

## OPTIMIZATION OF THE EXTRACTION PROCESS OF BIOACTIVE COMPOUNDS FROM BY-PRODUCTS OF WINE PROCESSING

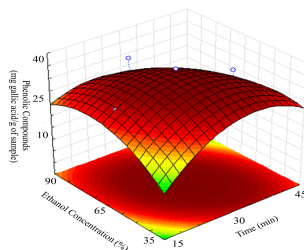
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**Abstract:** The aim of this study was to optimize the extraction of total phenolic compounds and antioxidant activity from the hydroalcoholic extract of grape marc from wine processing using full  $2^3$  factorial design and response surface methodology. Experimental data were processed using the STATISTICA 8.0 software and adjusted to a linear model. The dependent variables were the content of total phenolic compounds using the methodology of Folin-Ciocalteu [1], antioxidant activity using the DPPH free radical scavenging method [2]. The independent variables were temperature (35 to 85 °C), extraction time (15 to 45 min) and ethanol concentration (40 to 90%). The experimental design consisted of 18 experiments including four repetitions at the central point. Analysis of chemical characterization performed by high-performance liquid chromatography with photodiode array detection (HPLC-PDA) were carried out in optimal conditions of extraction. The extracts were prepared using 2 grams of grape marc and 25 mL of ethanol:water (80:20, v/v) and extracted in a water bath with temperature and time controlled as design. Then, samples were filtered and the assays performed in triplicate. The repeatability of the method was submitted to ANOVA and the results evaluated by response surface methodology to determine the influence of independent variables on the response variables. It was concluded that, at a 95% level of confidence, only the variable time was statistically significant for the extraction of total phenolic compounds ( $p \leq 0.05$  e  $p \leq 0.001$ ) with F value calculated 7.66 times higher than F tabular value. The higher extraction these phenolic compounds was obtained using time of 30 minutes (Figure 1). Thus, it is possible to suggest that the method of total phenolic compounds adjusts to model was statistically significant and predictive. Regarding the DPPH free radical scavenging method, none variable was significant. The extract evaluated in optimal condition (extraction time 30 min, ethanol concentration 65% and temperature 45°C) by HPLC-DAD presented two phenolic acids: caffeic acid ( $17.70 \mu\text{g ml}^{-1}$ ) and coumaric acid ( $46.13 \mu\text{g ml}^{-1}$ ) and stilbene trans-resveratrol ( $14.15 \mu\text{g ml}^{-1}$ ) (Figure 2). The results of this study confirm that the optimization of extraction conditions is important to obtain extracts rich in phenolic compounds and antioxidant activity from grape marc and this material is a potential source of bioactive compounds.

### References:

- [1] Singleton, V.L., Orthofer, R. and Lamuela-Raventós, R.M. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Methods Enzymol.* 299: 152-178.
- [2] Brand-Williams, W., Cuvelier, M.E. and Berset, C. 1995. Use of a free radical method to evaluate antioxidant activity. *LWT - Food Sci. Technol.* 28: 25-30.



**Figure 1:** Response surface for the effects of total phenolic compounds: (a) Ethanol concentration vs. time.