



## ***LC with Multi-Channel EC for the Determination of Resveratrol in Wine, Grape Juice and Grape Seed Capsules Following Automated Solid Phase Extraction***

### **Purpose**

Explore liquid chromatography/electrochemistry with multi-channel electrode detection for the determination of resveratrol in natural products. Multi-channel detection allows the collection of significantly more information per sample.

Resveratrol (trans-3,5,4'-trihydroxystilbene) is an important phenolic antioxidant in wine. It has been reported that resveratrol may be effective in inhibiting platelet aggregation, altering eicosanoid synthesis and modulating lipoprotein metabolism. It is considered to have possible value in preventive medicine.

### **Conditions**

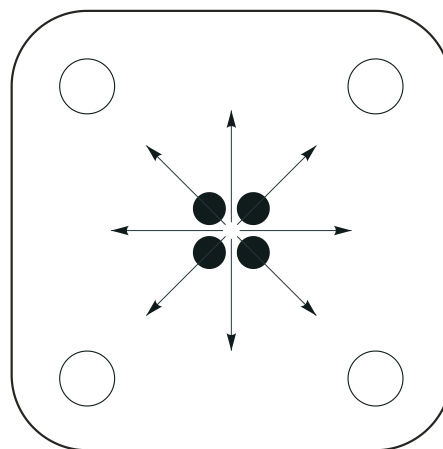
System: BAS MF480 chromatograph with multi-channel amperometric detection (epsilon™, BAS)  
Electrode: Four glassy carbon working electrodes  
Potential: +800, 700, 600, 500 mV vs. Ag/AgCl  
Column: C18, 3µm, 100 x 2.0mm (BAS, MF-8957)  
Mobile phase: 20 mM sodium acetate, 0.5 mM EDTA, pH 4.5, 18% acetonitrile (v/v)  
Flow rate: 0.4 ml/min  
Automated SPE system: Zymark RapidTrace workstation  
SPE cartridge: 100 mg C18 (1 ml) (Bond-Elut, Varian)

### **Sample Preparation**

A volume of 0.2ml of wine or 1.0ml of grape juice was transferred into 13 x 100mm borosilicate glass tubes and diluted to 2.0ml with water. For the grape seed capsules, to 0.1g of powder was added 2.0ml of methanol. The solution was sonicated for 15 minutes in the dark then centrifuged for 1 minute. A 0.1ml aliquot of the clear solution was transferred into a 13 x 100mm borosilicate glass tube and diluted to 2.0ml with water. Sample tubes from above were placed into the Zymark RapidTrace workstation, which was programmed to process the samples by solid phase extraction. Aliquots (20µl) of the extracts were injected into the LC system by autosampler.

### **Notes**

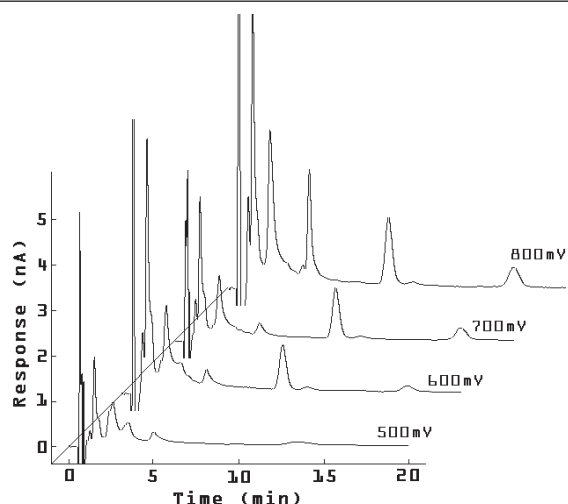
Four electrode detection was performed at applied potentials of +500, 600, 700 and 800 mV. A depiction of the working electrode configuration used in the four-channel electrochemical detector is shown in **F1**.



**Figure 1.** A depiction of the working electrode configuration for four-channel radial flow electrochemical detection (working electrode diameter = 2 mm).

Coincident chromatograms obtained from the individual channels are shown in **F2**. Monitoring four potentials gives a better voltammetric characterization of trans- and cis-resveratrol in different natural sources. Peak purity can be assured by comparing peak ratios at different energies for both standards and samples (1-3).

The calibration curve of resveratrol standard was linear over the range of 5 1000 ng/ml. The regression equation was  $Y = 258x - 18$  with correlation coefficient  $r^2 = 0.9999$ . **F3** shows chromatograms at +700 mV for red wine (A), grape juice (B) and grape seed capsules (C) after solid phase extraction. The recovery of known resveratrol concentrations added to the wine, grape juice and grape seed capsule varied between 96-102%.

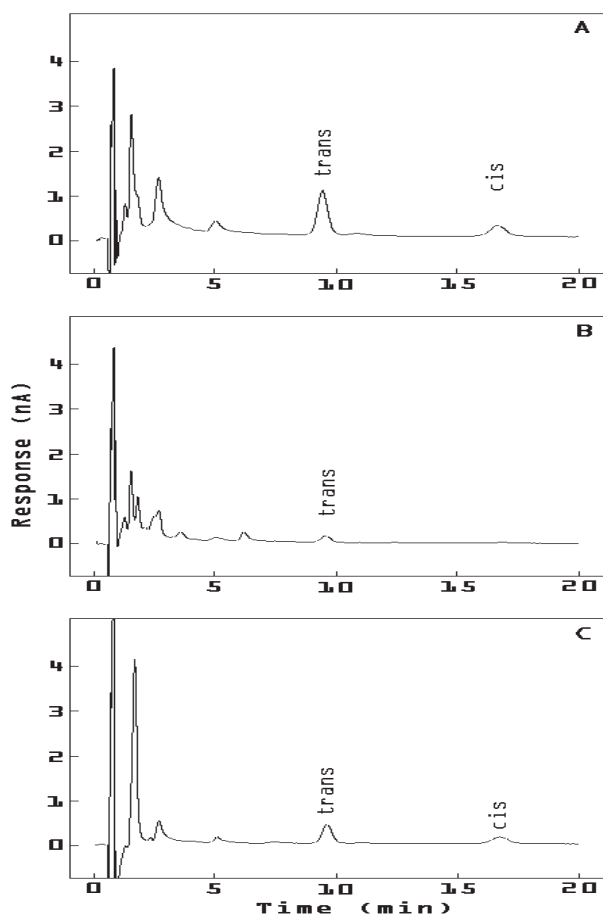


**Figure 2.** Chromatograms obtained from four-channel electrochemical detection.

The concentration of resveratrol in red and white wines, grape juice and grape seed capsules was determined by comparison with a standard solution. The results are comparable to those obtained by LCUV methods (4-5).

### References

1. D. A. Roston and P. T. Kissinger, *Anal. Chem.* 53 (1981) 1695-1699.
2. S. M. Lunte, *J. Chromatogr.* 2 (1987) 371-382.
3. S. M. Lunte, K. D. Blankenship, S. A. Read, *Analyst* 113 (1988) 99-102.
4. D. M. Goldberg, E. Ng, A. Karumanchiri, J. Yan, E. P. Diamandis, G. J. Soleas, *J. Chromatogr. A*, 708, 89-98 (1995).
5. D. M. Goldberg, E. Tsang, M. Levesque, G. J. Soleas, *J. Liq. Chrom. & Rel. Technol.*, 22, 1843-1855 (1999).



**Figure 3.** Chromatogram of extracts from red wine (A), grape juice (B) and grape seed capsules (C), trans-resveratrol (trans) and cis-resveratrol (cis).