



THE OPTIMIZATION OF ANTIOXIDANTS EXTRACTION FROM WINERY WASTE

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Abstract: Winery waste represents an important source of bioactive compounds that exhibit antioxidant properties and important antiradical activity. The concentration of ethyl alcohol in extraction environment has a significant influence on the rate of extraction and bioactive compounds as well as onto the measured parameters: the antioxidant activity of water-soluble substances; the antioxidant activity of fat-soluble substances; the total content of anthocyanins; total polyphenol index; the amount of tannins extracted; the DPPH antiradical activity and the ability of inhibiting hydrogen peroxide in an acid and in basic medium. On the basis of experimental data and by using the corresponding calculation relations, the first-order statistical characteristics were determined. The analysis of the trial data showed the existence of non-linear dependencies therefore, the spline functions were used to establish the mathematical models. The analysis of the information provided by the experimental data obtained allows the determining of influences of various parameters measured during the tests.

Introduction

The vegetable matter is essential for the survival of the entire population, being a source of aromatic, pharmaceutical and industrial compounds, and biologically active substances. These natural and biologically active substances can not be synthesized by the human body, therefore they must be consumed daily. The use of natural antioxidants in the diet, such as phenolic compounds, carotenoids, vitamins, minerals and other pigments, neutralizes the action of active forms of oxygen. The results of numerous clinical and epidemiological observations indicate an inverse dependence between the risk of developing cardiovascular disease, carcinogenic, and the presence in the diet of food rich in bioactive compounds with antioxidant properties diversity of natural polyphenols and health benefits being extensively researched. It was proved that the phenolic compounds of grapes as hydroxy acids (caffeic, coumaric, ferulic, caftaric, cutaric), flavanols (catechin, epicatechin), flavonols (quercetin) possess health improving properties, which are important in inhibiting carcinogenesis, mutagenesis, cardiovascular diseases, have the ability to inhibit peptic ulcer, different types of diabetes and even in lipid bio-absorption. These polyphenols have been associated with their antioxidant activities in vivo and in vitro. In vivo study demonstrate that the antioxidant effects of proanthocyanidins from grape seeds have a better protective effect than vitamin E, vitamin C, vitamin E, C and β -carotene against lipid peroxidation and DNA fragmentation in mice. By processing grapes, about 20-25% wine valuable raw material is usually obtained, consisting of secondary wine products (pomace, grape seed, etc.) rich in polyphenols.

Material and method

In order to obtain extracts, locally produced dried red grape winery waste was grounded to powder and sifted. The obtained powder was subjected to extraction using ethanol hydro-alcoholic medium in different concentrations: 20, 40, 50, 60, 80 and 96 %. The extraction was performed in solid-liquid ratio 1:8 for an hour in a dark place at laboratory temperature. The obtained extracts were filtered and transferred to the dark color packaging. The samples of winery waste extracts were kept in a dark place at a temperature of 4 ± 1 °C.

Table 1. Following indices were determined in the extracts of winery waste.

Index	Description
P1 (AASH)	The antioxidant activity of water soluble substances, C_{AAS}
P2 (AASL)	The antioxidant activity of fat-soluble substances, TROLOX
P3 (CTA)	The total content of anthocyanins
P4 (IPT)	The index of total polyphenols
P5 (AAA)	The antiradical activity, DPPH, in the acidic medium
P6 (AAB)	The antiradical activity, DPPH, in the basic medium
P7 (CIPHA)	The ability to inhibit hydrogen peroxide in an acid medium
P8 (CIPHB)	The ability to inhibit hydrogen peroxide in basic medium
P9 (CTE)	The amount of extracted tannins

On the basis of experimental data and by using the corresponding calculation relations, the first-order statistical characteristics were obtained: averages, dispersion, standard deviation (r-squared), minimum, maximum and median value. The analysis of the trial data showed the existence of non-linear dependencies, therefore spline functions were used to establish mathematical models. The analysis of the information provided by experimental data obtained allows the determining of influences from various parameters measured during the tests.

Methods:

Determination of the amount of tannins
 Index of total polyphenols (IPT) or D280
 Determination of the total amount of anthocyanins - pH variation method
 Photochemluminescence PCL method
 Antioxidant action of soluble compounds in water (ACW)-principle of the method
 Antioxidant action of lipids soluble compounds (ACL).
 Antiradical activity using free radicals DPPH•.
 Antioxidant activity (HPSA). The method for determining the capacity of inhibiting the hydrogen peroxide

Results and discussions

Table 2 presents the scientific results obtained from the extracts of winery waste depending on the concentration of ethyl alcohol.

Table 2. The scientific results obtained from the extracts of winery waste depending on the concentration of ethyl alcohol

Concentration of ethyl alcohol, %	Amount of tannins, mg/3g	Index of total polyphenols	Total quantity of anthocyanins mg/3g	Antioxidant activity of lipid-soluble substances, TROLOX, mg/3g	Antioxidant activity of water soluble substances, mg/3g
20	9.09±0.05	6.47±0.23	0.11±0.01	4.46±0.12	0.08±0.01
40	10.33±0.05	11.83±0.33	0.75±0.02	5.71±0.06	0.23±0.01
50	11.36±0.06	19.00±0.30	0.86±0.02	3.45±0.02	0.19±0.01
60	11.09±0.06	13.50±0.50	1.29±0.02	5.59±0.07	0.39±0.02
80	10.35±0.03	8.30±0.20	0.82±0.01	4.09±0.08	0.21±0.01
96	7.81±0.02	8.7±0.20	0.09±0.01	1.83±0.04	0.10±0.01

Analyzing the obtained data, the content of anthocyanins from the six concentrations of ethyl alcohol was determined. The values ranged between 0.09 mg/3g and 1.29 mg/3g. The highest anthocyanin content identified in the extract from winery waste has been determined for the concentration of 60%, followed by the one of 50%. The lowest value of the anthocyanin content -0.09 mg/3g was registered at the concentration of ethyl alcohol of 96%. The quantity of anthocyanins extracted from winery waste solution of ethyl alcohol 60% is 14.33 times higher than the one extracted with ethyl alcohol of 96% and 11.73 times higher than the one extracted with ethyl alcohol of 20%.

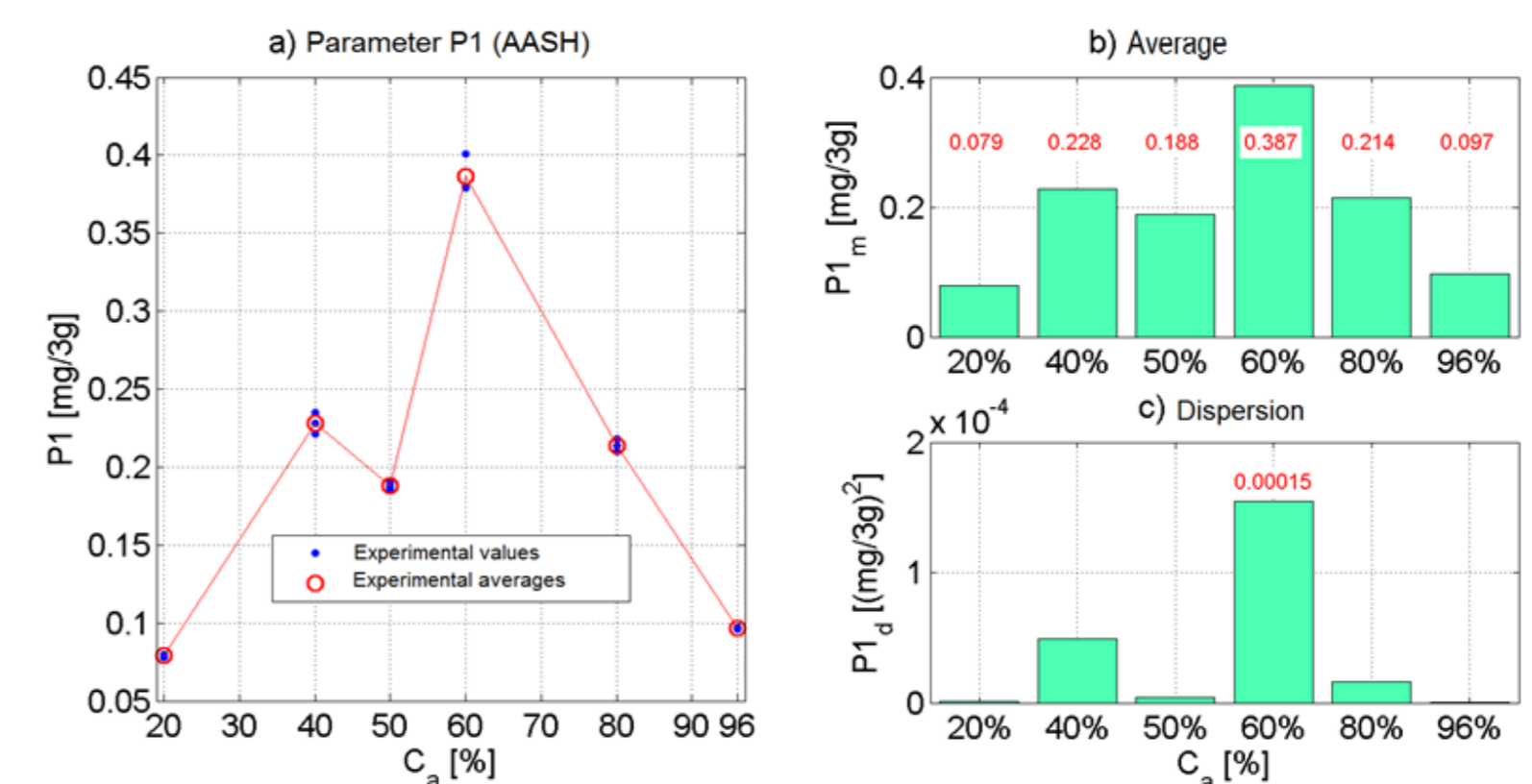


Fig. 1. The values, the average and dispersion of P1 (antioxidant activity of water-soluble substances, AASH) winery waste, depending on ethanol concentration.

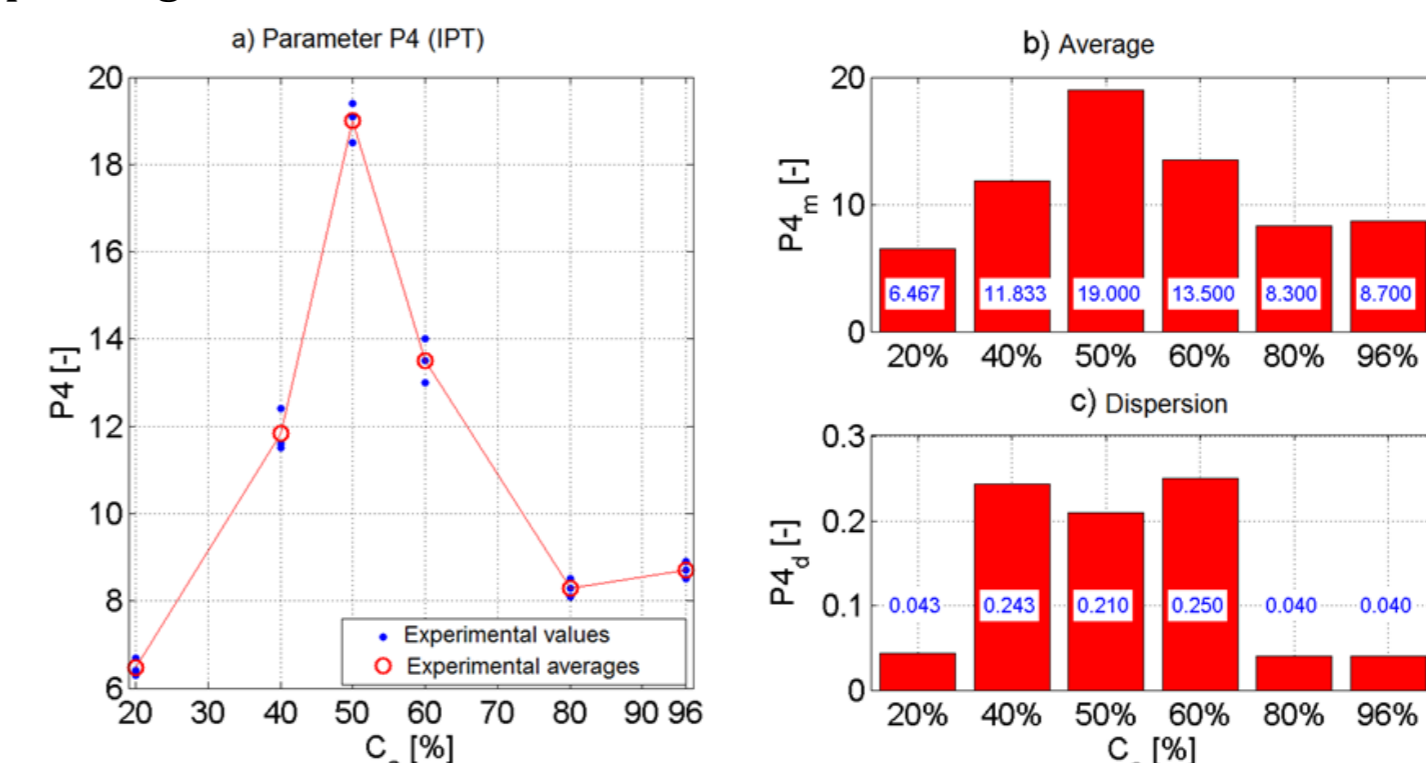


Fig. 4. The values, the average and the dispersion of P4 (index of total polyphenols, IPT) winery waste, depending on ethanol concentration.

Conclusions

The experimental results and analysis of the information lead to the following conclusions:

Winery waste is an important source of bioactive compounds that exhibit significant antiradical and antioxidant properties. The concentration of ethyl alcohol in the extraction environment has a significant influence on the rate of extraction of bioactive compounds and all the parameters measure was ascertained between antioxidant activity of water-soluble substances and the total content of anthocyanins, as well as between the antiradical activity in acidic activity and antiradical activity in the basic environment.

Acknowledgement: The project is co-funded by the AUF project (BECO-U-56135FT205 2012-53).