

## Sensory profile of wines produced from disease-resistant grapevine varieties grown in the

### Mediterranean area

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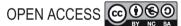
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**ORIGINAL ARTICLE** 

## **Abstract**

The transition to a sustainable viticulture is vital to address environmental issues such as the use of agrochemicals and greenhouse gas emissions. In viticulture, the use of disease-resistant varieties (PIWI) offers a promising solution because of their lower requirement for fungicides treatments, but little is known about their qualitative performance in Mediterranean environments. This study evaluated the quality of wines produced from Merlot Kanthus and Merlot Khorus varieties compared to their parental Merlot N. cultivar in Salento, an area of the Apulia region in southern Italy, characterized by a hot and dry climate. Two different sensory analyses, namely, an affective and a quantitative descriptive test, were performed. Resistant varieties tend to outperform Merlot N. in sensory preferences, acidity, and color, achieving high quality in the Apulian terroir. These results prove the potential value of resistant varieties like Merlot Kanthus and Merlot Khorus to produce high-quality wines in Mediterranean regions and to support a wider acceptance and adoption of PIWI varieties in Italian regions which currently restrict their use.

Keywords: Merlot Kanthus; Merlot Khorus; PIWI; Salento; Southern Italy; Wine sustainability

## Introduction

Global wine production must face the necessity to adopt more sustainable practices to counter environmental concerns such as the use of agrochemicals and greenhouse gas emission (IPCC, 2023; Mailly *et al.*, 2017; Ponstein *et al.*, 2019). Several strategies have been put in place at the European level to mitigate the effects of climate change. For instance, the Farm To Fork strategy (European Commission, 2020) outlines a comprehensive

agenda for a sustainable transformation of the food system including a 50% reduction of chemical and hazardous pesticides use by 2030 and total climate neutrality, that is, achieving net-zero greenhouse gas emission (Capros *et al.*, 2019) by 2050. In order to achieve the set goals, concrete regulatory actions are planned, including a reform of the legal framework for Geographical Indications (GIs) (Reinhardt and Ambrogio, 2023). In Italy, production regulations in the wine sector establish the standards to be followed to produce wines that aim

to obtain recognized GIs, defining essential criteria such as production area, permitted grape varieties, cultivation and winemaking techniques, organoleptic characteristics, and minimum alcoholic strength. Such standards are directly connected to the notion of "terroir effect," which stands for the relationship between the characteristics of an agricultural product (e.g., quality, taste, style) and its geographic origin. A multitude of factors including climatic and landscape conditions as a whole, soil and water availability, rootstock-scion combination, as well as human intervention take part in the definition of terroir as a cultivated ecosystem (Van Leeuwen, 2010). This concept is frequently at the base of the hierarchy of high-quality wines (Van Leeuwen and Seguin, 2006), as terroir components are paramount in the determination of grape and wine worth (Van Leeuwen, 2010).

Long-term strategies to reduce treatments involving spraying fungicides rely on the choice of proper terroirs and proper cultural practices, as well as on breeding efforts aimed at the generation of varieties that can be managed more sustainably. This last approach is considered the most promising tool and relies on intra- and inter-specific controlled hybridization with naturally occurring wild Vitis species from Asia and America with high degrees of disease resistance (Daldoul et al., 2020). As a result, obtaining grapevine resistant to fungal diseases (also called "PIWI," a German abbreviation of "Pilswiederstangfähige") has been proven possible (Bavaresco and Squeri, 2022; Foria et al., 2019; Ricciardi et al., 2024; De Rosso et al., 2023). Resistant varieties were obtained by crossing Vitis vinifera and other complex introgression lines of Vitis spp. and have been selected for their resistance to Plasmopara viticola and Erisyphe necator, as well as their high quality of grapes and wine (Pedneault and Provost, 2016; Teissedre, 2018). The deployment of resistant varieties in the vineyard should increase in the forthcoming years, confirming its role as a key strategy toward a sustainable viticulture (Miclot et al., 2022). On the other hand, Nesselhauf et al. (2019) noted that, despite the environmental credentials of resistant varieties, such as the reduction in pesticide usage and carbon emission associated with their production, their use could represent a commercial disadvantage. This is because of the implementation of wine names unknown to consumers (Fahey and Englefield, 2018) or unfamiliar and therefore unattractive sensory profiles sometimes characterized by "foxy" aromas. Several studies showed that the quality of wines obtained from resistant varieties is generally rated as equivalent to that of V. vinifera (Pedneault et al., 2012; Van Der Meer et al., 2010). For example, Celotti et al. (2020) compared two wines produced from resistant varieties with 12 wines derived from V. vinifera, showing that wines made from PIWIs exhibit a similar organoleptic profile to those originated from traditionally cultivated grapevines, supporting the use of disease-resistant grapevine varieties in accordance with the identity of the GIs.

The cultivation of resistant grapevines and the commercialization of the resulting wines are regulated by the European Union (EU), which permits the use of designations of origin for products made from both *V. vinifera* varieties and those originating from a cross with other species of the genus *Vitis* (European Parliament, 2021). Similarly, the Italian Ministry of Agriculture, Food and Forestry (MIPAAF) integrated resistant grape varieties into the National Vine Variety Register (Ministero delle Politiche Agricole, Alimentari e Forestali, 2015). Several Italian regions have authorized the cultivation of the disease-resistant grapevine varieties on their soil, such as Friuli-Venezia Giulia (Regione Friuli-Venezia Giulia, 2003), while others, such as Apulia region, still prefer a more conservative approach (Regione Puglia, 2003).

On these premises, this work aimed to evaluate the quality of wines produced from two resistant varieties, Merlot Kanthus and Merlot Khorus, compared to wine produced from their shared and internationally acclaimed parent Merlot N. The employment of these genotypes was assessed in the Southern Italian region of Apulia, and specifically in the area of Salento, characterized by a hot and dry summer climate, mild winters with rainfall not exceeding 500–600 mm/year, and temperatures rarely falling below 0°C (Piarulli *et al.*, 2024). This investigation provides precious insight into the possibility of a wider application of disease-resistant grapevine varieties in Mediterranean regions.

#### **Materials and Methods**

### Experimental site and setup

The study was implemented in 2022 at the "Cantine Due Palme" Cooperative Society (Cellino San Marco, Apulia, Italy) involving the comparison of two Vitis spp. Diseaseresistant varieties, namely, Merlot Khorus and Merlot Kanthus, and traditionally cultivated Merlot N. (40° 22' N, 17° 27' E), all grafted on Kober 5BB (Figure 1). The distance between the vineyard with Merlot Khorus and Merlot Kanthus and the vineyard with Merlot N. was 9.30 km. The soil had about 55 g kg<sup>-1</sup> of skeleton, 61% of sand, 26% of silt, and 13% of clay as described by Blanco et al. (2024). Each vineyard was planted in 2012 with  $2.2 \text{ m} \times 1.0 \text{ m}$  spacing (4500 vines per hectare). The vines were cane-pruned to a Guyot system with 12 nodes per cane and trained to a vertical shoot positioning (VPS) system using two pairs of fixed catch wires. During the growing season, vines were trimmed and defoliated only on the east-facing leaf canopy. Soil was managed by plowing and milling performed between March and June.

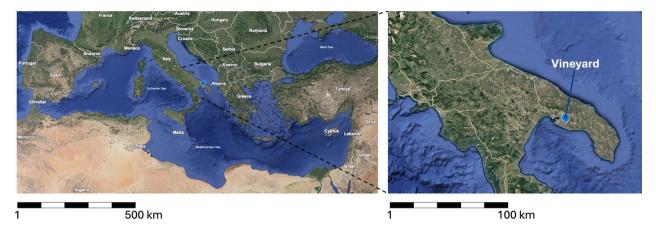


Figure 1. Satellite images of the Mediterranean basin (left) and the Apulia region (right) detailing the location of the vineyards under study (Google Earth).

The vines were grown without irrigation, and pesticide treatments applied according to integrate pest management procedures.

### Analysis of chemical parameters in grapes and wine

Chemical parameters were analyzed, both on grape at maturity and on wine throughout fermentation, using a WineScan Basic spectrometer (FOSS, Hillerød, Denmark). More specifically, three biological replicates of berries (about 500 g for each replicate) were randomly collected for each cultivar at grape maturity to determine total soluble solids concentration (g L<sup>-1</sup>) and pH. Must and wine analyses were implemented with additional parameters such as wine color intensity (dimensionless).

#### Vinification

A total of nine microvinifications, namely, three biological replicates for each cultivar (Merlot N., Merlot Khorus, and Merlot Kanthus) were performed using the same vinification protocol. In detail, 25 kg of grapes from each variety were harvested manually on the same day (August 22<sup>nd</sup>) and transported nearby to the "Cantine Due Palme" winery. Grapes were destemmed and crushed by hand, transferred to 5 L plastic fermentation containers, and 3.6 mL of liquid SO<sub>2</sub> (calculated based on the weight of destemmed grapes) was added to each container. Each fermentation container was filled to about 70% of its volume (3.5 kg of grape must) and inoculated with Viniferm 522 yeast (Agrovin, Ciudad Real, Spain). Musts were fermented into a refrigerated cell at 18-22°C for 8 days on the skins and punched once a day. During alcoholic fermentation, Nutriferm Vit (Enartis, Trecate, Italy), a fermentation activator of organic origin, was added to the musts twice 2 days apart. After alcoholic fermentation, the wines were pressed and 0.3 ml/L of liquid  $\mathrm{SO}_2$  (Potassium bisulfite 28%, Solfo K L, Vason, San Pietro in Cariano, Italy) was added. Subsequently, wines were cold decanted for 24 hours at 4°C. At the end of the decanting process, 2 bottles of 0.75 L were filled and 0.27 ml/L of liquid  $\mathrm{SO}_2$  was added to the wines. Each bottle was corked and stored in a cool and dry place.

### Sensory analysis

Two different sensory analyses were performed. All participants were of legal drinking age and were informed that the tasting involved alcoholic beverages. They were also informed that the study was an academic research project, that all data were going to be de-identified and only reported in the aggregate, and that the information collected would be used for research purposes only. All participants agreed to an informed consent statement to participate to the study. A first affective test was submitted to 61 judges of different gender (Figure S1), age (Figure S2), and frequency of wine consumption (Figure S3). The samples were presented to the panelists under ambient temperature and light, in a coded and randomized order. Each of them was asked to describe the tasted wine by a scale of preference (extremely liked, very liked, moderately liked, little liked, neither liked nor disliked, little disliked, moderately disliked, very disliked, extremely disliked). A second quantitative descriptive test was submitted to 10 expert judges including trained sommeliers and academic staff specialized in Viticulture and Enology from the University of Salento. The descriptive test was conducted using an unstructured graphical scale considering the following attributes: aroma (fruity, spicy, fresh, floral and dry vegetable, intensity and aromatic quality), taste (body of the wine, sweetness, acidity, bitterness, and astringency), and color intensity. The tasters rated the perception intensity in each experimental wine for each descriptor. A normalization based on the total variance of the perception of each taster for each parameter allowed to obtain the final ranking for each descriptor.

## Statistical analysis

Significant differences were assessed by ANOVA using the software SPSS v. 20 (IBM Corporation, Armonk, US). All ANOVAs were followed by Duncan's *post hoc* test at  $\rho \le 0.05$ .

### Results

#### Fruit composition

The comparison of grapes at maturity of resistant varieties Merlot Kanthus and Merlot Khorus as well as Merlot N. for the 2022 vintage revealed significant differences in their chemical composition (Figure 2). In particular, the concentration of sugar was significantly higher in Merlot Kanthus grapes (231  $\pm$  1.3 g  $L^{-1}$ ) than in the other cultivars. In contrast, Merlot Khorus grapes had a significantly lower sugar concentration than Merlot N. (180  $\pm$  2.7 g  $L^{-1}$  vs. 223  $\pm$  1.7 g  $L^{-1}$ ) (Figure 2A). Associated with a lower sugar concentration, Merlot Khorus also exhibited a significantly lower pH (3.43  $\pm$  0.02) than Merlot Kanthus and Merlot N. (3.69  $\pm$  0.03 vs. 3.67  $\pm$  0.03) (Figure 2B).

#### Fermentation kinetics and wine composition

Fermentation was conducted for 8 days. During fermentation, parameters such as total soluble solids and alcohol were determined to assess fermentation kinetics, which took place smoothly and without anomalies. Moreover, color intensity was evaluated to monitor the progress of maceration (Figure S4).

Consistent with the chemical characteristics of the grapes at maturity, the alcohol content was higher in Merlot Kanthus wines than in Merlot N., while Merlot Khorus displayed a lower alcohol content. Wine pH was higher in Merlot N. than both resistant varieties, while the lowest pH was detected in Merlot Khorus. The color intensity of the wines was significantly higher in wines made from Merlot Kanthus and Merlot Khorus compared to Merlot N. (Table 1).

#### Wine sensory evaluation

According to the affective test, representing the general liking of nonexpert panelists (Figure 3A), slight differences were observed. Specifically, Merlot Kanthus and Merlot Khorus wines were perceived as more pleasant than Merlot N., and Merlot Kanthus tended to be more generally preferred than Merlot Khorus.

The results of the quantitative descriptive test, conducted by a panel of expert tasters, showed statistically

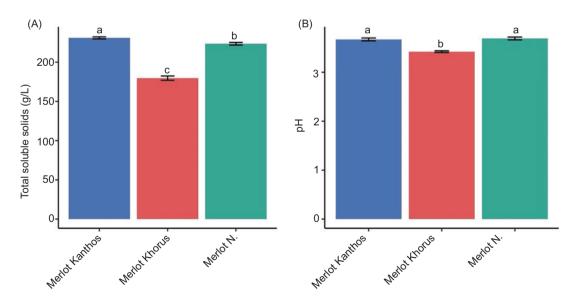


Figure 2. Chemical characteristics of grapes at maturity in terms of total soluble solids (A) and pH (B) among Merlot Kanthus, Merlot Khorus, and Merlot N.. Results are expressed as means  $\pm$  standard error. Duncan's *post hoc* test results among cultivars are indicated by the letter above the bars (p < 0,05).

significant differences for only a few of the parameters identified during the sensory analysis (Figure 3B). Fresh vegetable scents were perceived more strongly in the PIWI Merlot Kanthus than in Merlot Khorus, with Merlot N. displaying an intermediate perception for the same descriptor. No significant difference in acidic perception was found among Merlot Khorus and Merlot N. wines, while significantly higher levels were perceived in both when compared to Merlot Kanthus. Interestingly, color intensity was considered significantly more intense in disease-resistant grapevine wines than Merlot N.. No differences were found on bitterness, astringency, body, and sweetness. Although with no significant difference, Merlot Khorus wine exhibited a slightly more intense aroma profile with more pronounced fruity sensations.

Table 1. Values for technological and polyphenolic parameters of wines.

	Alcohol (%vol)	рН	Color intensity
	***	***	**
Merlot Kanthus Merlot Khorus Merlot N.	14.15 ± 0.05 <sup>a</sup> 10.88 ± 0.05 <sup>c</sup> 13.41 ± 0.05 <sup>b</sup>	3.62 ± 0.01 <sup>b</sup> 3.49 ± 0.01 <sup>c</sup> 3.68 ± 0.00 <sup>a</sup>	11.93 ± 1.10 <sup>a</sup> 14.51 ± 1.18 <sup>a</sup> 7.15 ± 0.05 <sup>b</sup>

Asterisks specify the level of significance evaluated with one way ANOVA with Duncan as *post hoc* test (\*\*\* = p < 0.001; \*\* = p < 0.05). Different letters indicate statistically different averages.

#### **Discussion**

The quality of disease-resistant grapevine variety wines has been the central topic of discussion since their development in the 19th century. With regard to the sensory analysis of these wines, Duley et al. (2023) recognized that disease-resistant grape cultivars are characterized by flavors and aromas that are somewhat different to that of classical V. vinifera cultivars. Other studies claim that wines made from wild Vitis species as well as aforementioned disease-resistant grapevine varieties often also have "foxy" and herbaceous characteristics. González-Centeno et al. (2019) analyzed the sensory potential of monovarietal red wines produced from resistant grape varieties, showing their promise to produce high-quality wines, as their phenolic and volatile composition appeared close to that of the traditionally cultivated monovarietal red wines. As fungus-resistant grape varieties carry non-V. vinifera genes, even at low amounts, they may be subjected to the perception that PIWI grapevines lead to low-quality wines (Fuller et al., 2014). However, many French-American disease-resistant grapevine varieties were thought to produce wine of "satisfactory commercial quality," winning medals in wine competitions (Paul, 1996).

In this study, wine produced in the hot and dry conditions of the southern Italian region of Apulia using traditionally cultivated Merlot N. was compared to that obtained from two disease-resistant grapevine varieties resulting from a cross involving the former as parent.

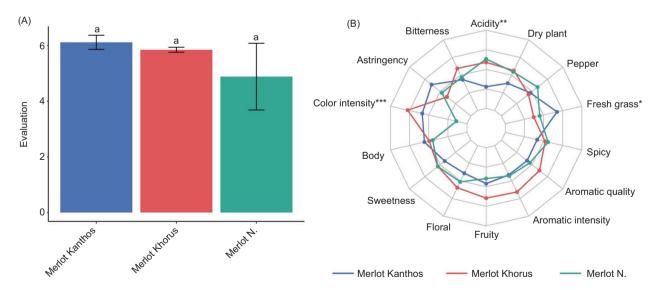


Figure 3. Evaluation of the general liking (affective test) of Merlot Kanthus, Merlot Khorus, and Merlot N. wines (A). Radar chart of sensory attributes of Merlot Kanthus, Merlot Khorus, and Merlot N. evaluated by the quantitative descriptive test (B). Results of the affective test are expressed as mean  $\pm$  standard error. Asterisks alongside the parameters specify the level of significance evaluated with one-way ANOVA with Duncan as *post hoc* test (\*\*\* = p < 0.001; \*\* = p < 0.05). Different letters indicate statistically different averages.

Firstly, the chemical analyses of the grapes showed a greater capacity for Merlot Kanthus to accumulate total soluble solids in the grapes than Merlot Khorus. Important differences emerged when comparing the pH of the clusters at maturity, where Merlot Khorus had a lower pH than Merlot Kanthus. Different results were shown by Bavaresco et al. (2023) where the two resistant Merlots showed comparable grape soluble solids. The contrasting results emerged in this work could be explained by the relevant role of genotype-environment interaction. As highlighted by Gonçalves et al. (2020) and Nikolić et al. (2017), the agronomic performance and sensory profiles of PIWIs are closely linked to the soil and climatic condition of the terroir in which they are grown. For this reason, assessing the genotype-environment interaction is crucial for accurately interpreting the differences found between studies conducted in different environments. Furthermore, climatic changes, such as the increase of average temperatures, lead to an anticipation of grapes ripening, which is often associated with a reduction in acidity and an increase in pH (Webb et al., 2011). On these premises, the use of Merlot Khorus, capable of maintaining a lower pH, could be an effective strategy to preserve the oenological balance of wines in Mediterranean terroirs.

In further support of the employment of such genotypes in warmer and dryer regions, wines from the resistant varieties Merlot Kanthus and Merlot Khorus presented a tendential higher general liking than Merlot N. in the affective test. This reinforces recent evidence suggesting that the quality of fungus-resistant grape wines can be equivalent to that of V. vinifera (Pedneault et al., 2012; Van Der Meer et al., 2010). In more detail, the quantitative descriptive test revealed a higher color intensity in wines produced from resistant varieties than in Merlot N.. Merlot Kanthus also tended to be more astringent, although no differences were noted about body and sweetness between the two resistant varieties and their V. vinifera counterpart. Generally, wines from resistant varieties are characterized by a higher anthocyanin level than red V. vinifera wines, while also carrying lower tannin levels (Casassa and Harbertson, 2014; Kilmister et al., 2014; Pedneault K. et al., 2014; Springer and Sacks 2014). This imbalance in polyphenol composition generally leads to color instability and lack of mouthfeel in the resulting wines (Manns et al., 2013; Springer and Sacks 2014). Although our study did not examine color instability, the tasting panel did not remark a difference in the smoothness of the compared wines. On the other hand, the aromatic quality and intensity of the Merlot Khorus wines tended to be superior, with a slightly higher perception of floral and fruity aromas compared to Merlot Kanthus and Merlot N.. Fruity notes have been previously described in many red resistant wines (Pedneault and Provost, 2016), but herbaceous notes have also been reported (Mansfield and Vickers, 2009; Rousseau et al., 2013).

Climate change is expected to have a significant negative impact on grapevine growth and yield (Daldoul et al., 2020). Moreover, because of the high pest sensitivity of this crop, 13% in mass of all synthetic pesticides used in Europe are applied in its cultivation (Muthmann and Nadin, 2007). Numerous environmental concerns are connected to pesticide use, like surface and groundwater pollution, contaminated runoffs from the fields, bee poisoning (Katherine and Roger, 2013), and/or emission of toxic active substances to the air compartment (Ducroz, 2006). Consumers do not only buy wine on the basis of various distinguishing features, such as country and region of origin, grape variety, price, and brand (Borrello et al., 2021; Mian et al., 2022), but are nowadays also more concerned with sustainability issues (Sellers-Rubio and Nicolau-Gonzalbez, 2016). In this scenario, breeding techniques aimed at the selection of resistant vines is a useful strategy for the control of major fungal diseases, improving the overall quality of the wine-growing sector and leading to a conversion of viticulture to more sustainable approaches. In Italy, grapevine resistant to fungal diseases is allowed in seven regions (Trentino-Alto Adige, Veneto, Friuli Venezia Giulia, Lombardy, Emilia-Romagna, Marche, and Abruzzo) (Sillani et al., 2022). This study presents, although based on data collected in a single vintage, an example of successful employment of resistant grapevine varieties in a hot and dry terroir such as that of Salento in the Apulia region of southern Italy. Future studies could focus on the quality potential of these resistant grapevine varieties in vintages with different seasonal conditions. Red varieties Merlot Kanthus and Merlot Khorus exhibited a great potential to produce high-quality wines in the Mediterranean area, introducing a concrete case in support of their application in terroirs with such characteristics. Ideally, this evidence could aid the evaluation procedures which lead to the authorization of such genotypes in areas in which their cultivation is currently not permitted, thereby contributing to advancing the sustainable transition of viticultural practices demanded by modern times.

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## **Competing Interests**

The authors had no relevant financial interests to disclose.

## **Author Contributions**

All authors contributed equally to this article.

### **Conflicts of Interest**

None.

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None.

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# **Supplementary**

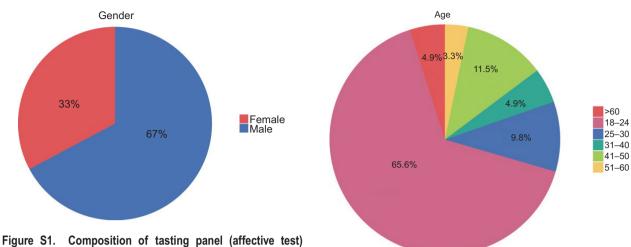


Figure S1. Composition of tasting panel (affective test according to gender.

Figure S2. Composition of tasting panel (affective test) according to age.

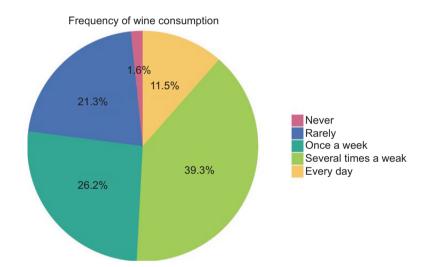


Figure S3. Composition of tasting panel (affective test) according to the frequency of consumption.

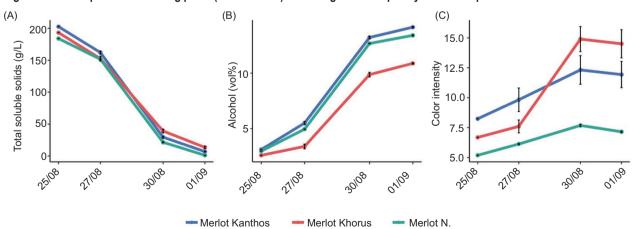


Figure S4. Trend of chemical parameters during fermentation and maceration monitoring. (A) total soluble solids; (B) alcohol; (C) color intensity. Results are expressed as means ± standard error.