

# CHARACTERIZATION OF MERLOT WINES BASED ON PHENOLIC COMPOSITION DETERMINED BY HPLC-DAD-ESI-MS AND MS/MS TECHNIQUE

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## INTRODUCTION

The concentration of phenolic compounds in wine depends on many factors, such as the variety, climate, soil, oenological practices applied for winemaking, aging and storage conditions. The purpose of this investigation was to determine the detailed phenolic profile of Merlot wines (vintage 2008), produced in Tikveš wine region in Republic of N. Macedonia. High-performance liquid chromatography with diode array detection coupled to mass spectrometry (HPLC-DAD-ESI-MS/MS) was used to identify and quantify the phenolic compounds in wines. Identification of the individual phenolic compounds was carried out by comparison of their UV/Vis spectra and retention times with those of the available standards, as well as by comparing the ESI-MS and MS/MS data with the standards analyzed under the same experimental conditions and those found in the literature.

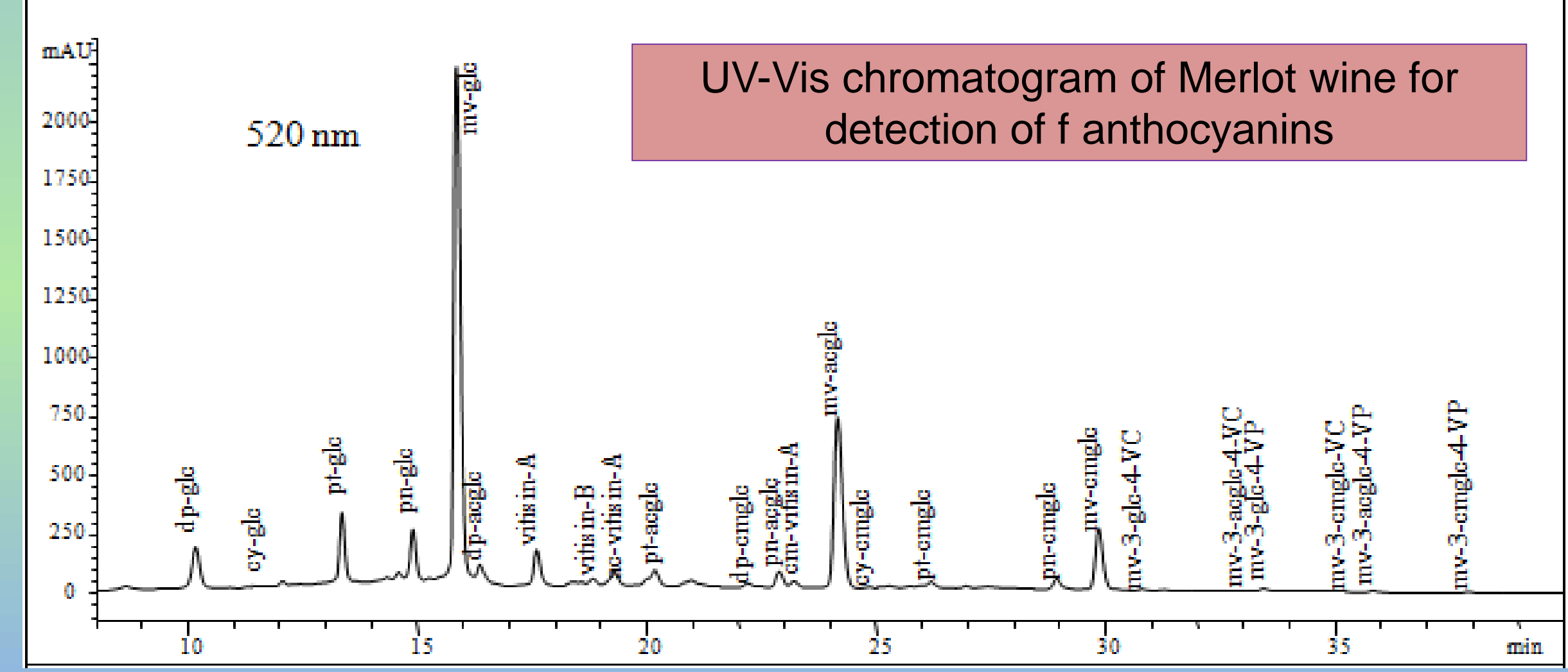
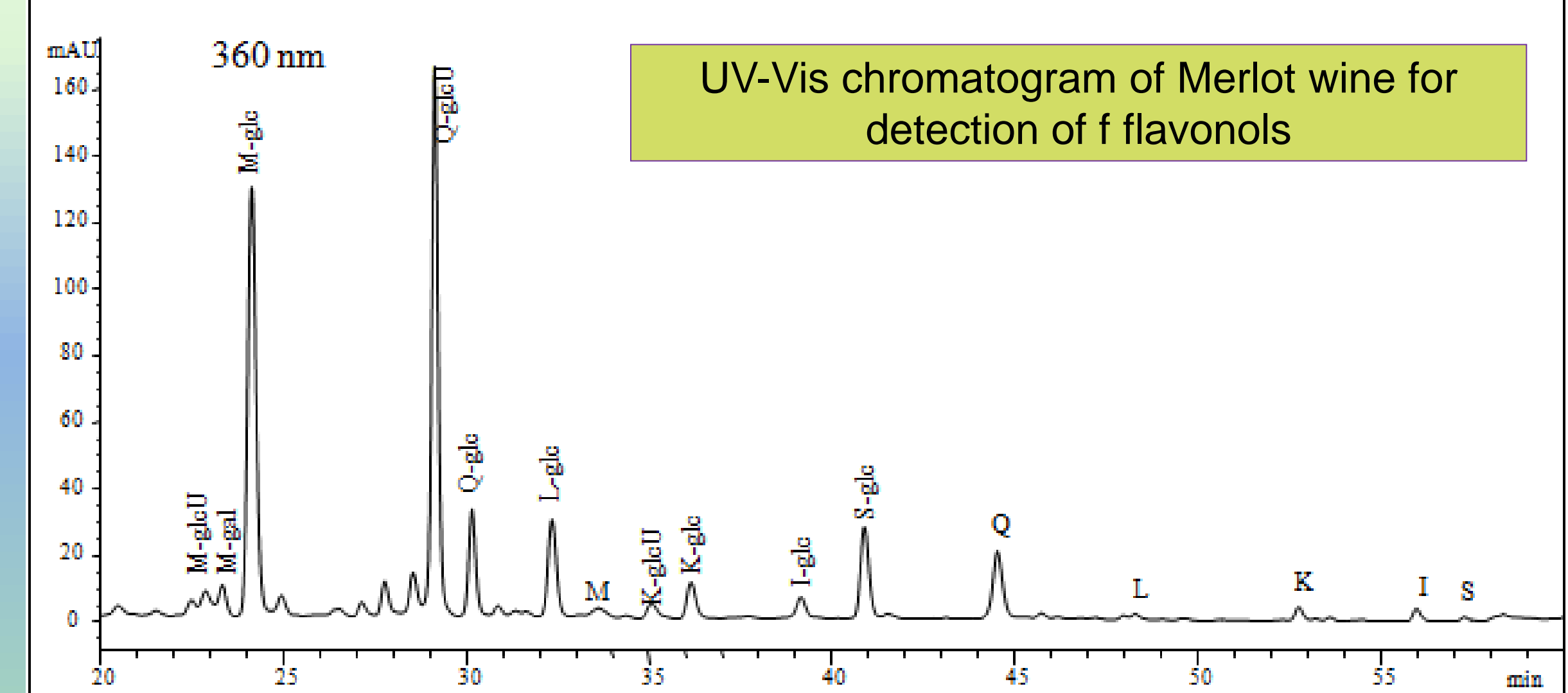
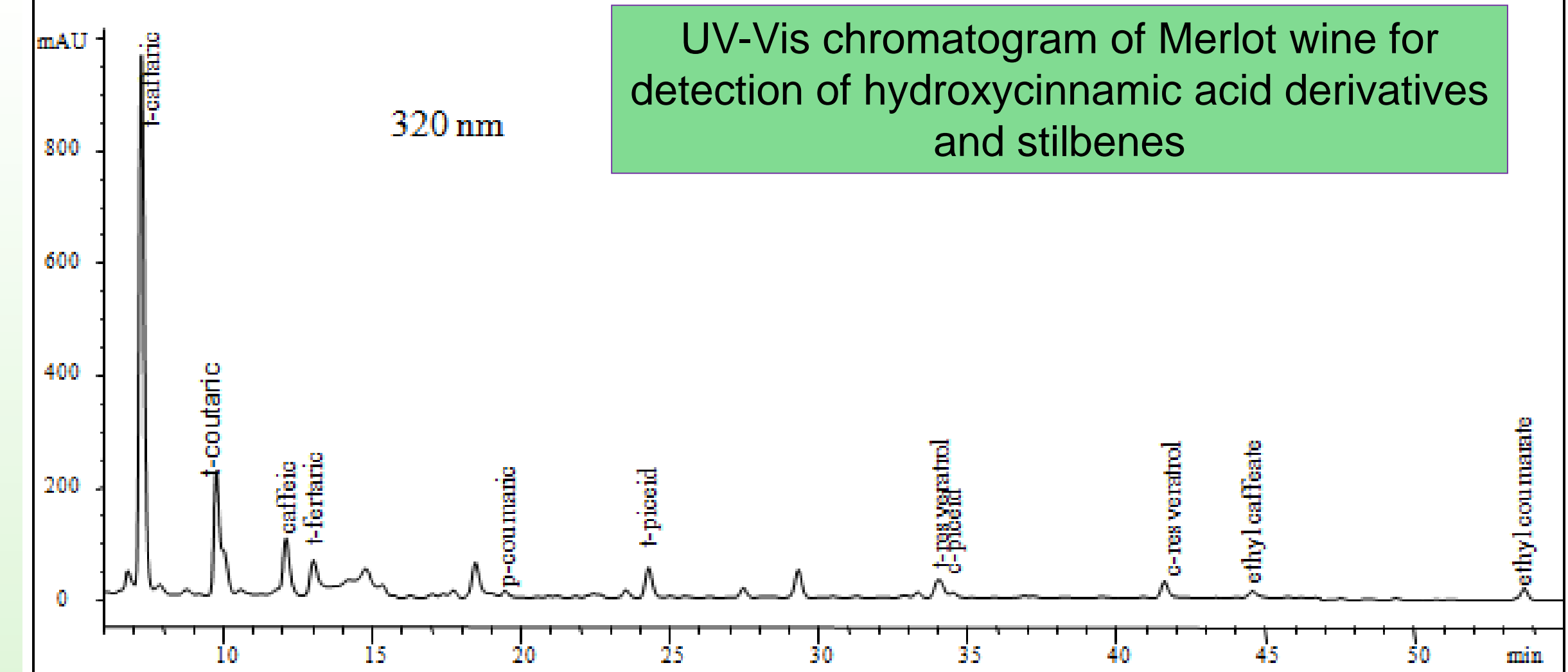
## MATERIALS AND METHODS

**Wine samples:** Merlot wines (vintage 2008)

### HPLC-DAD-ESI-MS/MS

✓ Solid-phase extraction of **non-anthocyanin phenolics** in wine. Mobile phase: solvent A (acetonitrile/water/formic acid, 3:88.5:8.5, V/V/V), solvent B (acetonitrile/water/formic acid, 50:41.5:8.5, V/V/V), and solvent C (methanol/water/formic acid, 90:1.5:8.5, V/V/V).

✓ For analysis of **anthocyanins and related pigments**, samples were only diluted with 0.1 M HCl solution (1:4, V/V). Mobile phase: solvent A (water/acetonitrile/formic acid, 87:3:10, V/V/V) and solvent B (water/acetonitrile/formic acid, 40:50:10, V/V/V).



## RESULTS AND DISCUSSION

### HPLC results for pigments in Merlot wines (vintage 2008)

| Anthocyanins (molar %)    |              | Vitisins and hydroxyphenyl-pyranoanthocyanins (molar %) |              | Nonflavonoids (molar %)          |              |
|---------------------------|--------------|---|--------------|----------------------------------|--------------|
| Compounds/Wines           | Merlot, 2008 | Compounds/Wines   | Merlot, 2008 | Hydroxycinnamic acid derivatives | Merlot, 2008 |
| Dp-3-glc                  | 4.83 ± 0.03  | Vitisin-A   | 46.5 ± 0.43  | <i>trans</i> -Cafutaric acid     | 59.3 ± 0.5   |
| Cv-3-glc                  | 0.52 ± 0.01  | Ac-vitisin-A  | 24.0 ± 0.21  | <i>trans</i> -Coutaric acid      | 14.9 ± 0.12  |
| Pt-3-glc                  | 5.74 ± 0.03  | p-Cm-vitisin-A  | 11.0 ± 0.10  | <i>cis</i> -Coutaric acid        | 5.3 ± 0.05   |
| Pn-3-glc                  | 4.51 ± 0.02  | Vitisin-B   | 10.4 ± 0.09  | Caffeic acid                     | 9.7 ± 0.09   |
| Mv-3-glc                  | 48.9 ± 0.55  | Ac-vitisin-B  | 8.17 ± 0.07  | <i>trans</i> -Fertaric acid      | 6.7 ± 0.06   |
| Dp-3-acglc                | 1.83 ± 0.01  | Total vitisins*   | 41.6 ± 0.35  | <i>p</i> -Coumaric acid          | 1.0 ± 0.01   |
| Pt-3-acglc                | 1.63 ± 0.01  | 10-DHP-pymv-3-glc (pinotin A)                           | 29.3 ± 0.30  | Ethyl caffeate                   | 1.5 ± 0.01   |
| Pn-3-acglc                | 1.98 ± 0.02  | 10-DHP-pymv-3-acglc                                     | nd           | Ethyl coumarate                  | 1.5 ± 0.01   |
| Mv-3-acglc                | 20.7 ± 0.38  | 10-DHP-pymv-3-cmglc                                     | nd           | Total HCAD (mmol/L)              | 248 ± 3.29   |
| Dp-3-cmglc                | 0.56 ± 0.01  | 10-MHP-pymv-3-glc                                       | 40.1 ± 0.37  | Stilbenes                        |              |
| Cv-3-cmglc                | nd           | 10-MHP-pymv-3-acglc                                     | 19.1 ± 0.15  | <i>trans</i> -piceid             | 40.2 ± 0.41  |
| Pt-3-cmglc                | 0.68 ± 0.01  | 10-MHP-pymv-3-cmglc                                     | 11.5 ± 0.09  | <i>trans</i> -resveratrol        | 14.6 ± 0.15  |
| Pn-3-cmglc                | 1.30 ± 0.01  | Total HP-pyranoanthocyanins (mg/L)                      | 6.28 ± 0.06  | <i>cis</i> -piceid               | 38.9 ± 0.39  |
| Mv-3-cmglc                | 6.81 ± 0.04  |   |              | <i>cis</i> -resveratrol          | 6.2 ± 0.06   |
| Total anthocyanins (mg/L) | 194 ± 2.3    |   |              | Total stilbenes (mmol/L)         | 19.2 ± 0.14  |

## CONCLUSION

- ✓ A total of 52 phenolic compounds have been identified and quantified in the wine samples applying HPLC-DAD-MS and MS/MS technique which included 14 anthocyanins, 16 pyranoanthocyanins, 16 flavonols, 8 hydroxycinnamic acids and their derivatives and 4 stilbenes
- ✓ Malvidin-3-glucoside and its derivatives were the major compounds.
- ✓ 10-carboxy-pyranomalvidin-3-glucoside (vitisin A) and 10-*p*-hydroxyphenyl-pyranomalvidin-3-glucoside were the main compounds from the family of vitisin-like and hydroxyphenyl-like pyranoanthocyanins, respectively.

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