EXTRACTION OF ANTIOXIDANT AND POLYPHENOL COMPOUNDS FROM TOKAJI ASZÚ MARC

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Abstract
Grapes and even wine-making wastes such as marc and stalks, are rich in phenols. The polyphenolic content has many favourable effects on human health,such as the anti-carcinogenic effects and the inhibition of the oxidization of low-density lipoproteins. An antioxidant is a molecule that hinder the oxidation of unlike molecules. Ouraim was to find the optimal conditions of the extraction of the antioxidant and phenolic compounds from Tokaji aszú marc. Absolute ethanol and deionized water were used to prepare the solvent, 4:1 solvent-to-sample ratio was choosen. The solvent contains different volumes of ethanol (0 – 25 – 50 – 75 – 100%). The temperature was 30 °C and 60 °C.The time of the extraction was half-, one-, two-, three-, four- and five-hours long. The extractions were more efficiency using ethanol solvent compared with the water solvent. In all casesthe phenol concentration and antioxidant capacity were two and three times higher at higher temperature (60 °C) than at lower temperature (30 °C).The maximum value of total phenol content (67830±509 μM GS/L) was reached at 60 °C temperature, 25% ethanol solvent after 3 hours.The maximum value of antioxidant capacity (11126±145 μM AS/L) was reached at 60 °C temperature, 50% ethanol solvent after 5 hours.

Introduction
Grapes are one of the world's largest fruit crops, and even wine-making wastes such as marc (theremainsofgrapesorotherfruitthathavebeenpressedforwine-making) and stalks, are rich in phenols. Grapes, wine, grape seeds and skins extracts have many favourable effects on human health due to their polyphenol content,such as the anti-carcinogenic effects and the inhibition of the oxidization of low-density lipoproteins, thereby decreasing the risk ofcardiovascular diseases.Therefore, phenolic compounds can be considered to be added-value by products,corroborating their isolation from the industrial waste [1, 2].
Furthermore, the activity of these compounds as food lipid antioxidants is well known. By adding antioxidants is a method which is increase the shelf life, especially of fats, oil and fat containing food products. Since synthetic antioxidants, such as BHA and BHT have restricted use in foods because their toxicological effects on different species and suspected carcinogenic potential, the search of natural and safe antioxidants, especially of plant origin, has increased latterly [1].
The goalof an extraction process is to provide the maximum yield of substances and of the highest quality (concentration of phenolic compoundsand antioxidant power of the extracts).

Experimental
The marc of Tokaji aszú was provided by theFitomark Ltd. (Tolcsva). The marc wasstored in freezer till the experiments.

Extraction measurements

Our aim was to find the optimal conditions of the extraction from Tokaji aszú marc. Absolute ethanol and deionized water were used to prepare the solvent,4:1 solvent-to-sample ratio was choosen. Continuous stirring was ensured during all experiments. The picture and flow sheet of the equipment can be seen at Figure 1.

In the experiments three parameters of the extraction were changed: the temperature, the solvent concentration and the time of the extraction. The temperature was 30 °C and 60 °C. To keep the temperature at constant value a Lauda Ecoline E100 Immersion Thermostat was used. The solvent contains different volumes of ethanol (0 – 25 – 50 – 75 – 100%). The time of the extraction was half-, one-, two-, three-, four- and five-hours long.

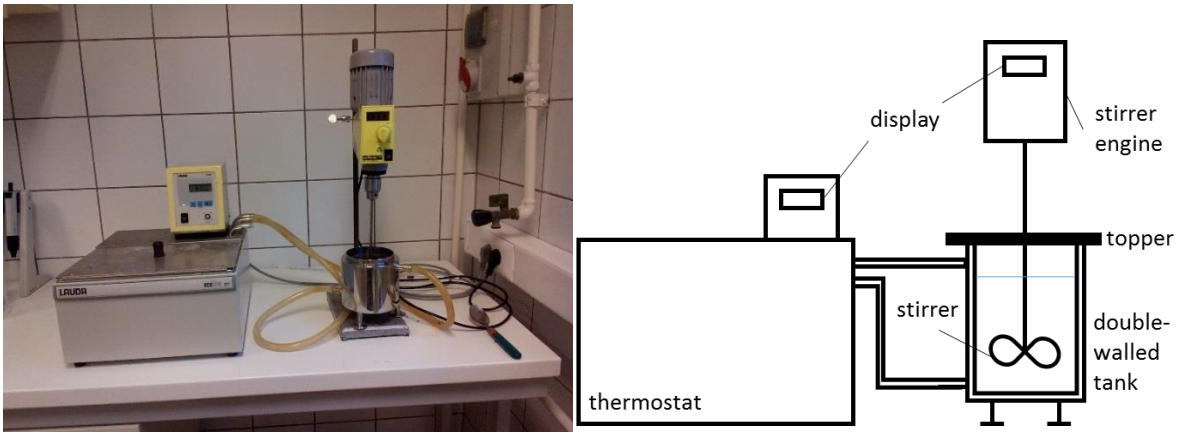


Figure 1. (a) picture of the experimental equipment and (b) flow sheetof the experimental equipment

Analitical measurements

The TPC, FRAP assays were run with a Nicolet Evolution 300 BB type spectrophotometer (Thermo Electron Corporation, Cambridge, UK) at the respective wavelengths. Measurements were run triplicate.

Analysis of total phenol content (TPC)

Total phenol content was determined by the Folin-Ciocalteu assay [3] applying gallic acid as the standard at 760 nm. Total phenol content was expressed in µmol equivalents of gallic acid (GS)/L.

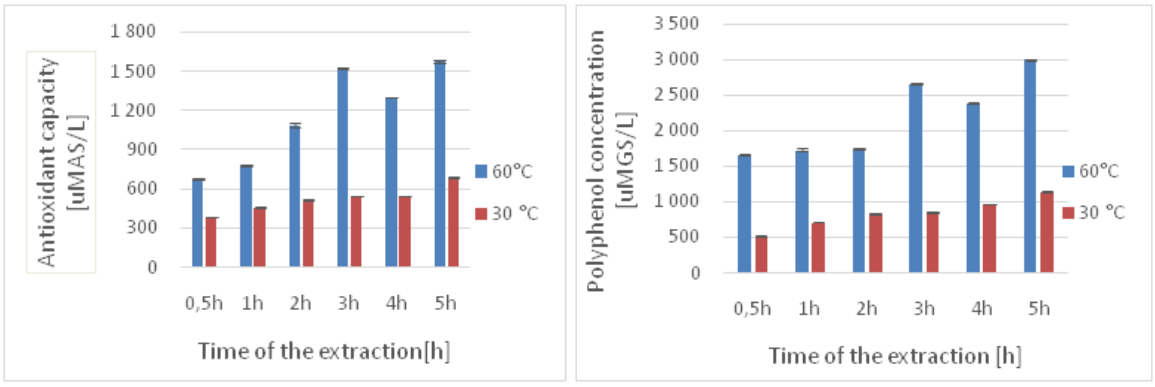
Antioxidant capacity measurements (FRAP)

The FRAP antioxidant capacity assay was run as described by Benzie and Strain [4] using ascorbic acid as standard. The absorbance was measured at 593 nm and results were determined in µmol equivalents of ascorbic acid (AS)/L.

Results and discussion

Figure 2. shows the phenol concentration (a) and antioxidant capacity (b) of the extracts in case of water solvent at different temperatures versus extraction time. At higher temperature (60 °C) the phenol concentration and antioxidant capacity were two and three times higher than at lower

temperature (30 °C). The extraction time generally increased the total phenol content and the antioxidant capacity. In case of water solvent the maximum values of polyphenol concentration was 3000±17 µM GS/L (after 5 hours), and the maximum of antioxidant capacity was 1575±33 µM AS/L (after 5 hours).

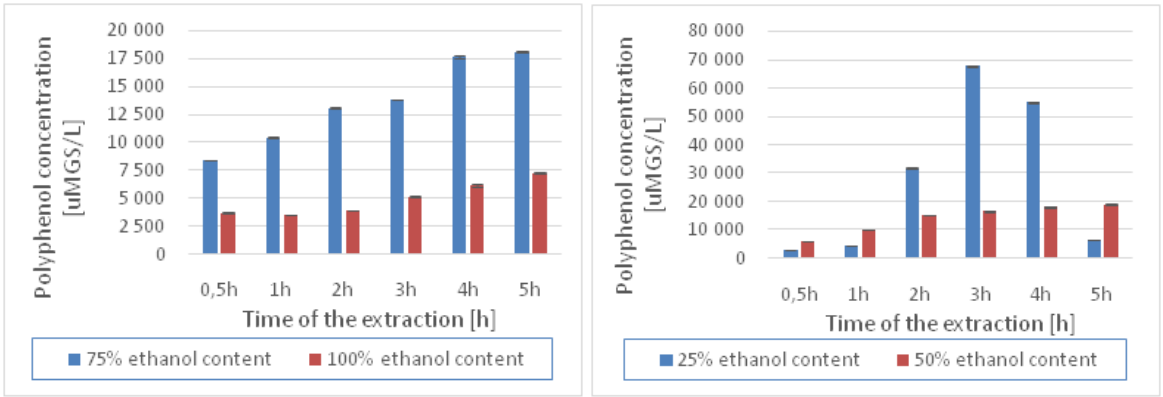


a) b)

Figure 2. (a) Polyphenol concentration incase of water solvent and b) Antioxidant capacity incase of water solvent

In our experiments the phenol content and the antioxidant capacity of the extracts were much higher at 60 °C temperature than at 30 °C temperature, therefore we only represent the results at 60 °C temperature comparing the various solvent concentrations.

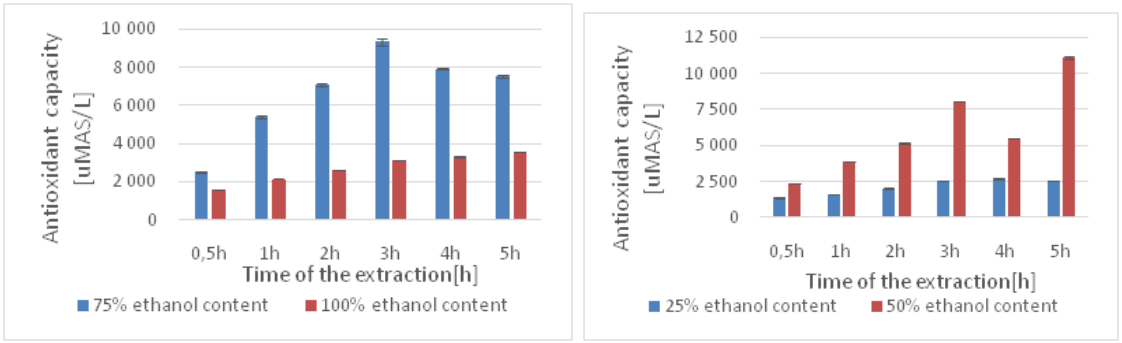
The total phenol content of the extracts can be seen in Figure 3. in case of 25% – 50% (a) and 75% – 100% (b) ethanol solvent versus extraction time. The different volumes of ethanol in the solvent reached a more varied result than the water solvent. In case of 25% ethanol solvent the total phenol content was increased in the first 3 hours and after that it was decreased. In case of 50%, 75% and 100% ethanol solvent a continuously raise was observed in total phenol content during the five hours. The maximum value of total phenol content (67830±509 µM GS/L) was reached at 60 °C temperature, 25% ethanol solvent after 3 hours.



a) b)

Figure 3. (a) Polyphenol concentration incase of 25% and 50% ethanol solvent (b) Polyphenol concentration incase of 75% and 100% ethanol solventat 60 °C temperature

The antioxidant capacity of the extracts can be seen in Figure 3. in case of 25% – 50% (a) and 75% – 100% (b) ethanol solvent versus extraction time.The same trend was observed in antioxidant capacity with 75% ethanol solvent than the total phenol content with 25% ethanol: the antioxidant capacity of the extracts was increased in the first 3 hours and after that it was decreased. In other cases the antioxidant capacity was increased with the extraction time. The maximum value of antioxidant capacity (11126±145µM AS/L) was reached at 60 °C temperature, 50% ethanol solvent after 5 hours.



a) b)

Figure 4. (a) Antioxidant capacity incase of 25% and 50% ethanol solvent (b) Antioxidant capacity incase of 75% and 100% ethanol solventat 60 °C temperature

Conclusion

The extraction experiments were achieved successfully. In all cases the concentration of total phenol content and antioxidant capacity was higher at higher temperature. The extractions were more efficiency using ethanol solvent compared with the water solvent. Determine the optimal parameters of the extraction (temperature, solvent concentration and extraction time) is not easy because of the different optimum of the total phenol content and antioxidant capacity.Comparing the proportions we can summarize that the total phenol content is about quarters of it's maximum value at the optimal parameters of antioxidant capacity (T = 60 °C, c_s= 50%, t = 5 hrs).The antioxidant capacity is about quarters of it's maximum value at the optimal parameters of total phenol content (T = 60 °C, c_s= 25%, t = 3 hrs). Our suggestion to choose the optimal operating parameters according to the more important component.

References

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