## **ORIGINAL PAPER**

# Spatial and temporal variability of cv. Tempranillo phenology and grape quality within the Ribera del Duero DO (Spain) and relationships with climate

M. C. Ramos<sup>1</sup> · G. V. Jones<sup>2</sup> · J. Yuste<sup>3</sup>

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**Abstract** The aim of this work was to analyze spatial phenology and grape quality variability related to the climatic characteristics within the Ribera del Duero Designation of Origin (DO). Twenty plots planted with cv. Tempranillo and distributed within the DO were analyzed for phenology from 2004 to 2013. Grape quality parameters at ripening (berry weight, sugar content, acidity and pH, and anthocyanins) were analyzed in 26 plots for the period 2003–2013. The relationships between phenology and grape parameters with different climatic variables were confirmed with a multivariate analysis. On average, bud break was April 27th, bloom June 17th, and veraison August 12th. However, phenology during the time period showed high variability, with differences between years of up to 21 days for a phenology stage. The earliest dates were observed in dry years (2005, 2006, and to a lesser degree in 2009) while the later phenology dates occurred in the wettest year of the period (2008). High correlations were found between veraison date and temperature variables as well as with precipitation-evapotranspiration recorded during the bloom-veraison period. These effects tended to be higher in the central part of the DO. Grape quality parameters also showed high variability among the dry and the wet years,

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- M. C. Ramos cramos@macs.udl.es
- Department Environment and Soil Science, University of Lleida, Lleida, Spain
- Department Environmental Studies, South Oregon University, Ashland, OR, USA
- Instituto Tecnológico Agrario de Castilla y León, Valladolid, Spain

and the influence of extreme temperatures on color development as well as the effect of available water on acidity were observed.

**Keywords** Climate variability and trends · Cluster analysis · Phenological dates · Ripening · River terraces and hillslopes · Water deficit

#### Introduction

Vineyards in the Ribera del Duero area (Spain) date back to the Roman period as indicates the Bacchic allegorical Roman mosaic found in Baños de Valdearados (Burgos) and the monastic frieze from Quintanilla de las Viñas (Burgos), The history of viticulture in the Ribera del Duero is strongly tied to landscape, climate, and culture. During the tenth and eleventh centuries, vineyards in the area were consolidated into larger operations and achieved relatively stable production, which became an increasingly important aspect of the economic and cultural development of the area. In the thirteenth century, vines in the Ribera del Duero exceeded the present limits with vines planted at elevations higher than 1000 m a.s.l. (Peñín 1992). However, significant fluctuations in production were observed throughout the centuries. The present DO Ribera del Duero was established in 1982, and from that moment, surface has increased from 6460 ha of vineyards officially registered in 1985 to approximately 21,700 ha in 2013 (Consejo Regulador DO Ribera del Duero 2013) and has become world renowned for being one of the highest quality red-wineproducing regions. The total grape production stands at around 90 million kg, with an average yield that approaches nearly 4500 kg/ha (MAGRAMA 2013). Most vineyards are cultivated under rainfed conditions. For this reason, climate

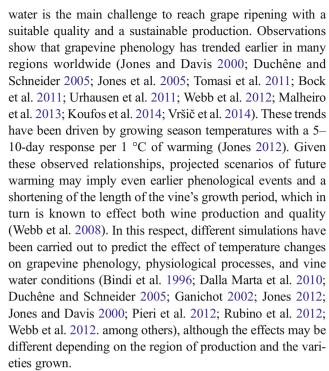


variability, especially that related to precipitation variability, may affect grape development and production.

Tempranillo is the dominant variety planted in the DO, accounting for over 95 % of the surface area. Its expansion has been to the detriment of other traditional varieties in the region such as Grenache (Garnacha Tinta) or Albillo Mayor, and authorized foreign varieties which were introduced some time ago, such as Cabernet Sauvignon, Merlot, and Malbec. The proportion of Tempranillo grapes in the red wines produced in the DO are required to be 85 % or more (www. riberadelduero.es, Regulation of DO), and typically many wines are 100 % Tempranillo. The result is that the DO Ribera del Duero is listed as the world's grape-growing region with largest contribution and importance of Tempranillo in its wines.

The expansion of the vineyards from higher, sloped landscapes (higher than 900 m a.s.l.), to new growing areas has resulted in the location of many newer planted vineyards at lower elevations and more fertile soils typically located between 750 and 800 m a.s.l. The climatic conditions in these areas, as well as the productive potential of the soils, require much greater attention to vine management, including microclimatic aspects affecting health and maturation of grapes. Various "green" interventions have been used in the region in order to control the grape yield (since the DO Regulations establishes a maximum of 7000 kg/ha), namely through cluster thinning, and optimize the vine microclimate through the vegetation management.

Of all of the terroir components, climate is arguably one of the most important on vine growth and grape development. Climate affects grapevine growth and fruit production in many ways. During the winter, grapevines need some dormant chilling to effectively set the latent buds for the coming vintage. During the growing season, grapevines need sustained average daily temperatures above 10 °C to initiate growth followed by sufficient heat accumulation to ripen the fruit. However, during the growth of the berries, temperature extremes can be detrimental inducing transpiration increases, stomatal conductance and photosynthesis reduction, plant stress, premature véraison, berry abscission, enzyme activation, and less flavor development (Coombe 1987; Mullins et al. 1992; Greer and Weedon 2013). Frost occurrence and timing is also significant for grapevines where low spring and fall frost risk is advantageous and where the length of the frost-free season should be 160–200 days or more. Most analyses relating phenology with climate focus mainly on the impacts of temperature on phenological events and on the suitability to different winegrape cultivars (Jones 2007). However, water available and soil water is also important. From a moisture standpoint, grapevines ideally should start off the growing season with adequate soil moisture for initial growth then receive nominal amounts (either naturally or via irrigation) throughout the growing season. But, in drylands, and under irregular rainfall distributions,



Given the importance of wine production in the region and the growth and spread of vineyards to newer locations, the main purpose of this research was to investigate the spatial and temporal variability of phenology and grape quality parameters at maturity within the Ribera del Duero Designation of Origin (Ribera del Duero DO) and its relationship with spatial and temporal climate variability. The study is focuses on the Tempranillo variety, which is the most important variety in the region.

## Materials and methods

## Study area

The Ribera del Duero DO covers approximately 300,000 ha running 115 km east-west along the Duero River, from the provinces of Quintanilla de Onésimo (Valladolid) on the west to San Esteban de Gormaz (Soria) on the east (Fig. 1). Geologically, the Ribera del Duero area is part of the large septentrional plateau formed by a large basement filled with Tertiary deposits, which consist of layers of loamy and sandy ochre and red clays, and middle and low terraces from the Duero River (Quaternary). The main soil types in the Ribera del Duero area are *Typic Xerofluvent* (in the alluvial deposits) and Typic Xerochrept, Calcixerollic Xerochrept, and Calcic Haploxeralf (on the low and middle Duero terraces). The landscapes over the Ribera de Duero have generally gradual to moderate slopes that are oriented to numerous aspects at elevations from about 700 m to more than 1300 m a.s.l. The climate is temperate with dry or temperate summers in the



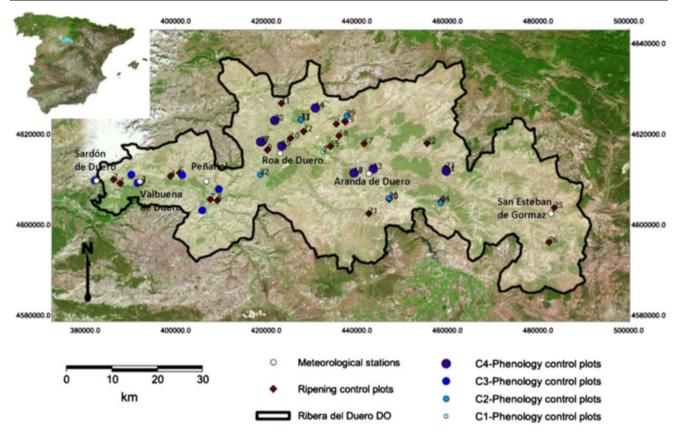


Fig. 1 Meteorological stations and phenology and ripening control plot locations. C1 through C4 represent the clustering of the phenology control plots according to the timing of the main phenological events during the growing season

western portion of the DO area and temperate with a dry summer season in the eastern portion of the DO area. The mean annual temperature ranges between 10.2 and 12.0 °C, with mean maximum temperatures around 18.4 °C and mean minimum temperatures ranging between 4.5 and 5.0 °C. The mean annual precipitation ranges between 413 and 519 mm with the main rainfall periods in April–May and October–November–December. Within this landscape and climate, the vine training system has evolved from a free vegetation shape in bush vines or gobelet, into a vine supported by a vertical trellis system, with both systems commonly used although gobelet is still used the most (Yuste 2008).

## Climate data and analysis

For this analysis, daily temperature and precipitation data for the period 1980–2012 from five meteorological stations belonging to the Spanish AEMET corporation were used: Sardón de Duero (RET; 41.6522° N, 4.7233° W; 723 m a.s.l.); Valbuena de Duero (VD; 41.6397° N, 4.2925° W; 733 m a.s.l.); Roa de Duero (ROA; 41.6969° N, 3.9278° W; 811 m a.s.l.); Aranda de Duero (AD; 41.6714° N, 3.6892° W; 798 m a.s.l.); San Esteban de Gormaz (SEG; 41.5753° N, 3.2080° W; 876 m a.s.l.) (Fig. 1). A shorter series recorded during the last 10 years at Peñafiel (41.5964° N, 4.1186° W;

754 m a.s.l.) were also used. For each meteorological station, the following indices were evaluated:

- Growing season (April–October) temperature [maximum (TGSmax); minimum (TGSmin); mean (TGSm)]
- Number of extremes: number of frost days (FD) and number of days with T>30 °C (NDT30)
- Bioclimatic indices: Winkler index (WI)=Σ((Tmax+Tmin)/2)-10 °C) and Huglin index (HI)=Σ((Tavg-10 °C)+(Tmax-10 °C)/2)×d, where d is a day length factor)
- Daily temperature range (DTR=Tmax-Tmin) during the ripening period (August-September)
- Annual precipitation (referred to the hydrological year October–September) (P<sub>HYD</sub>), growing season (April– October) precipitation (PGS), and precipitation in each phenological stage: [bud break–bloom (PBB), bloom– veraison (PBV), veraison–harvest (PVH)]
- Vine growing season evapotranspiration estimated according to Penman–Monteith equation and using the crop coefficients proposed by Allen et al. (1998) (ETcGS).

Means and standard deviations of each variable were calculated for each location and an ANOVA was used to identify significant differences throughout the Ribera del Duero area.



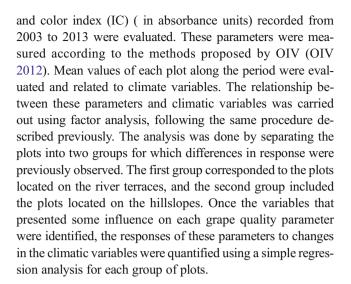
## Phenology data and analysis

Twenty plots distributed throughout the Ribera del Duero area (Fig. 1) and planted to the Tempranillo variety were analyzed for the period 2004–2013 from data provided by the Consejo Regulador of Ribera del Duero DO. Phenology dates (Baggiolini classification) corresponding to the C (bud break), G, I (bloom), K, L, and M (veraison) stages were averaged over each plot and analyzed. In order to characterize the spatial variability, the plots were classified using a hierarchical cluster analysis taking into account the dates referring to different phenological stages (C, G, I, L, and M) across all plots. Among the clustering algorithms, the Ward's minimum variance method was used (Ward 1963). This method calculates the distance between two clusters as the sum of the squares between them added to all of the other observations. All the data were standardized. The number of clusters to be retained was defined by taking into account the agglomeration distance, which measured the intercluster continuity, and the clustering coefficient. A cutoff point was established when the distance between one step and the next was greater than twice the average distance. The average values of each variable for the years included in each cluster were computed.

Due to the differences in phenology dates among the plots included in each cluster, the relationship between climate parameters and phenology was analyzed for each group of plots for which differences in phenology were previously established. A factor analysis including temperature and precipitation indices as well as phenological dates was applied to the different groups of plots. The variables included in the factor analysis were as follows: growing season temperatures (TGSmax, TGSmin, TGSm) and the different bioclimatic indices (WI, HI, DTR, and the number of extremes FD and NdT30) for temperature, and precipitation—evapotranspiration for the growing season (PGS) and for each phenological stage (P-ETcBB, P-ETcBV, P-ETcVH). Each climatic variable was interpolated for the location in which each plot was located using the recorded data in the available meteorological stations. The relationship between these parameters and climatic variables was carried out using factor analysis. Prior to applying the clustering method, all data were standardized to a mean of 0 and standard deviation of 1. In order to improve the orthogonality, a varimax rotation criterion was applied. The first few components were retained, which represented the majority of the variability in the whole of variables.

## Ripening grape quality data and analysis

Twenty-six plots distributed throughout the Ribera del Duero area (Fig. 1) were used for the ripening control analysis. Parameters such as 100 berry weight (g), sugar content (°Baumé), titratable (AcT), and malic (AcM) acids (g L<sup>-1</sup>), total (AntT) and extractable (AntE) anthocyanins (mg L<sup>-1</sup>),



#### Results

#### Climate variability among the analyzed observatories

Table 1 shows the average and standard deviation of temperature variables and bioclimatic indices for each location for the period 1980-2013. There were significant differences in mean TGSmax between the stations located at both extremes of the area (RET and SEG) with the lowest temperatures recorded in the eastern part of the Ribera del Duero area. Significant differences were also observed for minimum temperatures between AD and SEG but no differences were found among the rest of the locations. The mean number of FD revealed high variability from year to year, with a maximum number of days per year of 118 days in 2013 and a minimum of 48 days in 2002. Maximum extreme temperatures also showed significant differences between both extremes of the area with mean number of days per year ranging between 77 days in 1998 and 23.1 days in 2007, and a maximum of 81 days per year. The ROA location exhibited the highest values of warm extremes and the minimum number of frost days. The mean DTR for the ripening period presented significantly higher values in RET than in ROA but showed no statistical differences elsewhere. The differences in the WI among locations (between 1250 and 1311 GDD) were not significant. However, significant differences in the mean HI (between 2026 and 2219 GDD) existed between both extremes of the DO. Similar spatial variations were found for crop evapotranspiration.

Significant increases in minimum temperature were observed during the period 1980–2013 at the stations located at both extremes of the DO, but not in the central part of the region (Table 1), along with a decreasing number of frost days. The maximum temperature trends during the growing season were not significant in any location. The WI and HI increased



Table 1 Mean value±standard deviation and trends (change ratio per year) for temperature and precipitation variables for each location during the period 1980–2012

Location	Sardón de Duero - Retuerta (RET)	Valbuena de Duero (VD)	Roa de Duero (ROA)	Aranda de Duero (AD)	San Esteban de Gormaz (SEG)
TGSmax (°C)	25.49±1.16 a	25.13±1.07 ab	25.23±1.27 ab	25.25±0.89 ab	24.76±098 b
	(0.0254)	(0.0288)	(0.011)	(0.006)	(0.0316)
TGSmin (°C)	$8.79 \pm 0.86 \ ab$	8.99±1.17 ab	$8.83 \pm 0.61 \ ab$	$8.63\pm0.80~a$	9.11±0.83 b
	(0.0455)	(0.0231)	(0.011)	(0.0128)	(0.0262)
TGSm (°C)	17.12±0.81 a	17.08±1.02 a	$17.02 \pm 0.77$ a	16.95±0.74 a	$16.89 \pm 0.78$ a
	(0.0332)	(0.0288)	ns	(0.011)	(0.0326)
DTR (°C)	18.1±1.9 a	17.1±1.3 b	16.8±1.4 b	17.7±1.4 ab	17.7±1.4 ab
	(-0.044)	(-0.288)	(-0.317)	(-0.31)	ns
FD (days)	85.8±17.8 a	$76.9 \pm 19.6 \text{ ab}$	68.8±16.9 b	87.4±19.9 ab	82.7±16.7 ab
	(-0.523)	(0.597)	(-0.534)	(-0.2747)	(-0.300)
NDT30 (days)	52.9±13.7 a	47.8±14.7 ab	51.8±12.4 ab	$48.5 \pm 12.8 \text{ ab}$	43.1±10.8 b
	ns	ns	ns	ns	ns
WI (gdd)	$1311 \pm 173$	1299±219	$1287 \pm 163$	$1272 \pm 152$	$1250 \pm 155$
	(6.193)	(4.963)	(1.865)	(2.700)	(5.100)
HI (gdd)	2119±181 a	2081±197 ab	$2084 \pm 192 \ ab$	$2076 \pm 144 \ ab$	2026±162 b
	(6.949)	(5.957)	(3.065)	(2.746)	(6.733)
ETcGS (mm)	598.1±66.5 a	580.2±23.2 ab	588.0±43.4 ab	595.4±64.7 ab	565.9±25.8 b
	(1.854)	(0.598)	(0.648)	(1.700)	(0.644)
$P_{HYD}$ (mm)	$403.5 \pm 120.2$ a	345.9±100.8 b	417.8±95.6 a	422.1±94.5 a	472.2±103.8 c
PGS (mm)	$178.9 \pm 73.4$ a	144.9±57.2 b	$184.3 \pm 62.4a$	199.4±63.1 a	231.9±76.6c
	(-1.10)	(1.296)	(-1.208)	(-1.728)	ns
PBB (mm)	104.2±55.3 ab	83.1±47.2 a	101.3±49.4 ab	110.9±46.6 ab	133.9±56.1 b
	ns	ns	ns	ns	ns
PBV (mm)	38.1±28.1 ab	30.6±22.4 a	41.9±26.5 ab	48.1±36.9 ab	53.3±33.6 b
	(-0.350)	(0.3265)	(-0.466)	ns	(-0.43)
PVH (mm)	36.5±23.5 ab	31.2±19.9 a	41.0±21.9 ab	40.4±22.9 ab	44.7±28.1 b
	(-0.734)	(0.505)	(-0.666)	(-0.855)	(-0.44)

Different letters mean significant differences at 95 % level. Numbers in parenthesis indicate significant trends at  $\alpha$ <0.10, while numbers in italics indicate significant trends at  $\alpha$ <0.05 (see "Materials and methods" section for variable descriptions and formulations)

accordingly with significant trends in RET, VD, and SEG but not in the rest of locations. Statistics for average annual and growing season precipitation for each location are also shown in Table 1. During the period of study, high year to year variability in precipitation was recorded. For annual precipitation, the mean values ranged between 300 mm in 2005 and 595 mm in 1998, with differences among years greater than 400 mm. Precipitation recorded during the growing season ranged between 79 mm in 2005 and 312.9 mm in 2008, which on average represents between 43 and 49 % of annual precipitation. Significant differences were found between the extremes of the Ribera del Duero area and also between VD and the rest of locations. These lower values were also confirmed for a shorter series recorded in Peñafiel (located 17.5 km southwest of ROA) for the last 10 years.

Within the growing period, more than 50 % of the precipitation took place during the bud break-bloom period.

Precipitation during the bloom–veraison and veraison–harvest periods is relatively low (about 20–24 % of growing season precipitation in each period). Significant differences were found between growing season precipitation recorded in some stations located at both extremes of the DO (western and eastern parts).

During the period analyzed, years with different characteristics were recorded. Differences greater than 2 °C in TmaxGS and up to 5 °C in TminGS were observed between years, with similar differences in NDT30 values. Years from the 2000s were among the warmest years of the series (2000, 2003, 2005, 2006, 2010, 2011, and 2012), while 2007 and 2008 were within the coolest years. Regarding precipitation, large differences were also observed with PGS ranging between less than 100 mm and more than 300 mm. Years 2007 and 2008 were among the wettest years, while the rest of years in the 2000s were among the driest years of the series (with PGS<



155 mm). In particular, years 2002, 2005, 2009, and 2012 were very dry.

## Phenology variability and climate relationships

Table 2 shows the average dates for different phenological stages recorded during the period 2004–2013. On average bud break stage occurred on April 27 while bloom averaged June 17 and veraison averaged August 12. During this period of study, the phenology exhibited high variability with differences between plots and between years. Differences in the dates between years of up to 21 days were observed with the earliest dates observed in dry years (2005, 2006, and to a lesser degree in 2009). On the other hand, later dates occurred in the wetter years, especially in 2008.

Based on the mean phenological dates, the 20 plots were classified into three groups plus one isolated plot in one cluster (cluster 1), which was linked to cluster 3 (Fig. 2). Average differences of 2–3 days for all dates between groups were observed (Table 2). The plots included in group 2 are all located in the western extreme of the Ribera del Duero DO. These plots typically experienced earlier phenology, particularly with stages G through M. For the remainder plots, there were differences between locations on the terraces of the river (group 3) and those located at higher elevations on the hillslopes (group 4) which exhibited a slight delay in phenology. The analyzed years represented the situations previously established according to temperature and precipitation: dry

and hot years such as 2003, 2005, 2009, and 2012; wet years such as 2007 and 2008 (and also 2013) and years with intermediate conditions (such as 2004, 2010, 2011, and 2012).

The relationship between phenology and the different climate variables were confirmed with a multivariate analysis. As it was shown in Table 2, the differences in phenology among years were smaller in the earlier stages than in the later stages of the growing season (veraison). For this reason, the relationships between M stage (veraison) dates and climate parameters were evaluated for the three groups of plots established in the previous analysis. For each of the three areas, three factors were retained, which described more than 85 % of variance. The results are shown in Table 3. F1 was associated with maximum temperatures, WI and HI, while the other two factors (F2 and F3) were largely associated with water availability (precipitation-evapotranspiration) in each phenological stage, FD and DTR. However, the variables included in each group were not the same for the three areas. For the western part of the DO, veraison dates were only influenced by temperature variables with the results indicating an earlier veraison when all climatic variables associated with factor F1 increased. However, for the plots located in the central part of the DO, veraison dates were also influenced by water accumulated in the soil during the bloom-veraison period (F1) and during the dormant period (F3) and by the decrease in frost days (F3). The effects of temperature were greater in the plots located on the river terraces, while the effect of available water was higher on the plots located at

Table 2 Mean date±standard deviations of different phenological stages for each year averaged over all plots and for the four groups of plots established in the cluster analysis

	Stage C bud break		Stage G		Stage I bloom		Stage L	Stage L Mean±std (days)		Stage M veraison		
	Mean±std (days)	n±std (days) Me		Mean±std (days)		Mean±std (days)				Mean±std (days)		
2004	29-Apr±4.8		26-May±3.2		20-Jun±3.8		13-Jul±6.6		11-Aug±4.6			
2005	30-Apr±2.6		16-May±4.6		8-Jun±2.9		6-Jul±3.0		3-Aug±1.9			
2006	25-Apr±3.7		12-May±4.9	12-May±4.9		8-Jun±3.8		18-Jul±6.0		2-Aug±2.5		
2007	1-May ± 2.9		20-May±7.2		22-Jun±6.3		22-Jul±1.7		21-Aug±2.7			
2008	29-Apr±5.9		27-May±4.5		28-Jun±3.6		11-Jul±3.5		23-Aug±2.1			
2009	5-May±4.0		21-May±4.6		15-Jun±2.7		23-Jul±4.1		9-Aug±3.6			
2010	27-Apr±1.8		20-May±7.0		21-Jun±4.4		6-Jul±4.6		18-Aug±4.1			
2011	15-Apr±4.4		8-May±5.6		7-Jun±2.7		5-Jul±3.3		9-Aug±3.5			
2012	4-May ± 6.2		29-May±2.6		18-Jun±2.8		16-Jul±4.5		13-Aug±5.6			
2013	20-Apr±5.1		29-May±7.0		26-Jun±4.2		25-Jul±4.5		14-Ago±6.2			
2004-2013	27-Apr±5.9		20-May±7.1		$17$ -Jun $\pm 7.3$		$14$ -Jul $\pm 7.1$		12-Aug±7.4			
Cluster 1	30-Apr		20-May		12-Jun		12-Jul		11-Aug			
Cluster 2	28-Apr±2.3	a	20-May±2.2	a	15-Jun±1.2	a	15-Jul±0.8	a	13-Aug±2.4	i		
Cluster 3	26-Apr±2.1	a	17-May±1.6	a	$13$ -Jun $\pm 1.4$	b	11-Jul±1.4	b	10-Aug±1.0	1		
Cluster 4	26-Apr±2.5	a	16-May±1.5	b	14-Jun±1.5	ab	13-Jul±1.9	c	10-Aug±0.5	1		

Different letters indicate significant differences between groups (see "Materials and methods" section for description of phenological events and measurement system)



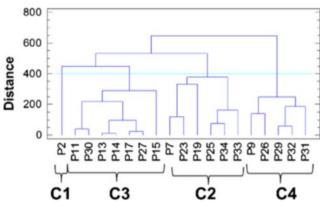


Fig. 2 Plot classification based on phenology dates of veraison using cluster analysis

higher elevations. The analysis of the influence of available water (P-ETc) on phenology showed that water deficits during the bloom–veraison period had higher effects on dates in the central part of the DO. The plot by plot analysis showed an advance of between 8 and 14 days for 100 mm of water deficit on the river terraces (with an average values of 11.9 days per 100 mm), and between 12 and 20 days for 100 mm water deficit in the plots located on the hillslopes (with and average values of 12.8 days for 100 mm of deficit).

# Grape quality variability and climate relationships

Grape quality parameters varied among years during the analyzed period (2003–2013) and among plots in each year, although differences between them were only significant for a reduced number of plots when all years were considered

together. At maturity, the overall average pH was 3.60, ranging between 3.18 and 3.91. The lower values were recorded in most plots during the years 2004, 2007, 2008, and 2013. In these years, titratable acidity recorded the highest values, while the lowest were recorded in years 2005, 2006, 2009, 2011, and 2012. Malic acid concentration varied between 1.37 and 5.45 g  $L^{-1}$ , with an average value of 3.48 g  $L^{-1}$ . The lower values were also recorded in 2005, 2006, 2011, and 2012, while the highest values were recorded in 2004, 2007, 2008, and 2010. The sugar content (°Baumé values) ranged between 10.6 and 15.0°, with an average value of 13.03°. Similarly, the highest values were recorded in 2007, 2008, and 2013. Berry weights did not show significant differences between plots. Similarly, results related to anthocyanin values presented differences between the same groups of years, but average values between plots were only significantly different for few plots. The lowest values for AntT were recorded in the years 2005, 2006, and 2012, and the highest values on 2007, 2008, 2010, and 2011. For AntE, the lowest levels were recorded in 2003, 2005, 2006, and 2009 and the highest values corresponded to 2007, 2008, 2010, and 2013. Similar differences were observed related to CI.

The factor analysis carried out with climate and grape quality parameters confirmed the influence of climate variables on grape ripening variables (Table 4). For the plots located on the terraces, the CI values exhibited positive correlations with water availability and negative correlations with the extreme temperature variables. However, anthocyanins did not exhibit any correlations with climate variables. Acidity parameters (AcT and AcM) and berry weight showed positive correlation with water availability in the earlier stages (bud break—

Table 3 Factor analysis of veraison dates and climatic variables for three areas within the DO defined by the cluster analysis and shown in Fig. 2b

	A			В			C			
	F1	F2	F3	F1	F2	F3	F1	F2	F3	
Stage M	-0.877	0.064	0.051	-0.701	0.336	0.403	-0.687	0.490	0.290	
TGSmax (°C)	0.984	0.042	0.022	0.886	0.339	0.005	0.890	0.332	-0.020	
TGSMin (°C)	0.645	0.226	0.652	0.732	-0.234	0.489	0.765	-0.184	0.433	
TGSm (°C)	0.897	0.149	0.372	0.934	0.278	0.080	0.937	0.273	0.034	
DTR (°C)	0.008	0.109	-0.690	0.257	0.848	0.191	0.242	0.834	0.183	
FD (days)	-0.238	-0.259	-0.636	-0.035	0.003	-0.722	0.004	0.084	-0.780	
NDT30 (days)	0.603	-0.019	0.512	0.807	0.220	0.159	0.864	0.114	0.161	
WI (gdd)	0.895	0.077	0.389	0.950	0.205	0.047	0.952	0.205	0.017	
HI (gdd)	0.972	0.098	0.164	0.907	0.302	0.069	0.911	0.296	0.034	
P-ETcBB (mm)	0.143	0.950	0.007	-0.080	-0.041	-0.774	-0.096	-0.124	-0.736	
P-ETcBV (mm)	-0.244	-0.927	0.021	-0.725	-0.240	0.511	-0.758	-0.232	0.481	
P-ETcVH (mm)	-0.283	0.870	0.168	-0.237	-0.864	0.213	-0.272	-0.822	0.269	
Variance (%)	50.91	21.42	10.14	51.60	15.88	13.21	52.45	15.10	13.80	

Numbers in italics indicate significant loadings (see "Materials and methods" section for variable descriptions and formulations) *A* western part of DO, *B* central part of the DO on river terraces, *C* central part of the DO on hillslopes



**Table 4** Results of factor analysis of climate and grape quality parameters, period 2003–2013, and plots located on river terraces (Fi<sub>r</sub>) and on hillslopes (Fi<sub>h</sub>)

	F1r	F2r	F3r	F4r	F5r	F6r	F1h	F2h	F3h	F4h	F5h	F6h
рН	-0.844	0.003	0.131	-0.159	-0.003	-0.016	0.373	-0.084	0.366	-0.648	0.246	0.138
$AcT (g L^{-1})$	0.849	0.171	-0.168	0.273	0.059	0.031	-0.461	0.437	0.006	0.607	0.211	-0.079
$AcM (g L^{-1})$	0.728	0.354	-0.112	-0.035	0.077	0.153	-0.288	0.402	0.319	0.517	-0.033	-0.064
°Beaumé (°)	-0.564	-0.223	-0.081	0.074	0.455	0.230	0.365	-0.244	0.565	-0.336	0.199	0.087
AntT (mg $L^{-1}$ )	0.283	-0.014	-0.005	0.070	0.176	0.879	-0.056	0.013	0.935	0.134	-0.033	-0.036
AntE (mg $L^{-1}$ )	-0.053	0.000	0.038	0.002	0.017	0.942	-0.083	0.062	0.903	0.012	-0.032	0.005
CI (Abs.units)	0.110	0.015	-0.200	0.826	0.059	0.012	-0.197	0.286	0.285	-0.031	0.803	-0.067
100BW (g)	0.542	0.023	0.008	-0.053	-0.306	0.224	-0.020	0.044	-0.025	0.717	-0.036	0.136
TGSmax (°C)	-0.140	-0.060	0.956	-0.095	0.144	0.003	0.853	-0.343	-0.054	-0.054	0.038	0.130
TGSmin (°C)	0.050	-0.092	-0.968	-0.073	0.014	-0.015	-0.132	-0.020	0.097	0.029	0.040	0.932
TGSm (°C)	-0.088	-0.070	0.966	-0.078	0.014	0.021	0.158	-0.118	-0.247	0.006	-0.134	0.315
DTR (°C)	-0.232	-0.431	0.048	0.368	-0.098	-0.071	0.161	-0.371	-0.326	-0.075	0.751	0.062
FD (days)	-0.128	0.046	-0.195	-0.046	-0.751	-0.255	-0.300	0.258	-0.338	-0.606	-0.017	0.069
NDT25 (days)	-0.334	-0.472	0.005	-0.440	0.325	-0.160	0.652	-0.288	-0.228	-0.297	0.201	0.347
NDT30 (days)	0.054	-0.265	-0.046	-0.688	0.282	-0.068	0.749	-0.137	-0.013	-0.056	0.123	0.296
WI (gdd)	-0.170	-0.054	-0.013	-0.590	0.741	-0.045	0.904	-0.076	0.013	-0.054	-0.176	-0.319
HI (gdd)	-0.210	-0.070	-0.019	-0.537	0.724	-0.034	0.900	-0.070	0.018	-0.031	-0.155	-0.363
P_Etc_BB (mm)	0.441	0.472	0.101	0.662	-0.075	-0.017	-0.448	0.691	-0.016	0.365	0.210	-0.124
P_Etc_BV (mm)	0.013	0.850	0.043	-0.015	-0.042	-0.123	-0.105	0.914	-0.025	-0.060	0.014	-0.146
P_Etc_VH (mm)	0.107	0.818	-0.101	0.200	-0.095	0.078	-0.093	0.921	0.057	0.067	-0.097	0.080
P_Etc_Total (mm)	-0.197	0.902	-0.024	0.062	0.004	-0.035	-0.197	0.902	-0.024	0.062	0.004	-0.035
Variance (%)	23.435	18.845	13.74	8.368	6.895	4.971	27.734	13.964	11.277	9.376	7.559	4.971

Numbers in italics indicate significant loadings (see "Materials and methods" section for variable descriptions and formulations)

bloom), but no correlation was found with most temperature variables. Sugar content (°Baumé) correlated positively with the HI and WI indexes and negatively with water availability during bud break—bloom period.

In the plots located on the hillslopes, the acidity parameters (AcT and AcM) and CI were correlated with the available water during bud break—bloom period, while sugar content (°Baumé values) was inversely correlated with this parameter. Temperature variables (Tmax, WI, HI, and the number of days with  $T>30\,$  °C) correlated positively with sugar content (°Baumé values) and negatively with CI and acidity. DTR exhibited a strong correlation with CI. However, anthocyanins did not reveal any correlations with climate parameters.

The average response of each group of plots to changes in each parameter is shown in Table 5. Water availability for the crop had greater impacts on the plots located on the hillslopes than in the plots on river terraces. From this data, 100-mm water deficit typically reduces CI between 1.00 and 0.52 absorbance units, respectively, in the two groups of plots; changes range between 0.96 and 0.34 g L<sup>-1</sup> for AcT and between 0.56 and 0.17 g L<sup>-1</sup> for AcM; and berry weight for the same change in water availability is commonly five times higher in the hillslopes compared to the terraces. However, the increase of temperature and the corresponding increase in WI and HI

had greater effect on the vine's ripening characteristics when located on the river terraces for some parameters. For 1 °C warmer growing season temperature, CI may change in -0.73 and -0.27 units, respectively in both situations; AcT may change between -0.453 and -0.287 g  $L^{-1};$  and AcM may change between -0.234 and -0.252 g  $L^{-1}.$  On the other hand, berry weights and sugar content (°Baumé) may suffer changes of the same magnitude in all areas related to changes in temperature.

## Discussion

The five long series of climatic data analyzed confirmed that both maximum and minimum temperatures had significant differences between the central portion and both east and west portions of the region. The differences in mean temperatures, frost risk and precipitation found in this analysis are in agreement with the general traits commented by Gómez-Miguel (2003). Despite the high variability from year to year, increasing trends in minimum temperatures were observed in the region as well as some changes in the temporal rainfall distribution. Changes were greater at both extremes (east and west) of the area than in the stations located in the central portion of



Table 5 Response of each grape quality parameter to changes in climatic parameters for plots located in the river terraces and on the hillslopes

Terraces					
	100-mm deficit BV	1 °C ΔTmaxGS	100GDD∆WI	100GDD ΔHI	1 °C ∆DTR
CI (Abs.units)	0.52	-0.753	-0.51	-0.49	
$AcT (g L^{-1})$	0.34	-0.453	-0.20	-0.24	
$AcM (g L^{-1})$	0.17	-0.234	-0.06	0.10	
рН	-0.093	0.038	0.02	0.02	
100BW (g)	2.1	-2.90	-2.38	-1.23	
°B (°)	-0.11	0.338	0.19	0.2	
Hillslopes					
	100-mm deficit BB	1 °C ΔTmaxGS	100GDD∆WI	100GDD ΔHI	1 °C ΔDTR
CI (Abs.units)	1.03	-0.269	-0.35	-0.21	0.619
$AcT (g L^{-1})$	0.959	-0.287	-0.36	-0.296	
$AcM (g L^{-1})$	0.556	-0.252	-0.20	-0.167	
pН	-0.083	0.047	0.03	0.03	
100BW (g)	10.23	-3.22	-2.04	-1.64	
°B (°)	-0.407	0.29	0.18	0.157	

100-mm deficit BB: water deficits of 100 mm during the bud break-bloom period; 100-mm deficit BV: water deficits of 100 mm during the bloom-veraison period; 1 °C  $\Delta$ TmaxGS: increase of 1 °C on TMaxG growing season; 1 °C  $\Delta$ DRT: increase of 1 °C on daily range temperature; 100DDG $\Delta$ WI: increase of 100 degree days on Winkler index; 100DDG $\Delta$ HI: increase of 100 degree days on Huglin index (see "Materials and methods" section for variable descriptions and formulations)

the area. The majority of the years with the greatest temperature and greater water deficits were found to occur in the last decade. The average increase in temperatures (about 0.03 °C per year for the last 30 years) as well as the changes observed in the bioclimatic indexes (WI and HI) in some locations are in agreement with observations in other viticultural areas of Spain (Ramos et al. 2008; Lorenzo et al. 2013) and in other regions around the world (Duchêne and Schneider 2005; Jones and Goodrich 2008; Makra et al. 2009; Santos et al. 2012; Schultze et al. 2014, among others).

The variability in the climate during this period gave rise to clear differences in phenology within the area and also between years. Differences of up to 21 days in a given phenological stage were found between the driest and the wettest years and up to 7 days between the hottest and coolest years with similar precipitation. For this study, spatial differences were also seen between the western, central, and eastern portions of the Ribera del Duero area, with the western locations generally earlier (2 or 3 days on average for each phenological stage) driven by warmer climatic conditions.

The results obtained in this study confirmed the relationships between phenology and all temperature variables, but in particular with Tmax, WI and HI indexes, and the number of extremes. This result agrees with the advance of harvest dates and dates of other phenological stages with increasing temperatures found by other authors in other areas. The IPCC (2007) reported an advance between 1 and 12 days for every 1 °C increase in spring temperature, with average values

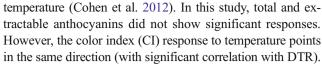
ranging between 2.5 and 6 days per °C. In vineyard areas, similar changes in phenology or in harvest dates have been observed. Jones et al. 2005 indicated earlier events (6–18 days) with shorter intervals between events (4–14 days) across most regions in Europe. Bock et al (2011) found that the phenology of grapevines in Lower Franconia has tended toward earlier occurrence with a shortening of phenological intervals and pointed out that grapevines were most influenced by mean maximum temperatures preceding the event, whereas precipitation and sunshine appeared less important. García-Mozo et al (2010) for southern Spain indicated a trend toward earlier foliation, flowering, and fruit ripening. Malheriro et al (2013) in Portugal also confirmed the role of temperature on phenology, and in particular, the accumulated thermal effects in phenological timing with flowering as the most sensitive phase. Urhausen et al. (2011) in the Moselle viticultural area found significant changes in vine phenology, in particular budburst date and flowering events occurred earlier by about 2 weeks, which were related to accumulated degree days in the previous period. Duchêne and Schneider (2005) found that harvest in Alsace (eastern France) was 2 weeks earlier in 2002 than in 1972, which represents a period during which temperature increased by 1.8 °C. Vršič et a (2013) in Slovenia found trends toward earlier harvest maturity of 12–25 days based on the data maturity associated with the content of soluble solids and total acidity, which were associated to the 1.2-1.8 °C temperature increase in the last 30 years in the region. Koufos et al (2014) in Greece also confirmed earlier harvest,



mainly driven by changes in maximum and minimum temperatures, with the areas with later ripening varieties being less sensitive to climate changes.

It has also been observed that phenology was also related to water deficits in some specific stages. The effects were not uniform in all Ribera del Duero areas. The plots located in the hillslopes, where water deficits are probably greater than on the river terraces, water deficits during bud break to bloom and during bloom to veraison periods had significant influences on phenology. However, delays in phenology were related to water deficits during bloom to veraison and veraison to harvest periods. In this respect, less attention has been paid to the effects of water availability on grape development. Nevertheless, Webb et al. (2012) highlight concomitant warming and decline in soil water content as the main driving factor of ripening trends. The results observed in this study are in agreement with those found in other Spanish viticultural areas located in NE Spain that were related to water impacts (Camps and Ramos 2012) and also with those observed by Castellarin et al. (2007) in ripening acceleration. In relation to grape quality parameters, differences between years were found, which are likely associated with different temperature and precipitation characteristics. For example, the analyzed grape quality parameters (berry weight, sugar content, acidity, and anthocyanins) exhibited differences between the same groups of years, which had in common different precipitation characteristics. While 2005, 2006, 2009, 2011, and 2012 were years with low precipitation, 2004, 2007, 2008, and 2013 were the wettest of the series. Differences in water availability were also affected by differences in temperature. Years 2003, 2005, 2006, and 2009 were the hottest years of the last decades while 2007 and 2008 were more similar with the cooler years recorded during the 1980s.

When the grape quality values corresponding to each plot were analyzed within these two groups, less variability was observed for each plot in the wet years than in the dry years for all acidity parameters (AcT, AcM, pH) and for berry weights. However, the differences were opposite for sugar content (°Baumé values) and anthocyanins. Higher significant differences between plots were revealed during the dry years than when all years were considered together. In addition, differences among plots within the Ribera del Duero DO were observed. In most years, pH values were greater in the western portion of the DO than in the center or in the eastern portion. Similarly, sugar content and anthocyanins were also greater in the western than in the eastern portion of the DO. It is known that grapes grown in warmer climates have lower acidity than grapes grown in cooler climates. On the other hand, the warmer the climate the higher the sugar content of the grapes, so that alcohol degree will be higher The impact of increasing temperature on anthocyanin accumulation in grape skins has been indicated by some authors (Spayd et al. 2002; Tarara et al. 2008), and in particular, the effects of changes in day and night



In addition, water deficits could affect berry weight and anthocyanin accumulation in grapes. In this respect, Downey et al. (2006) indicates that water deficits are related to berry size reduction and changes in the ratio of skin/berry weight, which would affect phenolic concentrations. Some authors have shown that water deficits increased anthocyanin and procianidin concentrations in berries (Nadal and Arola 1995; Ojeda et al. 2002; Roby and Matthews 2004; Castellarin et al. 2007), while other authors did not find significant effects of water deficits on the accumulation of these compounds (Kennedy et al. 2002). In this study, berry weights were related to water availability during the bud break-bloom period. However, the analyzed total and extractable anthocyanins did not show relationships with climate parameters while CI exhibited an opposite trend to that observed by Castellarin et al. (2007). These authors reported that water deficits promote higher concentrations of anthocyanins in red wine grapes and their wines, but also alter the timing of particular aspects of the ripening process.

## **Conclusions**

Strong relationships between climate, available water, and phenology have been documented in this research in the Ribera del Duero DO. The results show the important role of warmer temperatures and bud break to bloom and bloom to veraison period water deficits, producing advanced phenology in all areas, and most notably in the central portion of the Ribera del Duero DO. The relationship between climate and grape quality has also been shown along with the parameters that most affect acidity retention and color development in each area throughout the DO. Understanding relationships between climate and grapevine phenology over a region is useful to continually assess the role that climate variability and change play in vine growth, fruit production, and wine quality.

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