

Vujovic, D.; Pejin, B.; Popovic Djordjevic, J.; Velickovic, M.; Tesevic, V. Phenolic Natural Products of the Wines Obtained from Three New Merlot Clone Candidates. *Natural Product Research* **2016**, *30* (8), 987–990. <a href="https://doi.org/10.1080/14786419.2015.1079191">https://doi.org/10.1080/14786419.2015.1079191</a>

## SUPPLEMENTARY MATERIAL

# Phenolic natural products of the wines obtained from three new Merlot clone candidates

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#### **Abstract**

This work aimed to evaluate the total contents of polyphenolics (the Riberau-Gayon-Maurié procedure), anthocyanins (using pH differential method) and tanins (the Nègre procedure) as well as the content of phenolic acids (using UPLC/MS chromatography) respectively of the wines obtained from three new Merlot clone candidates in the perennial clonal selection. The aforementioned chemical parameters were determinated in the samples covering the period 2009-2012. In comparison both with the standard Merlot wine (mother vine) and the wines obtained from other two clone candidates, the Merlot wine of the clone candidate No. 022 was found to have the highest total content of all three examined components /1.89±0.05 g/L (polyphenolics), 185.59±5.00 mg/L (anthocyanins) and 1.11±0.03 g/L (tanins)/ as well as six phenolic acids including gallic acid (25.49±0.27 mg/L). These findings are in good agreement with the observed trend for the viticultural parameters indicating the clone candidate No. 022 as more promising than mother.

**Keywords:** clonal selection, Merlot wines, total polyphenolics, total anthocyanins, total tanins, phenolic acids

## **Experimental**

## Chemical analyses

The total contents of polyphenolics (the Riberau-Gayon-Maurié procedure), anthocyanins (by spectrophotometric procedure, using pH differential method) and tanins (the Nègre procedure) as well as the content of phenolic acids (using UPLC/MS chromatography with TQ analyser) respectively of the wines obtained from three new Merlot clone candidates (Danicic 1988; Lee et al. 2005).

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## HPLC/MS/MS analysis

The analysis of phenolic acids in selected wine samples was carried out using a Waters Acquity UPLC H-Class (WAT-176015007) equipped with quaternary pump (Waters Quaternary Solvent Menager), injector (Waters Sample Menager-FTN /Flow Through Needle/), column compartment with ZORBAX Eclipse XDB C18 column (150×4.6 mm; 5µm), Waters 2998 PDA (Photodiode Array) detector and mass detector (Waters TQ /Tandem Quadrupole, WAT-176001263/). For acquisition and processing data, MassLynx V4.1 software was used. Quantitative determinations were performed using the external standard method with commercial standards. The calibration curves were obtained by injection of standard solutions, under the same conditions as for the analysed samples, over the range of concentrations observed. Based in the area under peaks of each extracted transition of ions from certain compounds of standard solutions in ESI-mode, standard curves for each standard solution compounds were plotted. Identification of individual compounds relied on the retention times of original standards and spectral data.

## References

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- Lee J, Durst WR, Wrolstad RE. 2005. Determination of total monomeric anthocyanin pigment content of fruit juices, beverages, natural colorants, and wines by the pH differential method: collaborative study. J AOAC Int. 88:1269–1278.