

COMPARISON OF WINES OF GEORGIAN (KAKHETHIAN) AND EUROPEAN TYPES ACCORDING TO QUANTITATIVE CONTENT OF PHENOLIC COMPOUNDS AND ANTIRADICAL EFFICENCY

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ABSTRACT

On the basis of the archeological data, the winemaking in Georgia originates 8000 years ago. According to ancient Georgian traditional technology of preparation of the wine, the squeezed grape is placed in a clay vessel (qvevri) dug in the ground, and alcoholic fermentation is carried out together with components of a cluster. During fermentation, from a stem, skin and seeds of a grape in plenty are extracted phenolic compounds which define the composition and essence of Kakhethian wine. The aim of this study is to estimate total amount of phenols, catechins, proanthocyanidins and anthocyanins in wines of the European and Kakhethian type, and to compare their anti-radical efficiency with the help of 2,2-diphenyl-1-picrylhydrazyl (DPPH*) radical. According to the received data, by comparison of European and Kakhethian wines for the content in them of phenolic compounds, Kakhethian wines considerably surpass the European wines. In the Kakhethian white wines prepared from various grapes, the total content of phenolic compounds varies within the limits of 1330-2430 mg/L, and in the European white wines - within the limits of 210-468 mg/L. In the Kakhethian red wines these limits make 2898-4416 mg/L, and in the European red wines – 1630-2340 mg/L. The high content of phenolic compounds in the Kakhethian red wines specifies their medical and preventive properties that proves to be true by comparison of anti-radical efficiency of European and Kakhethian wines.

Se référant aux techniques traditionnelles anciennes géorgiennes de production du vin, le raisin pressé est placé dans un pot en argile enterré (kvevri) pour donner lieu à une fermentation alcoolique avec les constituants de la grappe de raisin. Lors du processus de la fermentation, la râpe, la pelure du raisin et le pépin dégagent en grande quantité des composés polyphénoliques définissant la composition et l'esprit du vin du type kakhétien. Se référant aux données obtenues et en comparant la teneur quantitative des composés phénoliques dans les vins des types kakhétien et européen, il a été mis en évidence que les vins du type kakhétien excèdent considérablement les vins du type européen par leur teneur en phénols. Dans les vins blancs du type kakhétien produits à partir de diverses espèces de raisin le total des phénols varie de 1330 mg à 2430 mg par litre alors que le même indice dans les vins blancs du type européen varie de 210 mg à 468 mg par litre. Dans les vins rouges du type kakhétien la différence se situe entre 2898 mg et 4416 mg par litre contre 1630 mg et 2340 mg par litre pour les vins rouges du type européen. C'est notamment cette teneur en grande quantité des polyphénols que confère aux vins du type kakhétien une qualité curative et préventive ce qui s'affirme d'ailleurs par les résultats d'une comparaison des effets anti-radicaux des vins des types kakhétien et européen.

INTRODUCTION

Adverse ecological conditions observable in the most part of the modern world, unbalanced nutrition and the various illnesses break the counterbalanced free-radical processes proceeding in living cells. The reasons causing this problem are pollution of an environment, the stressful influences, radiation, chronic intoxications, smoking and other conditions, as a result of which the uncontrollable free-radical reactions develop. Under influence of these reactions the toxic effects of xenobiotics are amplified, and carcinogenesis, mutagenesis, atherosclerosis and autoimmune diseases are stimulated. In this respect, the rather important factor is revealing such foodstuff which contains in large quantities compounds having antioxidant activity. In this aspect the Kakhethian technology of making of wine which is since old days used in Georgia is especially interesting (Beridze, 1970) the wines, exclusively rich with flavonoids, are prepared by means of this technology which are characterized by high antioxidant activity and considerably reduce injury of tissues and cells (Middleton, et al. 2000, Robles-Sardin, et al. 2010, Williams, et al. 1997).

According to ancient Kakhethian traditional technology of making of the wine, the crushed grape is placed in a clay vessel (qvevri) dug in the ground, and alcoholic fermentation is carried out together with components of a cluster. During fermentation, from a stem, skin and seeds of a grape in plenty are extracted phenolic compounds which define the composition and essence of Kakhethian wine, and interaction of these compounds with the oxidizing enzymes contained in a skin, pulp and stem of a grape, defines taste and aroma, characteristic for Kakhethian wine. Grape seeds play a main role during formation of a wine of the Kakhethian type as seeds basically increase the content of phenolic compounds in a wine, and give to it characteristic aroma. Stems promote clarification of a wine and enrich it with flavonoids and extractive substances. The grape skin gives to a wine gentle, specific, varietal aroma. Flavonoid compounds define character of a wine, at ageing these compounds are oxidized, therefore the wine becomes soft, velvety and pleasant taste. Quality of the wine made in a clay vessel (qvevri) is much better than quality of the wine made in a wooden vessel. The wine made in qvevri has specific fruit taste and is characterized by integrity. Therefore, qvevri is the best vessel for making of the Kakhethian wine. Besides, its advantage consists still that in the qvevri dug in the ground fermentation occurs in more normal conditions, because of smaller fluctuation of temperature.

The aim of this study is to estimate total amount of phenols, catechins, proanthocyanidins and anthocyanins in wines of the European and Kakhethian type, and to compare their anti-radical efficiency in the system forming a radical of 2,2-diphenyl-1-picrylhydrazyl (DPPH*).

MATERIAL AND METHODS

White and red wines of the Kakhethian and European type, made of a grape varieties (*Vitis vinifera L.*) cultivated in Georgia, Italy and Slovenia, were given to us by wineries and physical persons whom we express gratitude. The technology of making of white (W) and red (R) wines, variety of a grape, the country of cultivation, wine factory and year of making are given in Tab. 1.

In samples of analyzed wines the total content of phenols is determined with Folin-Chiocalteu reagent (Singleton, & Rossi, 1965), catechins and proanthocyanidins - by method of Swain & Hillis, 1959), anthocyanins – by Durmishidze & Sophromadze (1983). Standard

curves are constructed: for phenols - on a basis of gallic acid ("Sigma", maximum absorption 765 nm), for catechins - on a basis of (+) catechin ("Theodor Schuchard", maximum absorption 500 nm), for proanthocyanidins - on a basis of cyanidin isolated from a grape skin (maximum absorption 548 nm) and for anthocyanins - on a basis of malvidin-3-monoglucoside (maximum absorption 536 nm).

For determination of antiradical efficiency of wine the stable radical of 2,2-diphenyl-1-picrylhydrazyl (DPPH*) with maximum absorption 520 nm was used (Sanchez-Moreno, et al. 1998). Alcohol was removed from samples of analyzed wine (50 ml) by evaporation on the vacuum - rotational evaporator at 40°C and after that the volume of samples again supplemented up to initial volume with distilled water. Spectrophotometric measurements were carried out on spectrophotometer CФ-26 (Russia). Each experimental variant was repeated five times. Experimental data were processed statistically by computer program „MS Excel”.

Tab. 1

Wine Samples*

Wine	Tecnology of preparation	Grape Variety	Country and Winery	Vintage
1W	Kakhethian	<i>Rqatsitheli</i>	Georgia, Kakhethi, „Okros Kvanchkara”	2005
2W	Kakhethian	<i>Khikhvi</i>	Georgia, Kakhethi, Physical Person	2005
3W	Kakhethian	<i>Ribolla</i>	Italia, Azenda Agricola, Osalavia Francesco Joško Grauner	2003
4W	European	<i>Rqatsitheli</i>	Georgia, Kakhethi, „Badagoni”	2005
5W	European	<i>Kakhuri Mtsvane</i>	Georgia, Kakhethi, Physical person	2004
6W	European	<i>Tsulukidzis tetra</i>	Georgia, Racha, „Okros Khvanchkara”	2004
7W	European	<i>Rebula</i>	Slovenia, Vipavska dolina, Vinorodni okolis, deadami Azelen	2006
8R	Kakhethian	<i>Sapheravi</i>	Georgia, Kakhethi, „Vazi+Ltd ”	2005
9R	Kakhethian	<i>Cabernet Sauvignon</i>	Georgia, Kakhethi, „Vazi+Ltd “	2005
10R	Kakhethian	<i>Ojaleshi</i>	Georgia, Samegrelo, „Vazi+Ltd ”	2005
11R	European	<i>Sapheravi</i>	Georgia, Kakhethi, Physical person	2007
12R	European	<i>Aleksandreuli</i>	Georgia, Racha, „Okros Khvanchkara ”	2003
13R	European	<i>Merlot</i>	Georgia, Kakhethi, „Besini ”	2008

* W – White wine; R – Red wine

RESULTS AND DISCUSSION

According to the received data (Tab. 2), by comparison of the quantitative content of phenolic compounds in the wines made on the Kakhethian and European technologies it is obviously visible that white and red wines of the Kakhethian type (1W, 2W, 3W, 8R, 9R, 10R), under the content of phenolic compounds considerably surpass to the appropriate wines made on the European technology (4W, 5W, 6W, 7W, 11R, 12R, 13R).

In white wines made on the Kakhethian technology, the total content of phenols varies from 1296 mg (a wine 3W) up to 2290 mg (a wine 1W), and in red wines of the Kakhethian type - from 2848 mg (a wine 10R) up to 4416 mg (a wine 8R) per liter, while in white wines made on the European technology, these parameters varies from 210 mg (a wine 7W) up to 456 mg (a wine 6W), and in red wines of the European type - from 1630 mg (a wine 12R) up to 3130

mg (a wine 11R) per liter. Thus, as a result of making of a wine by the Kakhethian technology, the wine is considerably enriched by phenolic compounds.

Tab. 2

The Content of Total Phenolics, Catechins, Proanthocyanidins, Antocyanins in White and Red Wines and Antiradical Efficiency

Wine	Total phenolics, mg/l	Catechins, mg/l	Proanthocyanidins, mg/l	Anthocyanins, mg/l	EC ₅₀ (g antioxidant Kg ⁻¹ DPPH*)	T _{EC50} (min)	AE (x10 ⁻³)
1w	2290±38	640±007	690±7.1		510±11.7	4.5	0.44
2w	2000±13	453±01	1097±2.4		515±16	4.5	0.43
3w	1296±46	509±4	392±16		847±35	5	0.23
4w	346±11	39±1	47.8±2		1191±14.1	5	0.16
5w	278±7	27±2	43.2±1.1		1447±22.4	5	0.12
6w	456±26	77±2	165±8.7		1219±18.4	4.5	0.18
7w	210±4	8±2	-		893±13.3	5	0.038
8R	4416±100	1010±23	1203±15	1270±45	516±16.5	3.2	0.62
9R	2848±72	798±2	728±13	317±24	382±14.9	5	0.52
10R	3700±85	862±11	872±18	414±20	342±11.5	5	0.58
11R	3130±76	582±5	610±55	1456±36	519±5.1	4.4	0.43
12R	1630±50	378±15	980±69	53.2±5	595±14.5	5	0.34
13R	2318±73	636±33	826±4	322±18	880±4.3	4.5	0.25
α-Tocopherol					625±22.7	5	0.32

Special interest represents study of phenolic compounds of the wines made on the Kakhethian and European technologies, from the same variety of a grape. We have compared the wines made by the Kakhethian (1W, 3W, 8R) and European (4W, 7W, 11R) technology, from the autochthonous Georgian varieties of grape (*Rqatsitheli*, *Sapheravi*) and from a variety of a grape cultivated in some countries of Europe (*Ribolla*) (Tab. 2).

In the Kakhethian wine (1W), made of a variety of a grape *Rqatsitheli*, total amount of phenolic compounds, catechins and proanthocyanidins, is higher in 6.6 times, 16 times, and 15 times, respectively, than in a wine (4W), made from the same variety of a grape by the European technology. Approximately the same patterns of relationship is observed and in case of the wines made in Italy by the Kakhethian technology from a grape of variety *Ribolla*, and in Slovenia from the same variety of a grape by the European technology. In the wine made by the Kakhethian technology (3W), the total phenols are 1296 mg/l, and in the wine made by the European technology (7W) from the same variety of a grape, this parameter is 210 mg/l. Kakhethian wine (3W) contains catechins and proanthocyanidins in amount of 509 mg/l and 392 mg/l, respectively, and the wine made of the same variety of a grape by the European technology (7W) contains only insignificant amount of catechins, and practically does not contain proanthocyanidins.

Among the white wines made by Kakhethian technology, the high content of proanthocyanidins characterizes a wine (2W), made of the autochthonous Georgian variety of grape *Khikhvi*, in which the content of these compounds makes 1097 mg/l, and the content of catechins - 453 mg/l (the total of phenols in this wine makes 2 g/l). The wine made by Kakhethian technology from a grape variety *Khikhvi* has straw color, contains high amount of extractive substances, is perfect and harmonious (Tabidze, 1954).

Among the white wines made from the autochthonous Georgian varieties of a grape by the European technology, the wine (6W), made of a grape variety *Tsulukidzis Tetra* is distinguished, in which the total content of phenols makes 456 mg/l, catechins - 77 mg/l, and proanthocyanidins - 165 mg/l. Also it is necessary to note a wine (5W) made of a grape variety *Kakhuri Mtsvane* by the European technology, in which the total content of phenols makes 278 mg/l, catechins - 27 mg/l, and proanthocyanidins - 43.2 mg/l. This wine tastes very gentle and aromatic.

In red wine made from grape variety *Sapheravi* by Kakhethian (8R) and European (11R) technologies, the total content of phenols makes 4416 mg/l and 3130 mg/l, respectively. The total content of catechins and proanthocyanidins in wine made by Kakhethian technology is 1.7 times and 2 times higher, than in wine made by European technology.

Partially other interrelation is observed in a case of anthocyanins. In red wine made by European technology (11R) the content of anthocyanins is 1456 mg/l, but in red wine made by Kakhethian technology (8R) their content makes 1270 mg/l. The reason of it, apparently, consists in long process of maceration in a qvevri that causes sedimentation of anthocyanins.

Among autochthonous red varieties of grape of Georgia is distinguished *Ojaleshi*. This wine made by Kakhethian technology is characterized by good color, typical viscosity for red wines, extract content, and harmonicity (Ramishvili, 1948). In this wine (10R), the total content of phenols makes 3700 mg/l, catechins - 862 mg/l, proanthocyanidins - 872 mg/l, and anthocyanins - 414 mg/l.

The characteristic of the wine made by Kakhethian technology from of the French variety of grape *Cabernet Sauvignon* cultivated in Georgia is also important. In this wine (9R), the total content of phenols makes 2848 mg/l, catechins - 798 mg/l, proanthocyanidins - 728 mg/l, and anthocyanins - 317 mg/l. Comparison of the wine made by European technology from the French variety of grape *Merlot* cultivated in Georgia with the wine made in Argentina from the same variety of a grape and by the same technology is rather interesting. In wine made in Kakhethi (13R) the total content of phenols, catechins, proanthocyanidins and anthocyanins makes 2318 mg/l, 636 mg/l, 826 mg/l and 322 mg/l, respectively (Tab. 2), while in red wine made in Argentina the total content of phenols is 1637 mg/l, the total content of catechins and proanthocyanidins is 13.30 mg/l, and content of anthocyanins is 52.61 mg/l (Sanches-Moreno et al., 2003). It is necessary to note also the wine (12R) made from the autochthonous Georgian variety of a grape *Aleksandreuli*, which is characterized by pleasant taste and delicate aroma. In this wine the total content of phenols makes 1630 mg/l, catechins - 378 mg/l, proanthocyanidins - 980 mg/l, and anthocyanins - 53.2 mg/l.

Thus, study of total amount of phenolic compounds, catechins, proanthocyanidins, and anthocyanins of white and red wines prepared by Kakhethian and European technologies has demonstrated that by content of said compounds Kakhethian wines considerably exceed European wines, that specifies in high medical and prophylactic properties of wines of Kakhethian type. This conclusion has been confirmed by investigation of antiradical efficiency (AE) of wines of Kakhethian and European type (Tab. 2). According to these findings average value of AE of white and red wines of Kakhethian type 2.3 and 1.7 times exceeds average value of AE of white and red wines of European type, respectively. Among wines, the white (1W, 2W) and red (8R, 9R, 10R) wines of Kakhethian type are especially distinguished by their antiradical efficiency.

The data of some authors on the enrichment of white wines of European type by polyphenols should be noted (Williams & Elliot, 1997) have carried out researches whose goal was the development of a seed-enhanced (polyphenol-enriched) white wine with greater positive health potential. This, seed-enhanced white wine is currently being evaluated in human

subjects with regard to its potential effect on the cardiovascular system and inhibition of platelet aggregation. According to Auger et al. (2005), white wine prepared from grape variety *Chardonnay*, enriched by polyphenols possesses a protective effect against early forms of atherosclerosis in hamsters, and according to Fuhrman et al. (2001), the white wine enriched by polyphenols possesses similar to red wine antiradical properties.

CONCLUSIONS

As a result of comparison of white and red wines made by Kakhethian and European technologies it is shown that by content of total amount of phenolic compounds, catechines, proanthocyanidins, and anthocyanins, and by antiradical efficiency, the Kakhethian white and red wines considerably exceed European white and red wines, that specifies in high medical and prophylactic properties of wines of Kakhethian type.

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