NIZO food research B.V.
Kernhemseweg 2, 6718 ZB Ede, The Netherlands
P.O. Box 20, 6710 BA Ede, The Netherlands
T +31 (0)318 65 95 11 F +31 (0)318 65 04 00
E info@nizo.nl W www.nizo.com

Sample preparation and injection techniques in combination with GCMS

Wilma Wesselink and Amparo Gamero Lluna
E-mail: wilma.wesselink@nizo.nl

Introduction

A wide variety of sample preparation techniques are available to analyze volatile organic compounds (VOC) or semi-volatile organic compounds (SVOC). Based on the research question and available techniques, a choice has to be made on how to analyze the samples. In this work, several extraction techniques were evaluated for their suitability to extract aroma compounds typically found in wines at low concentrations (ppb level). In addition, the influence of cryo refocusing at the begin of the column was investigated in combination with HS-SPME.

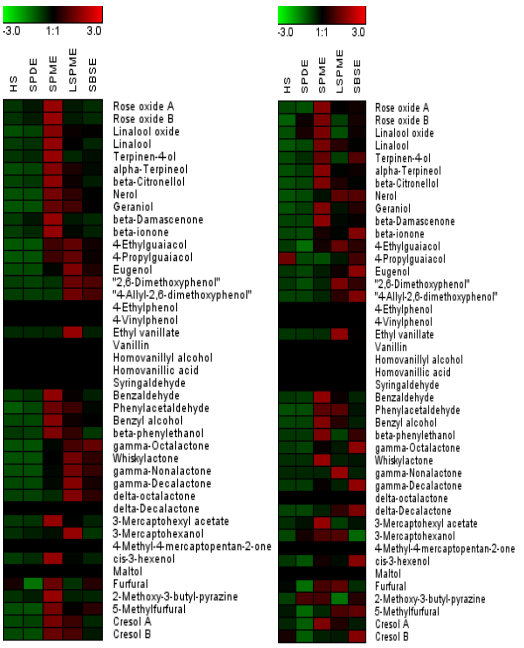
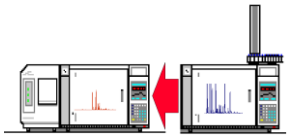


Fig. 1: Heatmap showing comparison of sensitivity of different sampling techniques for a 10 ppb reference matrix (left) and red wine matrix (right). SN-ratios increase from black (no difference) to green and are highest if colored in red.

Results and Discussion

Figure 1 shows a heatmap depicting the different signal to noise (SN) ratio intensities for each of the compounds of the reference mixture (left) and the red wine matrix (right). The results clearly point out that HS and HS-SPDE are the least sensitive techniques. As for SPME, L-SPME and SBSE, sensitivity depends on the volatile of interest. Hence, each technique presents advantages and disadvantages. Overall, L-SPME shows the highest sensitivity for detection of lactones.

Figure 2 shows the influence of cryo refocusing on the detection of selected volatile compounds. The more volatile the compound, the lower the required trapping temperature. For instance, methanethiol and acetaldehyde are not trapped at temperatures higher than -110°C. Contrarily, the less volatile compounds are not influenced by the refocusing temperature (e.g., 2-heptanone).



Samples

A reference mixture (10ppb) consisting of different volatile compounds (Figure 1; left hand side) was used, as well as a commercial red wine (Grand vin de Bordeaux, Tulipe de la Garde). The sample volume was 3 ml; each sample was analysed in triplicate.

Volatile extraction methods

HS (headspace)
The samples were incubated for 15 min at 60°C under agitation (500 rpm). 500 µl of the headspace was then injected into the GC column.
HS-SPDE (headspace solid phase dynamic extraction)
After incubation of samples for 15 min at 60°C under agitation, the volatiles were concentrated onto the phase of the needle (PDMS/AC) in a repetitive manner (5x) introducing the sample through the needle.
HS-SPME (headspace solid phase micro extraction)
A 50/30 µm DVB/CAR/PDMS fiber was exposed to the headspace of the sample for 15 min at 60 °C (agitation).
L-SPME (liquid solid phase micro extraction)
A 50/30 µm DVB/CAR/PDMS fiber was exposed to the liquid of the sample for 15 min at 60 °C (agitation).
SBSE (stir bar sorptive extraction)
Sample extraction was performed by placing a polydimethylsiloxane stir bar (10 mm, 1,0mm film thickness) into the liquid and let it stir for 16 h at 700 rpm at room temperature.

GC/MS

The GC analysis was carried out using a GC equipped with a 20 cm pre-column (CP-sil 0.53 mm x 1µm). All samples were thermally desorbed (splitless) into a hot injector (250°C). After cryo refocussing on the pre-column at -110°C (or other temperatures Fig 2), the trap was rapidly heated (50°C/s) to 200°C. The volatiles were transferred (splitless) to a VF-wax column (30m x 0.25 mm I.D., 0.5 µm film thickness). The GC column was programmed as follows: from 40°C (hold time 2 min.) to 250°C (hold time 5 min.) at 10°C/min. The carrier gas was helium 5.0, with a constant flow of 1.5 ml/min. The mass spectrometer (EI) was operated in full scan mode (m/z 35-250).
The signal to noise ratio (SN) was recorded for each compound in the different samples to compare sensitivity of the different techniques mentioned before.

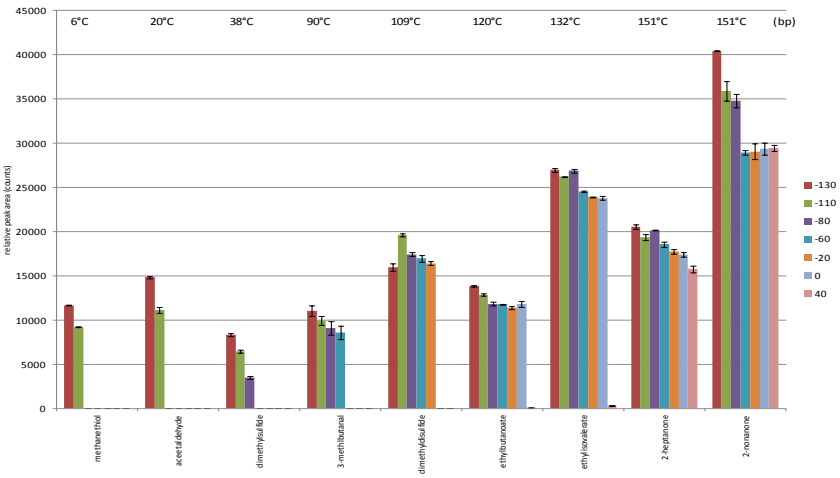


Fig. 2: : Influence of cryo refocusing in combination with HS-SPME.

Conclusions

- **HS-SPME, L-SPME and SBSE are more sensitive sampling techniques, compared to HS and HS-SPDE.**
- **SPME (liquid or headspace) covered the broadest range of volatile compounds.**
- **Cryo refocusing is crucial for the detection of highly volatile compounds.**