**Stability study as a basis for optimizing the extraction conditions of resveratrol and ε-viniferin**

Ema Kosovic1, Martin Topiař1

*1 Institute of Chemical Process Fundamentals of CAS v.v.i., Rozvojová 135, Prague 6, 16502, Czechia*

*\*Corresponding author: kosovic@icpf.cas.cz*

**Highlights**

* Comparison of several different extraction techniques under various conditions
* Performing the stability tests on extracted stilbenes
* Light induced formation of *cis*-viniferin by dimerization of resveratrol

**1. Introduction**

Czech Republic belongs among countries with long winemaking tradition. Unfortunately, the wine-processing industry produces large amounts of waste, which is usually burnt or used in compost [1]. However, wine waste formed during wine production can comprise valuable bioactive substances e.g., resveratrol and viniferin, important in food production and very useful in various branches of science including medicine, pharmacy and cosmetic industry thanks to their potential antioxidant activity [2]. The great interest in these compounds also stems from their biological properties and chemical stability. The mechanism that leads to production of these antimicrobial compounds synthesized and accumulated in different parts of the plant in response to biotic or abiotic stress is based on self-defense potential of the plants [3, 4]. This work is focused on the combination of several extraction techniques for the isolation of polyphenols with subsequent induction of stress conditions. Concern these factors, stability study is the basis for optimizing conditions of extraction methods of resveratrol and viniferin.



**Figure 1.** From grapes to main product and by-products.

**2. Methods**

To obtain a usable amount of stilbenes for subsequent processing it is essential to use highly effective extraction methods. Considering that, a comparison of several different extraction such as maceration, ultrasonic extraction, Soxhlet and pressured liquid extraction has been performed. The effects of stress conditions used in stability study were monitored and performed by newly developed HPLC-MS method, which is proved to be accurate, reproducible and efficient for determination of resveratrol and viniferin.

**3. Results and discussion**

Among extraction methods, the most noticeable results were obtained using maceration in dark and Soxhlet extraction method at atmospheric pressure providing the highest concentration of resveratrol. The outcome of stability study showed, that storage in dark didn’t affect the concentration of monitored compounds but light exposure induced visible decreasing in resveratrol and *trans*-viniferin concentration while the concentration of *cis*-viniferin increased considerably. The explanation of this phenomenon is lying in possible dimerization of two molecules of resveratrol induced by light and also photoisomerisation of *trans* form of stilbenes into *cis*, resulting in observable concentration increase of *cis*-viniferin [5]. To confirm this thesis, the sample without *cis*-viniferin was exposed to sunlight and UV light for defined time intervals. Process of formation of *cis*-viniferin was confirmed as well as supposed distinction between sunlight and UV light: UV light accelerates the entire process. Already known fact that higher temperature leads to degradation of stilbenes was also confirmed. Nevertheless, the short-time exposure of plant material to heating in solution enhances the release of resveratrol from other plant structures, such as carbohydrates.

**4. Conclusions**

Based on the stability study, the optimal conditions for obtaining individual stilbenes from grape cane extracts of *Vitis vinifera* L. cv. Cabernet Sauvignon were determined. Results showed that light-induced dimerization of *trans*-resveratrol leads to inverse concentration change between viniferin and resveratrol, while high temperature test showed complete degradation of stilbenes. Following photoisomerisation of *trans*-stilbenes into *cis* form also induced by light, explained the increase of *cis* form versus *trans.* Due to this, the basis for optimizing extraction conditions of resveratrol and viniferin was obtained.

**References**

1. C. Schonnenbeck, G. Trouve, M. Valente, P. Garra, J. F. Brilhac, Fuel 180 (2016) 324-331.
2. G.Y. Tang at al., Molecules 23 (2018) 2598.
3. M. Chalal et al., Molecules 19 (014) 7679-88.
4. G. Armijo et al., Front. Plant Sci. 7 (2016) 382.
5. L.M. Szewczuk, S.H. Lee, I.A. Blair, T.M. Penning, J. Nat. Prod. 68 (2005) 36-42.

**Acknowledgments**

The financial support from the Technological Agency of the Czech Republic via grant no. TJ01000249
is gratefully acknowledged.