	onparameti en treatmen	ric test (bot) its; n.s. indic	h $p < 0.05$ ). cates not sig	<i>F×I</i> : intention	raction bet *), significa	ween forcin nt at 5 perc	g and irrigs ent level; (**	tion effe ), signifi	cts; FF: fo cant at 1 p	rcing fac ercent le	or; IF: /el and
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FIGURE 5: Evolution of (a)-(A) total soluble solids (TSS) in 2017, (a)-(B) total soluble solids (TSS) in 2018, and (a)-(C) total soluble solids (TSS) in 2019; (b)-(A) titratable acidity (TA) in 2017, (b)-(B) titratable acidity (TA) in 2018, and (b)-(C) titratable acidity (TA) in 2019; (c)-(A) malic acid (MAL) in 2017, (c)-(B) malic acid (MAL) in 2018, and (c)-(C) malic acid (MAL) in 2019; (d)-(A) tartaric acid (TAR) in 2017, (d)-(B) tartaric acid (TAR) in 2019; (e)-(A) pH in 2017, (e)-(B) pH in 2018, and (e)-(C) pH in 2019. Each point represents the mean of 8 samples (4 blocks, 2 replicates). The bars represent the standard error of the mean. DOY: days of year.



FIGURE 6: Evolution of the content in (a)-(A) total anthocyanins (TAN) in 2017, (a)-(B) total anthocyanins (TAN) in 2018, and (a)-(C) total anthocyanins (TAN) in 2019; (b)-(A) total polyphenols (TPP) in 2017, (b)-(B) total polyphenols (TPP) in 2018, and (b)-(C) total polyphenols (TPP) in 2019. Each point represents the mean of 8 samples (4 blocks, 2 replicates). The bars represent the standard error of the mean. DOY: days of year.

they still maintain higher values than NF. Deficit irrigation acted as a stimulus for anthocyanin synthesis in all three pruning treatments in 2017 and in NF also in 2018 and 2019. The effect of forcing treatments on these substances was more evident in the last two harvests. In *F*1 and *F*2, the accumulation of these compounds was higher. In 2017 and 2018, in the forced grapevines, TAN dropped faster and final stabilization was shorter as we came closer to the harvest date.

3.4. Balance of Berry Traits. To analyze the effect of treatments on the balance between different berry compounds, regressions were fitted between TAN, TPP, TA, pH, MAL, and TAR with TSS throughout ripening (Figure 7). To simplify the presentation of the data, treatments C-F1 and C-F2 have been excluded from the figures, since the developments presented in the previous section (Figure 5) indicate that with the same green pruning date, ripeness development was similar in the two irrigation treatments, with greater differences with the F treatments in the case of RI. Table 5 shows Pearson's coefficients for the correlations between these compounds and the TSS and the significance. Table S1 of the supplementary information includes the equations, the coefficient of determination, and the comparisons of the slopes of the correlation lines. In all cases, the comparisons of the slopes were statistically significant, and at least two groups were established between the four treatments analyzed (Table S1).

The correlation between pH and TSS of the different treatments was different in the three years under study (Figure 7(a)). In 2017, all treatments maintained a similar balance between both compounds, except RI-F, which started from a significantly lower pH for the same TSS, and the rise in pH as ripening progressed was significantly

higher. In the two subsequent years, more differentiated relationships between treatments were identified. The pH of RI-NF was higher with 22 TSS. In these same years, RI-F2 reaches this TSS concentration at a similar or lower pH than C-NF.

The correlation between TA and TSS showed that as maturation progressed, TSS increased and TA decreased (Figure 7(b)). In 2017, the 4 treatments maintain a similar correlation. In 2018, with increasing TSS, the 4 treatments tend to equal TA with 24°Brix, while in 2019, RI-F1 and RI-F2 have higher TA with similar TSS throughout.

The correlation between malic acid and TSS also decreases linearly (Figure 7(c)). In 2017, at approximately 22°Brix, RI-F1 and RI-F2 match the C-NF line, while RI-NF shows lower malic acid values. In 2018, the trend is very similar to the previous year but, in this case, RI-F1 shows similar values to RI-NF. In 2019, C-NF is the treatment showing the lowest values across the line, followed by RI-NF. RI-F1 and RI-F2 are the treatments that show the highest values and are very similar to each other.

The correlation between TPP and TSS showed a decreasing trend (Figure 7(d)). The differences between treatments are appreciable throughout the line. C-NF shows the lowest values in all three years, followed by RI-NF (with the exception of 2018 which does not show RI-NF data). The treatment with the highest values is RI-F2 in 2017 and 2019, while in 2018 it is RI-F1, although the differences between these two treatments are small in all three years.

The correlation between TAN and TSS increases linearly (Figure 7(e)). RI-F2 is the treatment with the best results over the three years, followed by RI-F1. In 2017, RI-F1 is no different from RI-NF in 2017. In all three years, C-NF shows the lowest values in all cases, although in 2019 the RI-NF and C-NF values are very similar.



FIGURE 7: Regression between total soluble solids (TSS) and (a)-(A) pH in 2017, (a)-(B) pH in 2018, and (a)-(C) pH in 2019; (b)-(A) titratable acidity (TA) in 2017, (b)-(B) titratable acidity (TA) in 2018, and (b)-(C) titratable acidity (TA) in 2019; (c)-(A) malic acid (MAL) in 2017, (c)-(B) malic acid (MAL) in 2018, and (c)-(C) malic acid (MAL) in 2019; (d)-(A) tartaric acid (TAR) in 2017, (d)-(B) tartaric acid (TAR) in 2018, and (e)-(C) total polyphenols (TPP) in 2017, (e)-(B) total polyphenols (TPP) in 2018, and (e)-(C) total anthocyanins (TAN) in 2017, (f)-(B) total anthocyanins (TAN) in 2018, and (f)-(C) total anthocyanins (TAN) in 2017, (f)-(B) total anthocyanins (TAN) in 2019.

TABLE 5: Pearson correlation between total soluble solids content (TSS) and pH, total acidity (TA), malic acid (MAL), tartaric acid (TAR), total polyphenols (TPP), and total anthocyanins (TAN).

		C-NF	RI-NF	RI- <i>F</i> 1	RI-F2
	pН	0.908 (***)	0.918 (***)	0.836 (***)	0.845 (***)
	TA	-0.944 (***)	-0.930 (***)	-0.823 (***)	-0.841 (***)
2017	MAL	-0.934 (***)	-0.888 (***)	-0.842 (***)	-0.844 (***)
2017	TAR	-0.540 (**)	-0.834 (***)	-0.435 (n.s.)	-0.557 (*)
	TPP	-0.715 (***)	-0.734 (***)	-0.854 (***)	-0.739 (**)
	TAN	0.841 (***)	0.889 (***)	0.862 (***)	0.717 (**)
	pН	0.931 (***)	0.910 (***)	0.970 (***)	0.909 (***)
	TA	-0.935 (***)	-0.848 (***)	-0.929 (***)	-0.908 (***)
2010	MAL	-0.944 (***)	-0.686 (**)	-0.802 (***)	-0.784 (***)
2018	TAR	-0.691 (**)	0.349 (n.s.)	-0.485 (*)	-0.653 (**)
	TPP	-0.698 (**)	-0.025 (n.s.)	-0.501 (*)	-0.750 (***)
	TAN	0.949 (***)	0.776 (***)	0.852 (***)	0.770 (***)
	pН	0.943 (***)	0.852 (***)	0.972 (***)	0.951 (***)
	Τ̈́Α	-0.897 (***)	-0.907 (***)	-0.950 (***)	-0.928 (***)
2010	MAL	0.945 (***)	-0.906 (***)	-0.958 (***)	-0.950 (***)
2019	TAR	-0.695 (***)	-0.672 (***)	-0.818 (***)	-0.851 (***)
	TPP	-0.711 (***)	-0.882 (***)	-0.568 (***)	-0.735 (***)
	TAN	0.815 (***)	0.698 (***)	0.969 (***)	0.918 (***)

Pearson correlation: n.s. indicates not significant; (\*), significant at 5 percent level; (\*\*), significant at 1 percent level and (\*\*\*), significant at 0.1 percent level. Comparison of slopes was tested by performing the corresponding analysis of variance (ANOVA), in all cases p < 0.001.

3.5. *Berry Composition Effects of Treatments*. Table 6 shows the composition of the berries for a concentration of 22°Brix, considering this value as a reference for making wines that are less alcoholic.

It is noteworthy that a significant interaction of the forcing and irrigation effect was only observed in MAL in 2017 and in TPP in 2018 and 2019. In all three cases, minimum values were recorded in RI-NF. For the remaining parameters and years, the F \* I effect was not significant. This allows the effect of forcing and irrigation strategy to be analyzed separately.

The significance of the effect of forcing on berry acidity parameters depended on the parameter and season considered. In 2018 and 2019, the values of TA show an increase for F1 and F2 with respect to NF. However, differences between F1 and F2 only occur in 2019, the latter having a higher value. Overall, the effect of F1 and F2 relative to NF on TAR values was not clear. In 2017, the differences are found between NF and F2, while F1 results are intermediate. In 2018, NF and F1 achieve similar values and higher than F2. In 2019, it is F2 that shows a higher value, with differences with respect to NF, while F1 achieves intermediate values. For malic acid content in 2017, there is interaction between the two effects, with differences between C-NF and RI-NF, while all crop forcing treatments achieve intermediate values. In 2018, there is no difference in this parameter. The effect on malic acid is seen only in 2019, with higher results for F1 and F2 with respect to NF. All years showed the same tendency to higher berry pH values in forced grapevines with respect to the NF grapevines, and the effect was always greater in F2 than in F1. The effect on pH was significant in 2017 and 2019.

Regarding the effect of treatments on phenolic ripeness indicator parameters, a significant interaction was observed in TPP in the last two seasons, so that in both years the minimum values were recorded in C-NF. The values found in this treatment in 2019 were significantly lower than those corresponding to C-F1, C-F2, and RI-F2. In 2017, there is no interaction between the two effects, but there is an effect from forcing. The mean values of this parameter in F2 were significantly higher than those of the control grapevines. The results of forcing are clear and evident in the TAN values since in all years the contents of these F2 substances were significantly higher than those of NF.

The effect of irrigation strategy was less than that of forcing. In fact, most of the differences were found in 2018, when the lower volume of water applied in RI caused significant decreases in TA and MAL and increases in pH in berries with this treatment compared to those of treatment C. In addition, RI also caused increases in TAN in 2017 and 2018. In all years, RI-F1 and RI-F2 treatments resulted in the highest TAN values; therefore, a synergistic effect of forcing and deficit irrigation should be considered.

3.6. Principal Component Analysis (PCA) Classification of Treatments. Principal component analysis (PCA) was used to classify the different treatments in terms of values of acid (pH, TA, MAL, and TAR) and phenolic parameter values (TPP and TAN). The first PCA (Figure 8(a)) was performed with the data from the 2017 season. The figure reflects that PC1, 45.06% of the total variance, is strongly correlated whit pH and TAN (positive and negative side, respectively). According to the Figure 8(a), three groups can be distinguished: two of them located at the negative side of PC1 including F1 and F2 treatments. RI-F1 and C-F1 were associated with the highest of TAN and TPP values.

The second PCA was performed with the values obtained in 2018. In this year, PC1 correlated with pH and TA in

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Damatan	V 222	Statistical			Treat	ment			1 ~ 1		Forcing		11	Irriga	tion	11
rarameter	I ear	analysis	C-NF	C-F1	C-F2	RI-NF	RI-F1	RI-F2	$\Gamma \times I$	NF	F1	F2	гг	С	RI	IL
	2017	d	3.72	3.61	3.49	3.76	3.64	3.56	п.s.	3.74 a	3.63 ab	3.53 b	*	3.61	3.65	п.s.
Hd	2018	đ	3.48	3.50	3.45	3.74	3.56	3.56	п.s.	3.61	3.53	3.50	п.s.	3.48	3.62	* *
	2019		3.58	3.39	3.27	3.55	3.45	3.31	п.S.	3.56 a	3.42 b	3.29 c	* * *	3.42	3.44	п.s.
	2017	n.p.	5.77	5.65	5.53	5.65	6.28	5.60	п.s.	5.71	5.97	5.56	n.s.	5.65	5.84	п.S.
TA (g/L)	2018	đ	5.90	7.03	6.57	5.17	6.54	5.93	п.s.	5.53 b	6.78 a	6.25 a	* *	6.50	5.88	* *
•	2019		5.06	5.98	6.88	4.88	5.82	6.54	п.s.	4.97 c	5.90 b	6.71 a	* *	5.98	5.75	п.s.
	2017	d	3.31 a	3.08 ab	3.01 ab	1.93 b	3.06 ab	3.16 ab	*	2.62	3.07	3.08	n.s.	3.13	2.73	п.s.
Malic acid (g/L)	2018	þ	3.46	3.75	3.52	3.04	2.73	3.16	п.s.	3.25	3.24	3.34	п.s.	3.57	2.98	*
I	2019	þ	1.82	3.42	3.86	2.46	3.43	3.50	п.S.	2.14 b	3.43 a	3.68 a	* * *	3.03	3.13	п.s.
	2017	d	4.98	4.47	4.79	5.24	4.45	4.73	п.s.	5.04 a	4.46 b	4.76 ab	*	4.75	4.76	п.s.
Tartaric acid (g/L)	2018	þ	4.97	5.37	4.91	5.13	4.93	4.47	п.s.	5.05 a	5.15 a	4.69 b	*	5.08	4.84	п.s.
1	2019	þ	5.05	5.30	5.42	5.31	5.37	5.36	п.S.	5.18 b	5.34 ab	5.39 a	*	5.26	5.35	п.s.
	2017	Ρ	4.98	6.05	6.32	5.94	5.82	6.67	п.s.	5.46 b	5.93 ab	6.49 a	*	5.78	6.14	п.s.
TPP (mg/g)	2018	n.p.	5.00 ab	7.57 a	7.42 a	4.63 b	7.43 ab	6.48 ab	* *	4.82 b	7.50 a	6.95 a	* * *	6.66	6.18	п.s.
	2019	$\bar{P}$	4.06 d	5.90 abc	6.17 ab	4.78 cd	5.26 bcd	6.85 a	*	4.42 c	5.58 b	6.51 a	* *	5.37	5.63	п.s.
	2017	Ρ	0.97	1.17	1.08	1.12	1.24	1.47	п.s.	1.04 b	1.20 ab	1.28 a	*	1.07	1.28	* *
TAN (mg/g)	2018	Ρ	0.94	1.26	1.41	1.20	1.33	1.73	п.S.	1.07 c	1.29 b	1.57 a	* * *	1.20	1.42	* * *
	2019	Ρ	0.84	0.82	1.18	0.81	0.93	1.31	п.s.	0.83 b	0.88 b	1.24 a	* * *	0.95	1.02	п.s.
Statistical analysis: $p$ . in Different letters indicat percent level.	ndicates pa e the existe	urametric statis ance of statistic	tics $(p < 0.05)$ ally significar	); <i>n.p.</i> indicate it differences b	s nonparame etween treat	etric statistics ments; n.s., n	s ( $p < 0.05$ ). F; ot significant;	× I: interactic (*), significal	on betwee nt at 5 per	n forcing ar cent level; (	nd irrigation **), significar	effects; FF: f. ıt at 1 percer	orcing fa nt level, a	ctor; IF: ii ad (***), s	rrigation f significant	factor. : at 0.1

TABLE 6: Berry composition of pruning (NF, F1, and F2) and irrigation (C and R1) treatments: pH, titratable acidity (TA), malic acid, tartaric acid, total polyphenols (TPP), and total anthocyanins (TAN) at 22°Brix.



FIGURE 8: Principal component analysis on volatile families of compounds in Tempranillo wines in basis to forcing (NF, F1, and F2) and irrigation (C and RI) treatments: (a) 2017; (b) 2018; (c) 2019.

positive side discriminated the NF treatments (C-NF and RI-NF) from the rest. It is of note that C treatments were characterized by their values of TA.

Finally, Figure 8(c) shows that the first two main components (PC1 and PC2) explained 94.13% of the total variance (84.41 and 9.72%, respectively) of samples from 2019. RI-F1 and C-F1 and RI-F2 and C-F2 are located on the negative side of PC1 while NF treatments are on the positive side of the same axis. The F2 treatments strongly correlated with TA and TPP.

## 4. Discussion

High temperatures during ripening are typical of warm zone viticulture and often have negative effects on some of the desirable characteristics of red wine grapes. Severe water stress causes yield losses and, although it can improve some berry characteristics, it can also lead to an imbalance in grape ripening. With proper irrigation management, it is possible to improve both production and some of these characteristics, although it can also prejudice other traits. Numerous studies have aimed at designing irrigation strategies that make both aspects compatible. Such strategies are usually not universally valid and need to be adapted to the environment and the variety, often leading to varying results depending on the year. As a result of the work carried out in Extremadura, a strategy of regulated deficit irrigation with a reduction in water supply during preveraison is recommended to improve the content of polyphenols and total anthocyanins, yet with low titratable acidity values [43]. Another important aspect is that the application of RI causes faster ripening, which leads to an increase in TSS accumulation and alcoholic strength [12, 53]. This study aims to go a step further in the modification of berry characteristics, seeking to offer new enological possibilities or to alleviate some of the negative consequences of climate change on quality in hot areas.

The starting hypothesis of this work is that shifting grape ripening to a period with more suitable temperatures (lower and with a wider diurnal range) has a positive effect on berry characteristics. The crop forcing technique has been effective in achieving this displacement by lowering daily maximum temperatures during ripening (postveraison) by 5 to 8°C, particularly when applied after fruit set (F2). This change has modified berry composition by increasing phenolic compounds, like RI-NF, but reducing pH and increasing titratable acidity relative to this irrigation strategy [21-24, 54]. The effect of this technique was clearer in the F2 treatment because it caused a greater delay in the period of berry development.

Forcing displaced and modified the conditions of cluster development throughout the berry formation period, both preveraison and postveraison. Their composition at harvest depends on the whole process of berry formation: in the first phase after fruit set, organic acids, tannins, and some precursors of phenolic compounds accumulate [55]. At this stage, the treatments with crop forcing ripen at higher temperatures, between 5°C and 6°C, and with a shortening of the duration of this period. Figure 7 shows that the crop forcing treatments initiate veraison with a higher TSS content than the noncrop forcing treatments. In the postveraison stage, anthocyanins, compounds that are very sensitive to high temperatures, accumulate. At this time, the treatments with crop forcing are located in a period with significantly lower temperatures. On the other hand, summer pruning causes a shift of the clusters to a higher area of the trellis with greater exposure to solar radiation (photographs are included in the supplementary information). Bergqvist et al. [56] observed that, at high temperatures, excess sun exposure decreases colour in grapes. However, other studies [57] point to improvements in colour with increasing exposure as was the case in this study, possibly due to the combination of lower temperatures and higher lighting in postveraison.

The increase in phenolic compounds in grapes from deficit irrigation treatments has been attributed in part to the smaller size of the berries, as the proportion of skin and pips (where these compounds are located) increases in relation to the pulp [42]. As we have seen, the crop forcing treatments experienced a clear tendency to reduce berry size, reaching sizes similar to the RI-NF treatment (Figure 4). However, the dynamics of the different berry compounds throughout ripening were different between RI-NF and crop forcing treatments (Figure 7), so the smaller berry size was not solely responsible for the increase in phenolic compounds.

According to Sadras and Moran [11], high temperatures during ripening accelerate the accumulation of sugars, causing a decoupling of phenolic and technological ripeness. The crop forcing technique, although it effectively shifted ripening to a cooler period, did not prolong the duration of ripening and even shortened it in the 2017 season (Figure 3). The best coupling occurred when changing the dynamics of increasing or decreasing qualitative parameters (Figure 6). Figure 7 shows that the ripening process of the forced grapevines manages to improve the coupling between phenolic and technological ripeness, reaching a higher TAN and TPP content (in the three years of application) and TA (in 2018 and 2019) with a lower accumulation of TSS. The improvement of these parameters is greater with forcing (F1 and F2) than with deficit irrigation alone (RI-NF), when compared to the C-NF treatment. The combination of forced and deficit irrigation (RI-F1 and RI-F2) also improves the total anthocyanin content compared to the crop forcing treatments with control irrigation (C-F1 and C-F2), but decreases titratable acidity. Among the different crop forcing dates, the one applied after fruit set, F2, achieves the best results in terms of final fruit quality with a greater displacement of berry formation and ripening.

Although in this work we do not present results for wines made with grapes from forced vines, it is to be expected that, starting with better quality grapes, the vinification process will result in higher quality wines. At the very least, with this technique, the enologists are provided with different grapes that increase the possibilities for enological elaborations. In view of the figures shown in Table 6, F1 and F2 could be harvested at 22°Brix, in order to aim the harvest toward wines with lower alcoholic strength, obtaining improvements in TAN, TPP, and TA content, even when compared to the values obtained in NF at 24°Brix. These results are reaffirmed with those obtained in the PCA (Figure 8), where F2 stands out with better results in the content of these compounds. The composition is even more balanced if forced irrigation is combined with deficit irrigation during preveraison. These results demonstrate that it is possible to provide winemakers in this area with grapes of the cv. Tempranillo for the production of wines with lower alcoholic strength, higher acidity, and total anthocyanin and polyphenol contents than with conventional practices. Martínez De Toda et al. [58] managed to delay harvest by two weeks in the cv. Garnacha maintained the TSS of the unpruned control but with increases in anthocyanin content, while [59] applied this same technique on the cv. Tempranillo did not achieve notable increases in anthocyanin concentration, which highlights the importance of adapting this type of technique to the particular grape-growing area.

Although there are few published papers on this technique, in all cases, it implies lower production than treatments used as control with conventional pruning practices [21–24, 54]. This reduction in production also occurs with other viticultural practices aimed at improving berry quality, such as deficit irrigation, cluster thinning, or cluster tip removal. As with these other practices, for crop forcing to be of interest, it must demonstrate a significant and interesting effect for winemaking. When compared to an RI, these

production differences are smaller, since between C-NF and RI-NF, there is a drop in production of between 20 and 40 percent depending on the year. The forced treatments had a more stable production over the three years compared to C-NF. The table shows that F2 can reach 3 kg/grapevine, which with a planting layout of  $2.5 \times 1.2$  m, which means a production close to the maximum limit authorized by Spanish designations of origin (8000 kg/ha). In an attempt to reduce yield loss, the vines were forced by increasing the crop load (6 buds per shoot). This was intended to increase the number of bunches per vine. The number of bunches per shoot varied throughout the year, and although it was higher in F1 and F2, yields were not similar to those of the nonforced treatments, as the number of berries per bunch and consequently the average weight of the bunches were drastically reduced from 212.3 g in the C-NF treatments compared to 68.1 g and 77.7 g observed in F1 and F2 (p > 0.001). The basal inflorescences usually form the most flowers, and numbers decrease for higher inserted inflorescences [60] and elevated temperatures either before or after Budburst reduces the flower number [61] what happens when sprouting takes place after forcing pruning. Likewise, fruit set can be negatively affected not only by temperatures above 32°C [62] but also under conditions of high light intensity (due to its impact on the current supply of photoassimilates) as reported by Friend [63]. Both high temperature and high light intensity conditions occur during the flowering and fruit set process with the phenology shift in forced vines and could be responsible for the lower number of berries per bunch observed in this study.

In order to minimize the loss of production caused by green pruning, several authors have proposed a double harvest [25, 26]. With this technique, it is possible to obtain a higher production with the quality benefits of green pruning. However, the feasibility of this proposal will depend on the short- and mid-term on the vineyard, as well as cost overruns and harvest difficulties.

Unlike previous studies with this same technique [21–23, 54], in this study, the crop forcing treatments were maintained on the same grapevines throughout the three years. In 2019, the third consecutive year of application of the treatments on the same grapevines is when the greatest differences in grape composition between the crop forcing and noncrop forcing treatments were observed. These results agree with those of [24], who studied the effect of two types of crop forcing application in two consecutive years on the same grapevines of cv. Merlot, in Japan. The greater differences obtained in 2019 could either be a cumulative effect or coincide with the year in which there are greater differences in production between NF and F treatments, since the greatest differences in temperatures during ripening correspond to 2018, which is when the harvests in F1 and F2 were most delayed in relation to NF. Higher yields can decrease berry quality due to a dilution effect [42]. Gu et al. [21] and Tian and Gu [54] achieved an increase in TPP, TAN, and TA content in the first year of application of this technique for cv. Cabernet Sauvignon for the same degree of TSS accumulation (24°Brix). In 2019, Martinez De Toda et al. got similar results for cv. Garnacha, cv. Tempranillo, and cv. Maturana Tinta, with a significant improvement also in malic and tartaric acid content and with a difference in TSS between unforced (22°Brix) and forced (20°Brix) grapes. In a preliminary study in 2017, an increase in TPP, TAN, and TA content in Tempranillo grapes from forced vines at harvest for the same TSS value involved an increase in TPP, TAN, and malic acid content in wines from those same vines [64]. In this study, an improvement in TPP and TAN content has been achieved from the first year of application for cv. Tempranillo at 22°Brix. However, the TA content did not improve until the second year of application on the same grapevines. Malic acid content improved in 2019, as did tartaric acid and only in the case of crop forcing applied after fruit set.

Although the results obtained with the forced pruning technique may be interesting from the point of view of the characteristics of the berries, it should be taken into account that its application would only be recommended for areas that allow the development cycles of the vine to be extended and those where, after the harvest date, there are sufficient climatic conditions for the vine to continue its activity for a few more months. On the other hand, the varieties should not be long-cycle varieties, which naturally ripen during the coolest periods of the season. It is also necessary to previously evaluate the optimum moment for forced pruning in each variety, so that the fruiting buds are already developed, but with enough time to complete ripening before the leaves start to fall. The application of RI together with crop forcing maintained or improved berry characteristics and production compared to the respective irrigated treatment, so the combined use of both techniques is recommended to improve water use efficiency while maintaining compositional benefits. An important aspect to take into account for the application of this technique is the higher water consumption observed. Although ETo was lower in treatments F1 and F2 during the period analyzed, the volume of water consumed was higher than that in NF (Table 2). This effect was due to the fact that the canopies were more active during the summer period of higher evapotranspiration demand (July to September) with young leaves with a higher photosynthetic rate compared to NF canopies [65, 66]. In addition, the intercepted fraction of photosynthetically active radiation (PAR) was equal or higher in the case of C-F1 compared to C-NF (data not shown), which increases the water requirements of the vine [47]. All this indicates that in areas with a lack of rainfall during the summer months, this technique is probably not recommended in rainfed conditions.

## 5. Conclusions

Under the conditions in which the crop forcing experiment was carried out, it was an effective technique for delaying grape ripening to lower temperature periods, particularly when applied after fruit set. This technique limits vineyard yields but manages to improve phenolic content, anthocyanin content, and total berry acidity, with a lower total soluble solids content, thus restoring the coupling between phenolic and technological ripeness. When crop forcing is carried out, with water limitation during preveraison, the anthocyanins content of the grapes of cv. Tempranillo is increased while improving water use efficiency with respect to the application of crop forcing without water limitations.

#### Abbreviations

- CC: Climate change
- PDO: Protected designation of origin
- DOY: Days of year
- TSS: Total soluble solids
- TA: Titratable acidity
- MAL: Malic acid
- TAR: Tartaric acid
- TPP: Total polyphenol content
- TAN: Total anthocyanin.

#### **Data Availability**

Data are available upon request.

## **Conflicts of Interest**

The authors declare that they have no conflicts of interest.

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## **Supplementary Materials**

Table gathering coefficient of determination (*R*2) and equation between x = Total Soluble Solids (TSS) and y = pH; Titratable Acidity (TA), Malic Acid (MAL), Tartaric Acid (TAR), Total Polyphenols (TPP), and Total Anthocyanins (TAN), in year 2017, 2018, and 2019 (Table S1) and photographs of vines of treatments (a) C, (b) *F*1, and (c) *F*2 on 11th August 2017. (*Supplementary Materials*)

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