THE USE OF SOLID-PHASE MICROEXTRACTION (SPME) FOR THE ANALYSIS OF ALDEHYDE COMPOUNDS IN DRY-CURED HAM

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Background

In the last years, a considerable amount of research has been focused on dry-cured ham flavor not only by studying the mechanisms involved in its generation (Toldrá and Flores, 1998) but also analysing the volatile components using mainly headspace techniques. So, the study of the volatile compounds on the headspace has been mainly done at temperatures between 30 to 40°C in French (Berdague et al., 1991, Buscailhon et al., 1993), Parma (Barbieri et al., 1992, Bolzoni et al., 1996), Iberian (Lopez, et al., 1992, Sabio et al., 1998, Ruiz et al., 1999) and Serrano dry-cured hams (Flores et al., 1997) in order to mimic the release of volatile compounds on the mouth during mastication. The consumption of these traditional products is done without cooking and at room temperature. The temperature used has been higher (around 60°C) when other volatile extraction techniques such as vacuum distillation (Dirinck et al., 1997) and super critical carbon dioxide (Timon et al., 1998) have been used. In a previous study, the volatile compounds of dry-cured ham were extracted using SPME at different times and two temperatures (40° and 60°C) to assess its influence on extraction (Ruiz et al., 1998).

During the analysis of headspace by SPME, an exhaustive extraction does not occur instead an equilibrium is reached between matrix and the stationary phase coating the fiber (Zhang and Pawliszyn, 1993). There are many factors that affect SPME fiber performance such as the choice of the stationary phase and extraction conditions (Prosen and Zupancic-Kralj, 1999). It is necessary to take into account that competition effects between volatile compounds may skew results when one compound with a high affinity to the fiber (high constant Kfiber-air) is present at high concentrations (Roberts, et al., 2000). In meat samples, the use of SPME can be used for the analysis of lipid oxidation products such us the study of aldehydes in dry-cured ham due to their impact in meat flavor. The study has to be at mild temperatures to mimic the release of the volatile compounds during consumption in order to avoid artifact generation due to high temperature effect.

Objectives

Our objective is to develop and optimize a solid-phase microextraction sampling procedure for qualitative and quantitative determination of aldehyde compounds present in the headspace of dry-cured ham using different fiber coatings.

Methods

Samples. Dry-cured hams were selected from a local industry and processed according to a traditional methodology and ripened up to 12 months. The ham sample was divided in 50 g portions, vacuum packaged and stored frozen at -20° until analysis.

SPME fibers. The extraction of headspace volatile compounds was done using a SPME device (Supelco, Bellefonte, PA) using different fibers: 75µm Carboxen-PDMS and 50/30µm DVB/Carboxen/PDMS. Before the analysis, the fibers were preconditioned in the injection port of the GC as indicated by the manufacturer.

Procedure. For each experiment 3 g of dry-cured ham were cut in 2 mm cubes and weighed into a 10 ml headspace vial and sealed with a PTFE faced silicone septum (Supelco, Bellefonte, PA, USA). The vial was left at 30°C in a thermo block (J.P.Selecta, Barcelona, Spain) during 1 h 30 min to equilibrate its headspace. A SPME fiber was exposed to the headspace while maintaining the sample at 30°C during different times (30 min, 90 min, 3 h, 5 h and 17 h). The aldehyde compounds adsorbed by the fibers (75µm Carboxen-PDMS and 50/30µm DVB/Carboxen/PDMS) were quantified and identified by gas chromatography analysis using FID and MS detectors, respectively.

Identification and quantification of aldehyde compounds. The volatile compounds adsorbed by the fiber were desorbed in the injection port of the GC for 6 min at 220°C with the purge valve off (splitless mode). The compounds were separated in a DB-624 capillary column (J&W Scientific, 30 m, 0.25 mm i.d.; film thickness 1.4 µm) installed on a gas chromatograph GC 5890- HP series II equipped with a HP 5972 mass selective detector (Hewlett Packard, Palo Alto, CA) for their identification. Helium was used as a carrier gas with a linear velocity of 27.3 cm/sec. After desorption, the fiber was baked in a separate injection port at 220°C for an additional 10 min to eliminate the possibility of analyte carryover between samples. The GC oven temperature program begins when the fibers is inserted and holds at 38°C for 13 min, ramp to 150°C at 4°C per min, then to 210°C at 10°C/min and holds for 5 min; total run time 52 min. The GC-MS interface was maintained at 240°C. Mass spectra were obtained by electron impact at 70 ev. Mass spectral data were acquired across the range 25-400 amu.

The quantification of volatile compounds was done on a gas chromatograph GC 8000 CE Instruments (Rodano, Milan, Italy) equipped with a flame ionization detector (FID) equipped with a DB-624 capillary column (J&W Scientific, 60 m, 0.32 mm i.d.; film thickness 1.8 µm). The same temperature program was used for GC-FID and GC-MS analyses. Injector and detector temperatures were set at 220° and 240°C, respectively. The content of each of the aldehyde compounds in the ham sample was calculated from the FID areas and expressed as area units. Each sample was analyzed by three replicates. Kovats indices (KI) of the aldehyde compounds were calculated according to the method of Kovats (1965) and used for their identification by comparison with authentic aldehyde standards.

Results and Discussion

The extraction of dry-cured ham aldehyde compounds by the two fiber coatings was followed at 30°C during 5 h. The two fiber coatings exhibited increasing volatile extraction over 5 h with CAR/PDMS coating extracting between 2-3 times more amount of aldehyde compounds than DVB/CAR/PDMS coating (Figure 1). Both the fiber coating and the extraction time strongly affect total chromatographic area (see figure 1). Although the time of exposure of the fiber coating was more marked in the CAR/PDMS coating than in DVB/CAR/PDMS coating as seen by the increment in peak area of many aldehyde compounds with the time of exposure. The graph of total area counts suggests that absorption reached equilibrium after 5 h incubation although, some of the volatile compounds may still be increasing, such as hexanal in CAR/PDMS or nonanal in DVB/CAR/PDMS (Figure 2).

Both fibers were capable to detect all the series of aldehydes (from pentanal to nonanal) although the decanal was in very low quantities and only the DVB/CAR/PDMS fiber coating was able to extract it while CAR/PDMS only extracted decanal after 5 h of exposure (Figure 2). The DVB/CAR/PDMS fiber coating extracted increasing concentrations of octanal and nonanal with the time of exposure.

The CAR/PDMS fiber coating extracted increasing concentrations of pentanal, hexanal and heptanal while showed less affinity for octanal, nonanal and decanal.
In the case of hexanal, it has been study as an index of lipid oxidation in pre-cooked pork meat (Nielsen et al., 1997) using SPME, and it has a flavor of green cut grass and it was related to short processing stages in dry-cured ham (Flores et al., 1997). The study of hexanal by SPME in the headspace of dry-cured ham may be also useful to follow the lipid oxidation phenomenon although it is important to establish the appropriate conditions of analysis.
Therefore, the optimal time for extraction should be the time to reach equilibrium. In this case, the majority of aldehyde compounds extracted by DVB/CAR/PDMS coating reach equilibrium after 3 h of exposure while the compounds extracted by CAR/PDMS coating needed at least 5 h to reach the equilibrium.

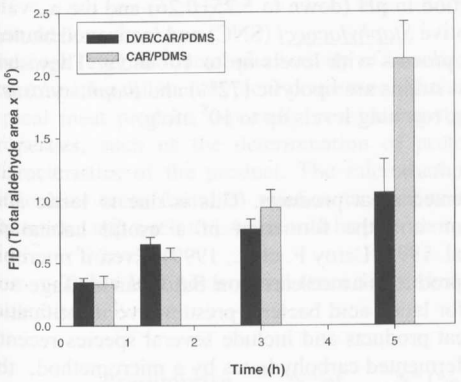

Figure 1. Absorption time profile of total aldehyde compounds using different fiber coatings of SPME.

Conclusion

SPME is an appropriate tool for qualitative and quantitative analysis of aldehyde compounds in the headspace of dry-cured ham. However, many factors affect coating efficiency such as the polarity, volatility and size of the analytes although competition effects may also be present in complex mixtures of volatile compounds such as dry-cured ham. Moreover, the extraction yields of dry-cured ham aldehyde compounds varied according to the fiber coating used. Therefore, extraction conditions should be carefully selected depending on the objective of the study.

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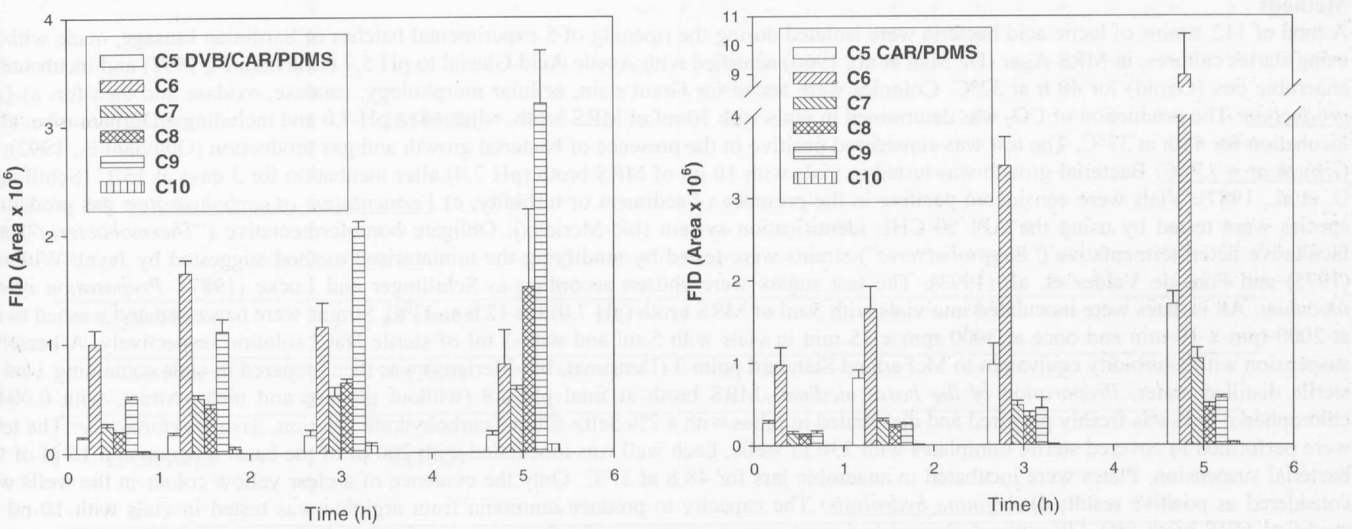

Figure 2. Absortion time profile of aldehyde compounds using different fiber coatings of SPME.

Pertinent literature

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