Detection and quantification of geosmin and other fungal metabolites in grape juice by HS-SPME coupled with GC/MS.

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(-)-Geosmin (GEO) (Figure 1) is an aromatic volatile metabolite the presence of which in grapes is associated with an earthy smell of crucial importance in grape products (e.g. grape juice). Two fungi are involved in the accumulation of geosmin in grapes: *Botrytis cinerea* and *Penicillium expansum* (La Guerche et al., 2005). Other compounds of fungal origin such as fenchone, fenchol, anisoles and methyl isoborneol (MIB) can also transmit undesirable aromas (Boutou and Chatonnet, 2007)

According to literature (Boutou and Chatonnet, 2007; Baghery *et al.*, 2007; Salto *et al.*, 2008 and Prat *et al.*, 2008), Head Space Solid-Phase Microextraction (HS-SPME) coupled with Gas Chromatography/Mass Spectrometry (GC/MS) methodology was used to detect and quantify GEO. The fiber used was divinilbenzene/carboxen/polidimetilsiloxane (DVB/CAR/PDMS).

Grape juice spiked with standards of the analytes was analyzed to validate the method. Optimization of the method was performed by determining the efficiency of GEO extraction. The influence of the ionization of the analytes was tested by extracting at different pH (from ca. 3.5 to 7.0) and/or addition of NaCl (from none to saturation). Also, the optimal ethanol concentration in the sample was assessed (from none to 6% v/v). Time of extraction (from 20 to 65 min) and temperature of extraction (from 30 to 70 °C) were assayed as was the position of the Head Space fiber. The results underwent ANOVA tests to evaluate the significance of the differences in geosmin extraction. Graphics of extraction of GEO as a function of the factors tested were plotted to choose those conditions that maximized the detection of GEO and permitted the detection of the other analytes. Finally, internal standards were tested (4-Nonanol; Methyl-4-pentan-2-ol and 2,3,4-Trichloroanisole) to obtain calibration curves.

The calibration curves of GEO were linear with high correlation coefficients. Geosmin, MIB and most of the other metabolites were detected. Quantification of GEO was possible in the range of concentrations in naturally contaminated grape juice.



Figure 1. Chemical structure of Geosmin

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