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Analysis of the polyphenolic composition of red wines with particular respect to the resveratrol concentration

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Summary: In our work we have studied red wines of some vine-districts. In the centre of interest was resveratrol. We declared, there was not significant difference between varietys in polyphenol comparison. There was significant variety in anthocyanin and colour-intensity.

Key words: resveratrol, polyphenol, red wines

Introduction

In wine-drinking countries, there is an almost constant debate about the characteristics, effects and role of wine played in our lives. Scientific researches are focusing more and more on the examination of compounds found in foodstuff, exerting physiological effects – including those in wine.

The aim of the present work is to analyse resveratrol, one of the several compounds found in wines, exerting physiological effects, in Hungarian wines.

Resveratrol

Polyphenolics and the colorants belonging to them form part of one of the most important compound groups. Based on Peri and Pompei's classification (1971), there are non-flavonoid phenolics, flavonoid-phenolics and tannins. These compounds get into the wine from the grape. Apart from the technology, their concentration is largely influenced – inter alia – by the way of cultivation, their variety, ripeness and vintage.

For a few years, research directions have been focusing mostly on *resveratrol* and its derivatives and *antocyanins* providing colorants among the above-mentioned polyphenolics.

Resveratrol, which has physiological effects, is a significant component among non-flavonoid phenolics. It belongs to the stilbene family and the chemical name of the basic structure is α,β-diphenylethylene. It contains a phenolic hydroxyl group typical of polyphenolic compounds. It has two geometric isomers, between which trans-stilbene can be found in nature. The cis-form is unstable due to the sterical hindrance of phenolic groups. It often exists in a glucoside form, where resveratrol is linked to a sugar molecule by β-glycoside linkage. It is the so-called piceide or polydatin that upon the decomposition of the glycoside linkage changes into resveratrol.

Within the grape berry, they are present particularly in the grape skin and in an insignificant quantity in the seed (*Creasy, Coffee,* 1988), thus the resveratrol-content of the wine depends almost entirely on the processing technology!

Research findings show that resveratrol has a dual physiological role: On the one hand, it plays an important role in the natural protective mechanism of grapevines against pathogen, fungal infections. On the other hand, it provides protection against the development of cardiovascular diseases (*Seigneur* et al., 1990). The natural immune effect of stilbenes was emphasized by Langcake and *Pryce* (1976, 1977), *Drecks & Creasy* (1989), Stein and Hoss (1984) and Stein and Blaich (1985) alike.

According to *Mattivi & Nicolini's* (1993) findings, red wines contain an average of 2.24 g/dm^3 t-resveratrol. They analysed the effect wine-making technology exerts on the resveratrol content. The analyses carried out in Hungary reveal similar findings. (*Kállay* et. al., 1998)

The purpose of our present work is to map if there is a difference among the various Hungarian "wine regions" with regard to the polyphenolic composition of wines as well as if there is a significant difference in the polyphenolic composition of the different grape varieties. Among polyphenolics, resveratrol was in the focal point of our analyses.

Materials and method

We collected samples for our analyses from the characteristic „regions". They included some of the northern, central and southern parts of the country: Eger, Sopron, Balatonboglár, the Great Plain and Villány.

We analysed the wine varieties Kékfrankos, Pinot Noir, Cabernet Sauvignon, Merlot, Zweigelt, Cabernet Franc and Portigieser. We tried to select them taking into account them to be available from all the previously mentioned territories if it's possible. The pure-bred wine samples collected from each region were taken from grapes of more or less the same ripeness level. We did not improve musts, the samples analysed had been drawn off once.

Our samples are originated from the vintage of 2005 and 2006; and the measurement results represent an average value.

We evaluated the differences among the samples by a statistic method, by variance analysis.

The basic analysis:

- measuring titratable acid content by acid-base titration,
- pH measurement using a combined glass electrode,
- measuring alcohol content by distillation,
- determination of the total polyphenolic content applying the Folin–Ciocalteau reagent, calibrated to gallus acid,
- analysis of colour intensity and hue; we performed it applying the absorbencies measured at 420 and 520 nm, spectrophotometrially, with a layer thickness of 1 cm, according to the procedure described in the chapter "Analysis of Wines" of the Hungarian Wine Book".

By a well-known and widespread method (or its modified version), we determined:

- the quantity of leucoantocyanins, after heating a 40:60 compound of hydrochloric acid-butanol containing iron (II) sulphate, spectrophotometrically, (*Aubert,* 1970, modified)
- the antocyanin content measured at 550 nm by a spectrophotometer, after dilution carried out with 96% ethanol containing 2 ^{v}/v% concentration of HCl. (*Aubert,* 1970, modified)
- the catechin content in wine diluted with alcohol, made it react with vanillin-sulphuric acid reagent, at 500 nm, spectrophotometrically, (*Tanner, Brunner,* 1979, modified)
- the polymer%, after the decolourization of monomer antocyanins, due to the effect of SO_2, by measuring the two absorbencies usually applied at colour intensity (*Somers,* 1971)

The resveratrol concentration was determined by the HPLC technique, a method developed at our Department. (*Kállay, Török,* 1997)

Results

We summarized the results in a table. *(Tables 1, 2).* In order to give a comprehensive view, we illustrated the most characteristic parameters in a chart, too.

Although we analysed wines from grapes of the same technological ripeness, differences between alcohol strengths are natural, due to sugar grades. *(Figure 1.)*

According to the parameters revealed on the basis of *(Figure 2.)* – total polyphenolics, leucoantocyanins and catechins –, there seems to be a significant difference among the wines of grape varieties, ranked in ascending order:

Table 1.: Analysis and phenolic compounds of redwine

Kékfrankos	**alcohol (v/v%)**	**alcohol (g/l)**	**t. acidity (g/l)**	**pH**	**polimer%**	**colour hue**	**cis-piceid (mg/l)**	**trans-piceid (mg/l)**	**cis-resveratrol (mg/l)**
Sopron	12.34	28.1	5.98	3.23	52.99	0.520	0.4	0.5	0.9
Balatonboglár	13.83	25.8	5.56	3.26	32.94	0.540	0.7	1.6	0.5
Villány	14.74	29.7	4.99	3.35	59.11	0.590	0.6	1.6	1.1
Eger	16.58	37.8	7.35	3.41	58.06	1.096	0.5	0.5	0.5
Alföld Kiskőrös	9.86	29.4	7.35	3.15	36.47	0.540	0.6	0.8	0.7
average	13.47	30.16	6.25	3.28	47.91	0.66	0.56	1.00	0.74
Standard deviation	2.535922	4.539053	1.0673	0.10198	12.34143	0.246656	0.114018	0.561248608	0.260768096
Pinot Noir	**alcohol (v/v%)**	**alcohol (g/l)**	**t. acidity (g/l)**	**pH**	**polimer%**	**colour hue**	**cis-piceid (mg/l)**	**trans-piceid (mg/l)**	**cis-resveratrol (mg/l)**
Sopron	13.48	25.3	6.36	3.25	50.60	0.720	0.5	0.6	0.8
Balatonboglár	13.30	32.0	6.22	3.45	70.58	0.800	0.5	0.7	0.6
Villány	14.29	28.1	5.18	3.74	77.19	0.950	0.5	1.6	1.3
Eger	14.02	23.7	5.09	3.54	68.42	0.920	0.5	0.6	0.5
Alföld Kiskőrös	11.21	24.5	7.26	3.41	49.25	0.850	0.6	1.0	0.7
average	13.26	26.72	6.02	3.48	63.21	0.85	0.52	0.90	0.78
Standard deviation	1.213569	3.385558	0.903283	0.180194	12.55782	0.092574	0.044721	0.424264069	0.31144823
Cabernet Sauvignon	**alcohol (v/v%)**	**alcohol (g/l)**	**t. acidity (g/l)**	**pH**	**polimer%**	**colour hue**	**cis-piceid (mg/l)**	**trans-piceid (mg/l)**	**cis-resveratrol (mg/l)**
Sopron	13.83	31.3	5.84	3.35	51.52	0.610	0.6	1.1	0.7
Balatonboglári	13.30	28.1	5.56	3.40	49.67	0.610	0.6	0.7	0.6
Villány	14.56	29.2	4.81	3.70	47.45	0.780	0.0	1.0	0.8
Eger	14.56	26.6	6.79	3.37	52.30	0.630	0.6	0.6	0.5
Alföld Kiskőrös	13.39	32.6	6.32	3.40	54.10	0.690	0.5	0.6	0.7
average	13.93	29.56	5.86	3.44	51.01	0.66	0.46	0.80	0.66
Standard deviation	0.610795	2.413089	0.753213	0.144672	2.546639	0.072664	0.260768	0.234520788	0.114017543

Table 2.: Analysis and phenolic compounds of redwine

Merlot	alcohol (v/v%)	alcohol (g/l)	t. acidity (g/l)	pH	polimer%	colour hue	cis-piceid (mg/l)	trans-piceid (mg/l)	cis-resveratrol (mg/l)
Sopron	13.74	30.0	5.75	3.30	34.95	0.610	0.5	0.5	0.6
Balatonboglár	12.96	28.4	5.37	3.36	44.73	0.660	0.8	1.1	0.5
Villány	15.57	28.4	4.62	3.59	47.05	0.710	0.9	1.7	1.3
Eger	15.01	26.8	6.79	3.40	51.19	0.850	0.5	0.5	0.5
average	14.32	28.40	5.63	3.41	44.48	0.71	0.68	0.95	0.73
Standard deviation	1.186676	1.306395	0.903267	0.125266	6.892324	0.103401	0.206155	0.574456265	0.386221008
Zweigelt	**alcohol (v/v%)**	**alcohol (g/l)**	**t. acidity (g/l)**	**pH**	**polimer%**	**colour hue**	**cis-piceid (mg/l)**	**trans-piceid (mg/l)**	**cis-resveratrol (mg/l**
Sopron	13.39	28.9	5.61	3.23	39.06	0.610	0.6	0.8	0.7
Balatonboglár	12.42	25.8	7.26	3.07	48.52	0.550	0.6	0.8	0.4
Villány	13.65	25.5	5.65	3.38	75.74	0.820	0.6	0.7	0.8
Eger	11.48	24.0	6.98	3.20	36.05	0.490	0.5	0.5	0.5
Alföld Kiskőrös	13.65	27.9	5.56	3.24	55.63	0.590	0.5	0.6	0.5
average	12.92	26.42	6.21	3.22	51.00	0.61	0.56	0.68	0.58
Standard deviation	0.950037	1.963925	0.835386	0.110589	15.85655	0.12498	0.054772	0.130384048	0.164316767
Cabernet Franc	**alcohol (v/v%)**	**alcohol (g/l)**	**t. acidity (g/l)**	**pH**	**polimer%**	**colour hue**	**cis-piceid (mg/l)**	**trans-piceid (mg/l)**	**cis-resveratrol (mg/l)**
Sopron									
Balatonboglár									
Villány	15.39	34.4	4.90	3.67	54.37	0.920	0.5	0.6	0.7
Eger	14.02	25.3	6.60	3.41	67.75	0.790	0.5	0.5	0.4
Alföld Kiskőrös	13.39	32.6	6.41	3.39	55.31	0.690	0.5	0.6	0.7
average	14.27	30.77	5.97	3.49	59.14	0.80	0.50	0.57	0.60
Standard deviation	1.022562	4.819059	0.931504	0.156205	7.468396	0.115326	0	0.057735027	0.173205081
Portugieser	**alcohol (v/v%)**	**alcohol (g/l)**	**t. acidity (g/l)**	**pH**	**polimer%**	**colour hue**	**cis-piceid (mg/l)**	**trans-piceid (mg/l)**	**cis-resveratrol (mg/l)**
Villány	13.65	29.4	8.58	3.52	61.36	0.720	0.0	0.4	0.9

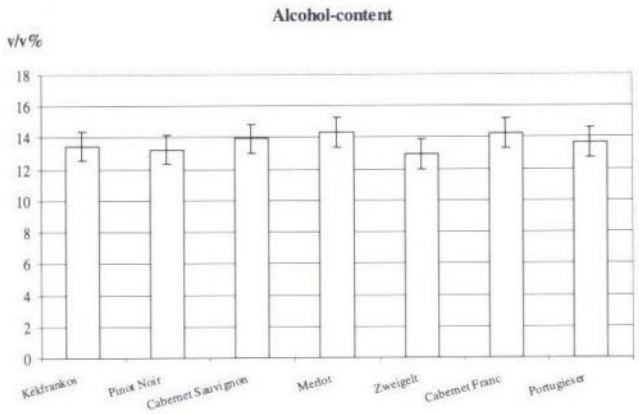

Figure 1.: Alcohol-content in"Wineregion"

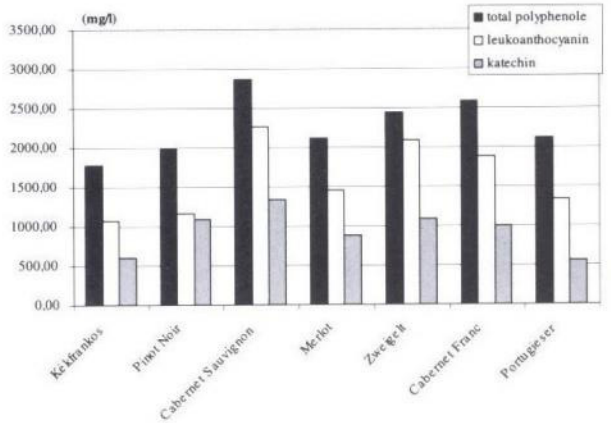

Figure 2.: Total phenol. leukoanthocyanin. katechin-content in "Wineregion"

Pinot Noir, Kékfrankos, Merlot, (Portugieser), Cabernet Franc, Zweigelt and Cabernet Sauvignon. As for Portugieser, we had only 1 sample item available, so it doesn't form part of the evaluation but the absolute value.

Based on *(Figure 3.)*, the order by antocyanin-concentration and colour intensity is: Cabernet Sauvignon, Zweigelt, Cabernet Franc, Merlot, Kékfrankos and Pinot Noir.

Figure 4. shows the order by resveratrol concentration, in ascending order:

Cabernet Franc, Zweigelt, Pinot Noir, Cabernet Sauvignon, Merlot and Kékfrankos. Despite the fact that Portugieser represents the highest absolute value, it's statistically invaluable (for having only 1 sample item). The relatively high t-resveratrol concentration of the wines Merlot and Kékfrankos has already been proven in the course of several other kinds of experiments of ours.

The development of the antocyanin concentration is illustrated by Chart 5. The significant difference presented on

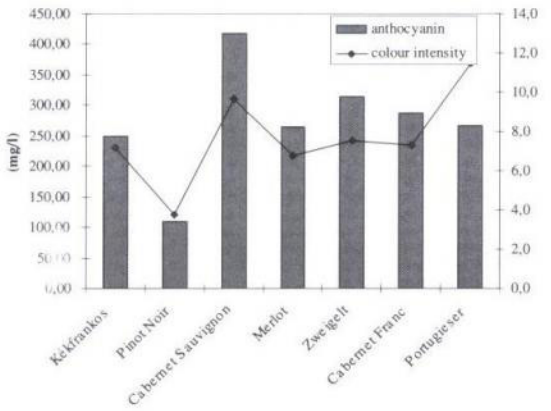

Figure 3.: Anthocyanin and colour intensity in "Wineregion"

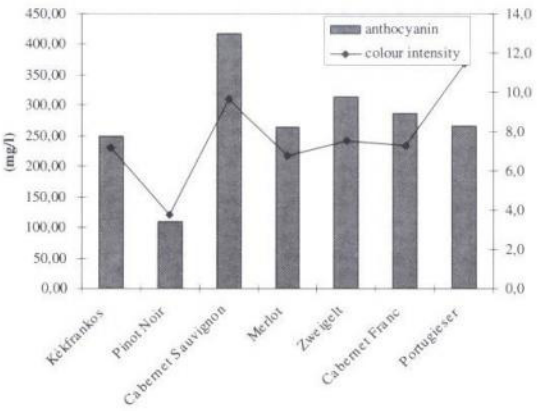

Figure 4.: Trans-rezveratrol

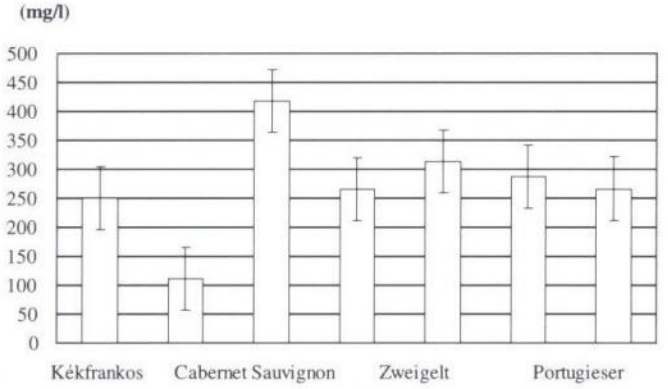

Figure 5.: Anthocyanin in "Wineregion"

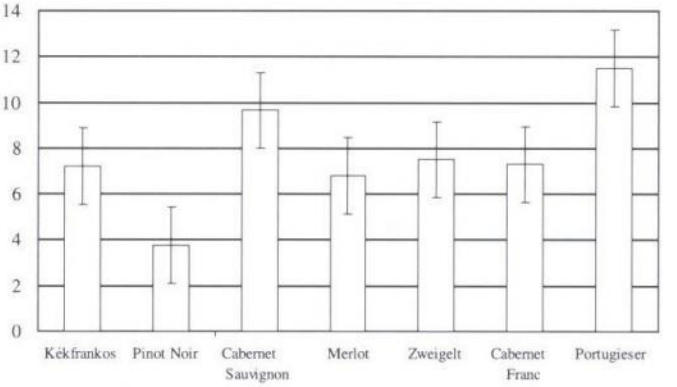

Figure 6.: Colour intensity

the basis of the statistical evaluation can be detected among the samples. This difference among the samples can be calculated by the „significance semi-matrix". As a result, on a significance level of 95%, there is significant difference detected among the sample pairs below:

Cabernet Franc – Zweigelt

Cabernet Franc – Kékfrankos

Cabernet Franc – Pinot Noir

Cabernet Franc – Cabernet Sauvignon

Cabernet Franc – Merlot,

Certainly, we did not analyse the Portugieser sample, as we had only 1 sample item available thereof.

In the colour intensity values, there is also significant difference detected on a significance level of 95% (Chart 6). We show the difference among sample pairs deriving from the „significance semi-matrix", without taking the Portugieser into account.

Pinot Noir – Zweigelt

Cabernet Franc – Pinot Noir

Pinot Noir – Kékfrankos

Pinot Noir – Cabernet Sauvignon

As a whole, we can state that the highest polyphenolic composition – total polyphenolics, leucoantocyanins, catechins – was typical of the Cabernet Sauvignon, Cabernet Franc and Zweigelt samples. However, we could not detect a really significant difference in these parameters.

In the case of antocyanin concentration and colour intensity, we have managed to detect a significant difference, where the highest antocyanin content was characteristic of the Cabernet Sauvignon samples.

As regards the t-resveratrol quantity in case of „Wine Regions", the following trend can be seen: Villány and Sopron contained remarkable quantities. Concerning the t-resveratrol quantity of the grape varieties, the order is as follows: Merlot, Kékfrankos and Pinot Noir are the ones which contained t-resveratrol to the highest degree.

We can state that Hungarian wines contain an average concentration of 1–3 mg/l t-resveratrol.

Conclusion

In our present work, we have compared some red wines of some „Wine Regions". We were looking for an answer to the question whether there is a significant difference among varieties concerning their polyphenolic composition. Our analyses were focused on the physiologically important resveratrol. Our measurement results show that except the antocyanin concentration and the colour intensity values, there is no significant difference among the varieties.

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