Scientific Papers. Series B, Horticulture. Vol. LXVI, No. 1, 2022
Print ISSN 2285-5653, CD-ROM ISSN 2285-5661, Online ISSN 2286-1580, ISSN-L 2285-5653

# THE RESVERATROL CONTENT IN BLACK GRAPES SKINS AT DIFFERENT DEVELOPMENT STAGES

**Ramona CĂPRUCIU, Daniela Doloris CICHI, Liviu Cristian MĂRĂCINEANU, Dorin Constantin COSTEA**

University of Craiova, Department of Horticulture and Food Science, Faculty of Horticulture, 13 A.I. Cuza Street, Craiova, Romania

Corresponding author email: ramona_capruciu@yahoo.com

***Abstract***

*The resveratrol (3,5,4'-trihydroxystilbene) is a phytoalexin belonging to the class of polyphenolic compounds called stilbene, produced in response to stress factors by a wide variety of plants, including Vitis vinifera. This study presents the high performance liquid chromatography (HPLC) system detection of the forms of resveratrol (cis- and trans-) isomers in the skin of the Cabernet Sauvignon and Merlot black grapes from the Şimnicu de Sus wine grape-growing area during the 2019 and 2020 period. The resveratrol isomers were determined throughout the period of maturation of the studied varieties (15 July to 5 September) of the grape berries skin samples. The resveratrol content of grapes skin decreased constantly from the green phase until the full maturity for both varieties, reaching zero for cis-resveratrol (0.043mg/L for Cabernet Sauvignon variety in 2020) in the matured fruits.*

***Key words****: cis- and trans- resveratrol, black grapes, grape skin.*

## INTRODUCTION

The resveratrol was isolated from the grape berries, skin and seeds in 1976 (Wu et al., 2013). In 1992, Siemann and Creasy reported that the resveratrol could appear in grape products and especially in wine being considered responsible, at least partially for the effects of wine on coronary artery disease. Other studies confirmed this aspect as well (St Leger et al, 1979; Rimm et al., 1991; Hertog et al., 1995; Rimm et al., 1996; Arthur et al., 1997; Hao et al., 2004; Nakata et al., 2012; Timmers et al., 2012; Mazza et al., 2021). Some varieties showed a *trans*-resveratrol content in skins of two to three times higher than the average content in all varieties (Torchio et al., 2013; Nguyen et al., 2020). Besides, resveratrol play a role in the prevention of human pathological processes such as inflammation, atherosclerosis and carcinogenesis ( Stivala et al., 2001; Donna et al., 2009; López-Nicolás et al., 2009; Holthoff et al., 2010; Shingai et al., 2011; Stark et al., 2011; Mobasheri et al., 2012; Carter et al., 2014). The *trans*-resveratrol was considered to have antimutagenic and chemoprotective effects against cancer proliferation (Melzoch et al., 2001; Borra et al., 2005; Chandra et al., 2015; Munir et al., 2015; Jiang et al., 2017), antiseptic, antioxidant and anti-inflammatory characteristics (Park et al., 2000; Pinto et al., 2005; Szewczuk et al., 2005; Bist et al., 2010; Zhu et al., 2012; Goutzourelas et al., 2015; Benayahoum et al., 2015; Nunes et al., 2017). As a result of these studies, the resveratrol is considered one of the constituents of black grapes and red wine that can be used to make pharmaceutical preparations in order to improve human health (Delmas et al., 2005; Ahn et al., 2007).

The in-depth study of resveratrol isomer stability was conducted by (Jeandet et al., 1991; Căpruciu et al., 2007; Cayuela et al., 2009; Kutil et al. 2015; Novelle et al., 2015; Oliveira et al., 2017; etc). Another important aspect for the synthesis of resveratrol is the influence of the viticultural area on the total polyphenols dynamics.

The influence of the viticultural area has been studied by many authors who have established direct relationships and correlations between climatic conditions (temperature, precipitations), soil conditions and resveratrol. It has been established that the duration of sunshine, precipitation, slope exposure and winds are essential factors for the synthesis and dynamics of total polyphenols (Sautter et al., 2008; Rastija et al., 2009; Ubalde et al., 2010; Duque et al., 2011; Geana et al., 2015; Cichi et al., 2016; Leeuwen et al., 2016).

The quality-oriented practices in viticulture produce grapes with high levels of stilbene (Bavaresco, 2003; Yaman et al., 2016; Storchi et al., 2019; Rocchetti et al., 2021).

It has been found that grape vines synthesize the resveratrol as a response to stress factors: hitting impact, high level of UV radiation (Rodriguez et al., 2006; Choi, 2011), fungal infections (Hoos and Blaich, 1990, Dai et al., 1995; Schoonbeek et al., 2001; Pezet et al., 2004), the presence of heavy metals such as copper, etc.

The exposure to UV radiation produces irreversible isomerization of *trans*-resveratrol in *cis*- resveratrol which exhibits less stability than the isomer which is not exposed to light (Delmas et al., 2011; Wu et al., 2013).

When *Botritys cinerea* attacks, the resveratrol is produced in grape as a means of self-defense. Some studies have shown that a 160 mg/L trans-resveratrol concentration is sufficient to inhibit the growth of the *Botritys cinerea* mycelium (Adrian & Jandet, 2012).

The objectives of this study were to determine through high performance liquid chromatography (HPLC) system, the *cis* and *trans* resveratrol from the grapes of Cabernet Sauvignon and Merlot cultivars at different stages of their development, as well to do statistical analysis of the obtained results at full grapes maturation.

## MATERIALS AND METHODS

### *Sampling Sites*

The experiments were organized in parcels placed in identical orographic conditions (plateau conditions), on a slightly levigated reddish brown soil. Geographically, the area is located in South West of Romania, at 44°24'23 "N, 23°48'09" E and it is characterized by mild winters and long summers, with high sunshine duration and low rainfall (Table 1), climatic conditions required for the synthesis of anthocyanins and total polyphenols, including the resveratrol.

Table 1. Description of the sampling sites for blake grapes

| Sampling sites | Șimnicu de Sus |
|---|---|
| Altitude (m) | 175 |
| Average July temperature (°C) | +22°C … +23°C |
| Radiation | Hight |
| Precipitation (mm) | 565 |
| Soil | terraced forest brick soils or heavily eroded slopes with moderate slopes rich in carbonates |

The organization of the experiments was done by the randomized block method, with 2 variants, representing the studied varieties, respectively 15 vine per variety in 4 repetitions. Soil preparation and grapevine work was done uniformly for both varieties.

The varieties included in the study are grafted on Berlandieri x Riparia Kober 5BB rootstock, with 2 x 1.2 m planting distance, semi-high growth with multiple Guyot cutting system, with a crop load of 40 buds/vine.

### *Fruit Sampling*

The biological material used for analysis was the grape berries skins of `Cabernet Sauvignon` and `Merlot` cultivars, which are basic varieties in the vineyards of south-western Romania, used in the production of high quality red wines.

The samples were harvested at 15 days between the veraison phase and full maturity (15 July - 5 September), the analyzed varieties being in the same stage of development.

For each variety, 100 grape berries were harvested at the same specified date, in 3 repetitions and were analyzed in viticulture lab of the Faculty of Horticulture, University of Craiova (3 samples).

### *Reagents*

The pure resveratrol (*trans* - 3,5,4' - trihydroxystilbene) was purchased from Sigma Chemical Co. and the stock solution (1000 mg/L was prepared by dissolving 25.00 mg of commercial product, without previous purification in 25 mL of methanol. The acetonitrile, methanol and acetic acid were used for liquid chromatography (Merck). The used water was ultra-pure, Basic TWF.

***The used protocol to resveratrol determination by HPLC***

A. *Extraction-purification method based on solvent extraction*

The extract obtained from the skin of 100 grapes berries (representing a sample) was used in order to identify the cis and trans-resveratrol forms by HPLC Agilent 1000. The grape berries obtained from the three samples were manually separated, their skin was weighed and placed in 50 mL of 96% ethanol. After homogenization, the samples were kept in the dark for 48 hours. To obtain the extracts, the centrifugation method was used at 4000 rpm for 20 minutes followed by filtration through 0.45 µm filters. Centrifugation was performed with Centurion Scientific Centrifuge LTD K2 R Series. The samples obtained were kept in the dark until the extract was subject to chromatographic analysis.

B. *HPLC method for determination*

This study involves a simple, accurate, sensitive and reproducible high performance liquid chromatographic method for the determination of resveratrol (*cis*- and *trans*-3,5,4' -trihydroxystilbene) in the skin of Cabernet Sauvignon and Merlot grapes. The calibration curves of *trans*-resveratrol and *cis*-resveratrol were obtained by plotting the peak area of each standard against concentration in the range of 5-100 and 2-50 mg/L, respectively. The number of calibration points was eight for *trans*-resveratrol and five for *cis*-resveratrol. Each calibration point was the average of three independent measurements.

The chromatographic separation was performed on an Econosil C18 10 U column (250 mm x 4.6 mm i.d., 5 µm particle size) at room temperature (20°C) and with thermostatic control. The total gradient time was 35 minutes, with a flow rate of 1.0 mL/min, with an injection volume of 5 µL. The gradient elution system consists of water (A) and acetonitrile (B) ($H_2O/CH_3CN$ 2: 8 v/v). The column effluent was monitored by the UV detector.

The *cis*-resveratrol is detected at absorbance of 280 nm and the *trans*-resveratrol at 307 nm absorbance. The identification was based on retention time and spectrum data. In order to confirm the accuracy of the method, repetitive analyzes were performed, calculating the mean relative standard deviation (RSD) for three replicated determinations. The calculation of the limit of detection (LOD), signal-to-noise ratio (S/N) = 3 of the individual compounds was reached at their maximum absorption.

***Statistical Analysis***

In order to determine the content of *trans*- and *cis*-resveratrol in the grapes of the Cabernet Sauvignon and Merlot, the analysis of variance (ANOVA) was used. The differences between the means values of *trans*- and *cis*- resveratrol were tested by the Duncan test (using the SPSS 16 program). The results were expressed in mg/L, as mean values ± standard deviation (SD). Also, the coefficient of variation (CV %) was calculated.

## RESULTS AND DISCUSSIONS

The results of the study regarding the dynamics of *trans*- and *cis*- resveratrol in the grape berries skins are presented in Table 2 and Table 3.

Table 2. The variability of *trans*- resveretrol (mg/L) in grape berries skin analyzed in Șimnicu de Sus viticultural areas during the 2019-2020

| Calendar dates | Variety of grapes | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Cabernet Sauvignon | | | | | | Merlot | | | | | |
| | 2019 | | | 2020 | | | 2019 | | | 2020 | | |
| | Mean | S.D. | CV % | Mean | S.D. | CV% | Mean | S.D. | CV % | Mean | S.D. | CV % |
| 15 July | 9.152 | 0.76 | 8.3 | 8.463 | 0.27 | 3.2 | 11.083 | 0.17 | 1.5 | 10.443 | 0.37 | 3.5 |
| 29 July | 5.828 | 0.65 | 11.2 | 4.539 | 0.17 | 3.7 | 7.581 | 0.25 | 3.3 | 6.778 | 0.27 | 4.0 |
| 8 August | 3.316 | 0,40 | 12.1 | 2.822 | 0.19 | 6.7 | 4.363 | 0.26 | 6.0 | 3.169 | 0.21 | 6.6 |
| 22 August | 1.997 | 0.10 | 5.0 | 1.240 | 0.18 | 14.6 | 2.491 | 0.42 | 16.8 | 1.897 | 0.20 | 10.5 |
| 5 Sept. | 0.633 | 0.08 | 12.7 | 0.430 | 0.04 | 9.3 | 0.996 | 0.04 | 3.9 | 0.675 | 0.06 | 8.4 |

Table 3. The variability of *cis*- resveretrol (mg/L) in grape berries skin analyzed in Șimnicu de Sus viticultural areas during the 2019-2020

| Calendar dates | Variety of grapes | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Cabernet Sauvignon | | | | | | Merlot | | | | | |
| | 2019 | | | 2020 | | | 2019 | | | 2020 | | |
| | Mean | S.D. | CV % | Mean | S.D. | CV % | Mean | S.D. | CV % | Mean | S.D. | CV % |
| 15 July | 7.119 | 0.13 | 1.8 | 6.459 | 0.13 | 2.0 | 8.071 | 0.14 | 1.7 | 6.994 | 0.3 | 4.3 |
| 29 July | 4.910 | 0.14 | 2.8 | 3.531 | 0.11 | 3.1 | 5.573 | 0.09 | 1.7 | 5.178 | 0.02 | 0.4 |
| 8 August | 2.213 | 0.04 | 1.9 | 1.788 | 0.1 | 5.7 | 3.828 | 0.02 | 0.6 | 2.123 | 0.01 | 0.7 |
| 22 August | 1.083 | 0.06 | 4.6 | 1.045 | 0.03 | 2.6 | 1.095 | 0.03 | 2.6 | 1.105 | 0.03 | 2.6 |
| 5 Sept. | 0.065 | 0.01 | 19.6 | 0.043 | 0.01 | 23.3 | 0.086 | 0.01 | 15.4 | 0.058 | 0.01 | 16.6 |

The presented data show differences in the resveratrol accumulation between both isomers and varieties, which change from one determination to another. The results of the study show that a maximum of *cis*- and *trans*-resveratrol content was recorded at the beginning of the grape maturation period for both Cabernet Sauvignon and Merlot varieties in both years of study (2019 and 2020). There is a continuous decrease in the content of *trans*- and *cis*-resveratrol from the initial stage of grape berries ripening to full maturity for both studied varieties. Jeandet et al. (1991), find that there is a decrease in the capacity of grape fruits to synthesize the resveratrol during the fruit maturation, and this appears to be produced at the skin level. Thus, a maximum content of 9.152 mg/L for *trans*-resveratrol was recorded on 15 July 2019 and 8.463 mg/L in 2020 for Cabernet Sauvignon in Șimnicu de Sus, the values continuing to decrease continuously until the full maturity in both years of study. A higher amount of *trans*-resveratrol quantity was extracted from the Merlot variety skins in comparison with the Cabernet Sauvignon variety under the same orographic conditions and climate of the Șimnicu de Sus vineyard plantation, i.e. 11.083 mg/L in the 2019 crop, and 10.443 mg/L in 2020. A continuous decrease in the *trans*-resveratrol content was observed during maturation also by Giuffrè (2013) on the four varieties of red grapes grown in the south-west Calabria (South Italy). The coefficient of variation of *trans*-resveratrol in the Cabernet Sauvignon variety skins is low or medium in all the analyzed data, between 5% and 12.7% in 2019 and between 3.2% and 14.6% in 2020 (Table 2). The variation coefficient of *trans*-resveratrol in the Merlot variety is low for most determinations periods, ranging from 1.5% to 16.5% in 2019 and from 3.5% to 10.5% in 2020 (Table 2). Higher levels of *trans*-resveratrol content in the Merlot variety have been observed throughout the study compared to the Cabernet Sauvignon variety, with a higher dynamic in 2020. It can also be observed that at the end of the maturity process, the lowest value of *trans*-resveratrol is detected by the HPLC system at the Cabernet Sauvignon variety in 2020 with a value of 0.430 mg/L In another study, the highest concentration of *trans*-resveratrol detected in the Cabernet Sauvignon grapes at full maturity in the Tekirdağ region of Turkey was 0.443 mg/L (Yaman et al., 2016). The resveratrol concentration in skins ranged from 0.5 to 14.1 μg/g fresh skin, with an average of 4.12 μg/g for the 36 skin samples from grapes grown in Japan (Okuda & Yokotsuka, 1996). The content of *cis*-resveratrol in Șimnicu de Sus recorded a maximum value at the beginning of the maturation period on July 15 for both Cabernet Sauvignon and Merlot. The content of *cis*-resveratrol decreases continuously for both varieties both in 2019 and in 2020 until September 5 (last date of the analysis). Lower values were observed at the technological maturity compared with *trans*-resveratrol content (Table 3). The coefficient of variation of *cis*-resveratrol for the Cabernet Sauvignon variety is low in most analyzed data, ranging from 1.8% to 4.6% in 2019 and between 2% and 5.7% in 2020 and it has medium values (19.6 %) at the end of the maturation period, in 2019 and very high values (23.3%) in 2020 (Table 2). The Merlot variety is characterized in both years of study by a higher capacity of *cis*-resveratrol synthesis compared to the Cabernet Sauvignon variety. The coefficient of variation of *cis*-resveratrol for Merlot variety is also low in most of the analyzed data, ranging between 0.6% and 2.6% in 2019 and between 0.4% and 4.3% in 2020 and medium at the end of the maturation period, both in 2019 (15.4%) and 2020 (16.6%) (Table 3). Cui et al. (2015), after a study on grapes and wine from three major wine regions in China, show a resveratrol content in Merlot wines higher than that recorded in Cabernet Sauvignon wines. A comparative analysis of the results obtained in the two analyzed varieties in the two years of study (Table 4 and Table 5) was performed on September 5 (the last date of the analysis). Thus, on September 5 2019, the Cabernet Sauvignon variety recorded an average content of 0.633 mg/L of *trans*-resveratrol, with 36.4% lower than the Merlot variety, which had an average content of 0.996 mg/L the negative difference being statistically assured to a significant degree. Also, in 2020, the Cabernet Sauvignon variety recorded an average content of 0.43 mg/L in *trans*-resveretrol, lower with 36.3% than the Merlot variety, which had an average content of 0.675 mg/Lbut the negative difference was not statistically assured (Table 4). Similar data for the *trans*-resveratrol content of the Cabernet Sauvignon and Merlot varieties in the Șimnicu de Sus wine grape-growing region were obtained at the end of the maturation period also by Geana et al. (2015), for the same varieties grown in Murfatlar vineyards in Dobrogea, the viticultural area located in the south-east of Romania.

Also, in 2020, the Cabernet Sauvignon variety recorded an average content in *cis*-resveratrol of 0.043 mg/L, lower with 25.9% than the Merlot variety, which had an average content of 0.058 mg/L the difference was not statistically assured.

The two varieties studied in the two years recorded similar *cis*-resveratrol content on September 5, the differences were not assured in any of the experimentation year (Table 5).

Table 4. The synthesis of the results on the *trans*- resveretrol content for the in grape berries skin analyzed in the Șimnicu de Sus viticultural area on full maturity

| Variety | 2019 | | 2020 | |
|---|---|---|---|---|
| | Content of *trans*-resveratrol mg/L | Relative content % | Content of *trans*-resveratrol mg/L | Relative content % |
| Cabernet Sauvignon | 0.633[b] | 63.6 | 0.430[a] | 63.7 |
| Merlot | 0.996[a] | 100.0 | 0.675[a] | 100.0 |

Note: Means separation by Duncan test at p≤0.05. Means with the same letter are not statistically significant.

Table 5. The synthesis of the results on the *cis*- resveratrol content for the in grape berries skin analyzed in the Șimnicu de Sus viticultural area on full maturity

| Variety | 2019 | | 2020 | |
|---|---|---|---|---|
| | Content of *cis*-resveratrol mg/L | Relative content % | Content of *cis* -resveratrol mg/L | Relative content % |
| Cabernet Sauvignon | 0.065[a] | 75.6 | 0.043[a] | 74.1 |
| Merlot | 0.086[a] | 100.0 | 0.058[a] | 100.0 |

Note: Means separation by Duncan test at p≤0.05. Means with the same letter are not statistically significant.

In 2019, the Cabernet Sauvignon variety recorded on 5 September a *cis*-resveratrol average content of 0.065 mg/L lower with 24.4% than the Merlot variety with an average content of 0.086 mg/L the difference not being statistically assured.

## CONCLUSIONS

Studying the dynamics of resveratrol from the beginning of the fruit maturation period until the technological maturity, it was observed that the *trans*-resveratrol form is quantified more than the *cis*-resveratrol form for the studied varieties, with the highest values for the Merlot variety in the Șimnicu de Sus viticultural area in the 2019 crop.

The *cis* and *trans*-resveratrol forms of the black grape varieties in the Șimnicu de Sus wine grape-growing area were on a downward trend during the maturation process, the highest values being recorded in the first maturation period. The resveratrol dynamics analysis of the studied varieties highlights the high potential of synthesis for this substance at the skin level, requiring further studies of these aspects. Thus, the Șimnicu de Sus viticultural area becomes a point of reference within the wine-growing area of South-West Oltenia region, Romania, for obtaining quality red wines. The resveratrol content in the grape berries skin at the time of technological maturity being a proof in this respect.

## ACKNOWLEDGMENTS

The authors thank the grape producer from the Șimnicu de Sus wine area for their collaboration, support and availability of the grapes from which the laboratory tests were performed. This research activity was carried out with the support of colleagues from the viticulture department of the Faculty of Horticulture, Craiova.

## REFERENCES

Adrian, M., & Jeandet, P. (2012). Effects of resveratrol on the ultrastructure of *Botrytis cinerea* conidia and biological significance in plant/pathogen interactions. *Fitoterapia.* 83 (8), 1345-1350.

Ahn, J., Lee, H., Kim, S. & Ha, T. (2007). Resveratrol inhibits TNF-α-induced changes of adipokines in 3T3-L1 adipocytes. *Biochemical and Biophysical Research Communications*, 364(4), 972-977.

Arthur, L., Klatsky, M., D., Armstrong, M.A., Gary D. & Friedman M.D. (1997). Red Wine, White Wine, Liquor, Beer, and Risk for Coronary Artery Disease Hospitalization fn1, *The American Journal of Cardiology,* Volume 80, Issue (4), 416-420.

Bavaresco, L. (2003). Role of viticultural factors on stilbene concentrations of grapes and wine. *Drugs Exp. Clin. Res,* 29(5-6), 181-7.

Benayahoum, A., Guebailia, H. A. & Houache, O. (2015). On the role of ethylene bridge elongation in the antioxidant activity of polyhydroxylated stilbenes: A theoretical approach. *Comptes Rendus Chimie*, *18* (2), 149-159.

Bist, R. & Bhatt, D.K. (2010). Augmentation of cholinesterases and ATPase activities in the cerebellum and pons-medulla oblongata, by a combination of antioxidants (resveratrol, ascorbic acid, alpha-lipoic acid and vitamin E), in acutely lindane intoxicated mice. *Journal of the Neurological Sciences*, 296(1-2), 83-87.

Borra, M.T., Smith, B.C. & Denu, J.M. (2005). Mechanism of human SIRT1 activation by resveratrol. *Journal of Biological Chemistry*, 280(17), 17187-17195.

Căpruciu, R., Olteanu I., Cichi, D.D., Costea, C.D., Cichi, M., Mărăcineanu, L.C., Genoiu, E., Căpruciu, F.D. (2007). Evaluarea prin sistem HPLC a formelor izomere ale resveratrolului din unele soiuri de vita de vie din centrul viticol Banu Maracine. *Lucrari Ştiinţifice/Univ. Agrara de Stat din Moldova.* 15(2), 184-187.

Carter, L.G., D'Orazio, J.A. & Pearson, J. (2014). Resveratrol and cancer: focus on in vivo evidence, *Endocr. Relat. Cancer,* 21(3), R209-25.

Cayuela, A., Vázquez, A., Pérez, A.G. & García, J.M. (2009). Control of Table Grapes Postharvest Decay by Ozone Treatment and Resveratrol Induction. *Food Science and Technology International*, *15* (5), 495-502.

Chandra, K., Singh, X.L. & Nihal, A. (2015). Resveratrol, in its natural combination in whole grape, for health promotion and disease management. *Ann NY Acad Sci,* 1348 (1), 150–160.

Choi, S.J. (2011). The identification of stilbene compounds and the change of their contents in UV-irradiated grapevine leaves. Kor. *J. Hort. Sci. Technol,* 29, 374-381.

Cichi, D.D., Costea, D.C. & Gheorghiu, N. (2016). The cold hardiness of some varieties of grapevine cultivated in the viticultural area Plenita (Southwestern Romania). *Analele Universităţii din Craiova, seria Agricultură – Montanologie – Cadastru (Annals of the University of Craiova - Agriculture, Montanology, Cadastre Series),* XLVI (1), 62-67.

Dai, G.H., Andray, C., Mondolet-Cosson, L. & Boubals, D. (1995). Histochemical studies on the interaction between three species of grapevine, *Vitis vinifera*, *V. rupestris* and *V. rotundifolia* and downey mildew fungus, *Plasmopora viticola*. *Physiol. Mol. Plant Pathol*, 46, 177- 188.

Delmas, D., Jannin, B., & Latruffe, N. (2005). Resveratrol: Preventing properties against vascular alterations and ageing. *Molecular Nutrition & Food Research*, 49 (5), 377-395.

Delmas, D., Aires, V., Limagne, E., Dutartre, P., Mazué, F. & Ghiringhelli, F. (2011). Transport, stability, and biological activity of resveratrol. *Annals of the New York Academy of Science,* 1215, 48-59.

Duque, A.L., Pinto, M.D.C. & Macias, P. (2011). Lipoxygenase inhibition by red wine phenolics compounds. *Journal of Food Biochemistry*, 35 (2), 542-555.

Geana, El., Dinca, O., Ionete, R.E., Artem, V. & Niculescu, V.C. (2015). Monitoring *trans*-resveratrol in grape berry skins during ripening and in corresponding wines by HPLC. *Food Tecnol Biotechnol*, 53(1), 73-80.

Donna, L.D., Mazzotti F., Benabdelkamel, H., Gabriele, B., Plastina, P. & Sindona, G. (2009). Effect of H/D Isotopomerization in the Assay of Resveratrol by Tandem Mass Spectrometry and Isotope Dilution Method. *Analytical Chemistry,* 81 (20), 8603-8609.

Goutzourelas, N., Stagos, D., Spanidis, Y., Liosi, M., Apostolou, A., Priftis, A., Haroutounian, S., Spandidos, D.A., Tsatsakis, A.M. & Kouretas, D. (2015). Polyphenolic composition of grape stem extracts affects antioxidant activity in endothelial and muscle cells. *Mol Med Rep.Epub,* 12(4), 5846-56.

Hao, H.D., & He, L.R. (2004). Mechanisms of Cardiovascular Protection by Resveratrol. *Journal of Medicinal Food*, 7(3), 290-298.

Hertog, M.G.L., Kromhout D., Aravanis C., Blackburn H., Buzina R.& Fidanza F. (1995). Flavonoid intake and long-term risk of coronary heart disease and cancer in the seven countries study. *Arch Intern Med*, 155, 381− 6.

Hoos, G. & Blaich, R. (1990). Influence of resveratrol on germination of conidia and mycelial growth of *Botrytis cinerea* and *Phomopsis viticola. Journal of Phytophatology,* 129(2), 102-110.

Holthoff, J.H., Woodling, K.A., Doerge, D.R., Burns, S.T., Hinson J.A., & Mayeux P.R. (2010). Resveratrol, a dietary polyphenolic phytoalexin, is a functional scavenger of peroxynitrite. *Biochemical Pharmacology*, 80(8), 1260-1265.

Jeandet, P., Bessis, R. & Gautheron, B. (1991). The production of resveratrol (3,5,4'-trihydroxystibene) by grape berries in different development stages. *Am. J. Enol. Viticult*, 42, 41-46.

Jiang, Z., Chen, K., Cheng, L., Yan, B., Qian, W., Cao, J., Li, J., Wu, E., Ma, Q. & Yang, W. (2017). Resveratrol and cancer treatment: Updates. *Ann. N Y Acad. Sci*, 1403, 59–69.

Kutil, Z., Kvasnicova, M., Schuster, V., Marsik, D.P., Cusimamani, E.F., Lou, J.D., Vanek, T. & Landa, P. (2015). Effect of Dietary Stilbenes on 5-Lipoxygenase and Cyclooxygenases Activities In Vitro. *International Journal of Food Properties*, 18 (7), 1471-1477.

Leeuwen, C., & Darriet, P. (2016). The Impact Of Climate Change On Viticulture And Wine. *Journal Of Wine Economics,* 11(1), 150-167.

López-Nicolás, J.M., Pérez-Gilabert, M., & García-Carmona, F. (2009). Effect of Protonation and Aggregation State of (E)-Resveratrol on Its Hydroperoxidation by Lipoxygenase. *Journal of Agricultural and Food Chemistry,* 57 (11), 4630-4635.

Mazza, A., Nicoletti, M., Lenti, S., Torin, G., Rigatelli, G., Pellizzato, M. & Fratter, A. (2021). Effectiveness and Safety of Novel Nutraceutical Formulation Added to Ezetimibe in Statin-Intolerant Hypercholesterolemic Subjects with Moderate-to-High Cardiovascular Risk. *Journal of Medicinal Food*, 24(1), 59-66.

Melzoch, K., Hanzlikova, I., Filip, V., Buckiova, D. & Smidrkal, J. (2001). Resveratrol in parts of vine and wine originating from Bohemian and Moravian vineyard regions. *Agric. Conspec. Sci*., 66 (1), 53-57.

Mobasheri, A., Henrotin, Y., Biesalski, H.K. & Shakibaei, M. (2012). Scientific Evidence and Rationale for the Development of Curcumin and Resveratrol as Nutraceutricals for Joint Health. *International Journal of Molecular Sciences*, 13(4), 4202-4232.

Munir, N., Safdar, I. & Naz, S. (2015). Effect of induced mutation for varietal improvement in some local grapevine cultivars. *J. Anim. Plant Sci,* 25(1):234-242.

Nakata, R., Takahashi, S. & Inoue, H. (2012). Recent advances in the study on resveratrol. *Biol. Pharm. Bull*., 35, 273-279.

Nguyen, T.N., Dubreucq, E., Perrier, V., Tran, Q.H., Charpentier, C., Charnay, C., Terki, F., Jay-Allemand, C. & Bidel, L.P.R. (2020). Interactions between trans-resveratrol and CpLIP2 lipase/acyltransferase: Evidenced by fluorescence and in silico. *Food Chemistry*, 318, 126482.

Novelle, M.G., Wahl, D., Diéguez, C., Bernier, M. & De Cabo, R. (2015). Resveratrol supplementation: Where are we now and where should we go? *Ageing Res Rev,* 2015 May; Epub Jan 24, (21), 1-15.

Nunes, S., Danesi, F., Del Rio, D. & Silva, D. (2017). Resveratrol and inflammatory bowel disease: the evidence so far. *Nutr Res Rev,* Dec 1, 1-13.

Oliveira, A.L.B., Monteiro, V.V.S., Navegantes-Lima, K.C., Reis, J.F., Gomes, R.S., Rodrigues, D.V.S., Gaspar, S.L.F. & Monteiro, M.C. (2017). Resveratrol role in autoimmune disease-A mini-review. *Nutrients,* 2017; 9(12), 1306.

Okuda, T. & Yokotsuka, K. (1996). *Trans*-resveratrol concentrations in berry skins and wines from grapes grown in Japan. *Am. J. Enol. Viticult,* 47(1), 93-99.

Park, J.B. (2000). Inhibition of glucose and dehydroascorbic acid uptakes by resveratrol in human transformed myelocytic cells. *J. Nat. Prod*, Nr. 64, 381-384.

Pezet, R., Gindro, Viret, O. & Spring, J.L. (2004). Glycosylation and oxidative dimerization of resveratrol are respectively associated to sensitivity and resistance of grapevine cultivars to downy mildew. *Physiol. Mol. Plant Pathol*, 65, 297-303.

Pinto, M.C.& Macias, P. (2005). Oxidation of Dietary Polyphenolics by Hydroperoxidase Activity of Lipoxygenase. *Journal of Agricultural and Food Chemistry*, 53(23), 9225-9230.

Rastija, V. & Srečnik, G. (2009). Polyphenolic composition of Croatian wines with different geographical origins. *Food Chem*, 115, 54–60.

Rocchetti, G., Ferrari, F., Trevisan, M. & Bavaresco, L. (2021). Impact of Climatic Conditions on the Resveratrol Concentration in Blend of *Vitis vinifera* L. cvs. Barbera and Croatina Grape Wines. *Molecules*, 26*,* 401.

Rodriguez, M.R., Romero, P.R., Chacon, V.J.L., Martinez, G.J. & Garcia, R.E. (2006). Phenolic compounds in skin and seeds of ten grape *Vitis vinifera* varieties grown in a warm climate. *J. Food Compos*, Anal. 19, 687-693.

Rimm, E.B., Giovannucci, E.L., Willett, W.C., Colditz, G.A., Ascherio, A. & Rosner, B. (1991). Prospective study of alcohol consumption and risk of coronary disease in men. *Lancet*, 338, 464− 8.

Rimm, E.B., Katan, M.B., Ascherio, A., Stampfer, M.J., Willett, W.C. (1996). Relation between intake of flavonoids and risk for coronary heart disease in male health professionals. *Ann Intern Med*, 125, 384− 9.

Santos-Buelga, C., González-Manzano, S., González-Paramás ,A.M. (2022). White wine polyphenols and health, *White Wine Technology,* 10.1016/B978-0-12-823497-6.00017-X, 205-220.

Sautter, C.K., Storck, L., Rizzatti, M.R., Mallmann, C.A. & Brackmann, A. (2008). Síntese de trans-resveratrol e controle de podridão em maçãs com uso de elicitores em pós-colheita. *Pesquisa Agropecuária Brasileira*, 43(9), 1097-1103.

Schoonbeek, H., Del Sorbo, G.& De Waard, M.A. (2001). The ABC transporter BcatrB affects the sensitivity of *Botrytis cinerea* to the phytoalexin resveratrol and the fungicide fenpiclonil, *Mol. Plant Microbe Interact,* 14(4), 562-571.

Shingai, Y., Fujimoto, A., Nakamura, M. & Masuda, T. (2011). Structure and Function of the Oxidation Products of Polyphenols and Identification of Potent Lipoxygenase Inhibitors from Fe-Catalyzed Oxidation of Resveratrol. *Journal of Agricultural and Food Chemistry*, 59(15), 8180-8186.

Siemann, E. H. & Creasy, L. L. (1992). Concentration of the phytoalexin resveratrol in wine. *Am J Enol Vitic,* 43, 49-52.

St Leger, A.S., Cochrane, A.L. & Moore, F. (1979) Factors associated with cardiac mortality in developed countries with particular reference to the consumption of wine. *Lancet*, 1, 1017-20.

Stark, T., Wollmann, N., Lösch S., & Hofmann, T. (2011). Quantitation of Resveratrol in Red Wines by Means of Stable Isotope Dilution Analysis−Ultra-Performance Liquid Chromatography−Quan-Time-of-Flight Mass Spectrometry and Cross Validation. *Analytical Chemistry*, 83(9), 3398-3405.

Stivala, L.A., Savio, M., Carafoli, F., Perucca, P., Bianchi, L., Maga, G. & Vannini, V. (2001). Specific structural determinants are responsible for the antioxidant activity and the cell cycle effects of resveratrol. *J Biol Chem*. 276(25), 22586-22594.

Storchi, P., Puccioni, S. & Santini, C. (2019). 10 - The case of resveratrol-enriched wines: Is it possible to create a new niche or to revitalize mature products? Case Studies in the Wine Industry A volume in the consumer science and strategic marketing series Woodhead Publishing Series in Food Science, Technology and Nutrition, 129-138.

Szewczuk, L.M., Lee, S.H., Blair, I.A. & Penning, T.M. (2005). Viniferin Formation by COX-1: Evidence for Radical Intermediates during Co-oxidation of Resveratrol. *Journal of Natural Products*, 68(1), 36-42.

Timmers, S., Auwerx, J. & Schrauwen, P. (2012). The journey of resveratrol from yeast to human. *Aging,* 4(3), 146-153.

Torchio, F., Vincenzi, S., Tomasi, D., Gaiotti, F., Lovat, L., Giacosa, S., Torchio, F., Río Segade, S. & Rolle L. (2013). Comparative study of the resveratrol content of twenty-one italian red grape varieties. *S. Afr. J. Enol. Vitic,* 34(1), 30-35.

Ubalde, J. M., Sort, X., Zayas, A. & Poch, R.M. (2010). Effects of soil and climatic conditions on grape ripening and wine quality of Cabernet Sauvignon. *J Wine Res,* 21(1), 1-17.

Wu, C.F., Yang, J.Y., Wang, F. & Wang, X.X. (2013). Resveratrol: botanical origin, pharmacological activity and applications. *Chinese Journal of Natural Medicines,* 11, 1-15.

Yaman, Ü.R., Yaman, B.Ç., Adıgüzel, U., Yücel, N. & Çetinkaya (2016). Effect of vegetation time and climatic conditions on *trans*-resveratrol concentrations in Cabernet Sauvignon and Merlot wines from different regions in Turkey. *S. Afr. J. Enol. Vitic*, 37(1), 85-92.

Zhu, L., Zhang, Y., Deng, J., Li , H. & Lu, J. (2012). Phenolic concentrations and antioxidant properties of wines made from North American grapes grown in China*,* Mar 14; 17(3), 3304-23.