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## QuEChERS extraction and simple clean-up procedure for the GC-MS/MS quantification of polycyclic aromatic hydrocarbons (PAHs) in cheese

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Summary: Polycyclic Aromatic Hydrocarbons (PAHs) are chemical compounds associated with risks to human health, especially carcinogenesis. PAHs in food originate from environmental deposits or arise from food processing. In this study a fast and effective method to quantify PAHs in difficult fatty food matrices, such as cheese, was proposed.

Keywords: PAHs, cheese, GC-MS/MS

#### Introduction

Polycyclic Aromatic Hydrocarbons (PAHs) are chemicals composed of hydrogen and carbon atoms arranged in two or more fused aromatic rings. More than a hundred different PAH compounds exist and almost always in mixtures. PAHs represents environmental carcinogenic compounds, and can arise as a result of human activity as well as during food processing. The major route of human exposure to PAHs for non-smokers is food. Because of their lipophilic nature, PAHs can accumulate easily in fatty food matrices such as milk, meat, edible oil, fish and cheese [1].

The aim of the presented study was to develop an effective and easy sample preparation procedure for the determination of PAHs in commercial cheese samples.

#### Experimental

For this work, 60 samples of non-smoked hard cheese were collected from the market. The optimised protocol for PAHs quantification was composed of three steps: 1) fast QuEChERS extraction using cyclohexane as solvent, 2) easily and fast mechanically clean-up by Silica Gel (70-200 mesh ASTM) and 3) concentration under gentle stream of nitrogen.

PAHs analysis was carried out on an Agilent Intuvo 9000 GC system coupled with an Agilent 7000 Series Triple Quadrupole MS using a HP-5MS Ultra Inert (30 m × 0.32 mm id × 0.25  $\mu$ m film thickness) capillary column. Chromatograms were acquired in selected-ion monitoring (SIM) mode and target compounds were identified according to their ions and retention times.

For method development were considered 18 different PAHs: naphthalene (NaP), 2-methylnaphthalene (MeNaP2), 1-methylnaphthalene (MeNaP1), acenaphthene (Ace), acenaphthylene (Acp), fluorene (Flu), phenanthrene (Phen), anthracene (Ant), fluoranthene (Fla), pyrene (Pyr), benzo[a]anthracene (B[a]a), chrysene (Chr), benzo[b]fluoranthene (B[b]f), benzo[k]fluoranthene (B[k]f), benzo[a]pyrene (B[a]p), indeno[1,2,3-c,d]pyrene (I[cd]p), dibenzo[a,h]-anthracene (D[ah]a), benzo[g,h,i]perylene (B[ghi]P).

#### Results

The developed method was validated in terms of linearity, repeatability, reproducibility, recovery, limit of detection (LOD), and limit of quantification (LOQ). Linearity was evaluated between 0 and 500  $\mu$ g/kg; the values of the R<sup>2</sup> were higher than 0.99 for all the considered compounds.

The repeatability and reproducibility were calculated from five spiking samples analysed on the same day and from three different days, respectively. The repeatability was lower than 10% for all analytes, whereas the reproducibility was in most cases better than 10%, going from 4 to 15%.

The recovery was calculated at two different concentration levels: 20 and 100  $\mu$ g/kg. All results were found within acceptable limits and ranged from 75% to 110%. The LOD and LOQ were estimated at 0.3 and 0.9  $\mu$ g/kg, respectively.

Once validated, the method was applied to analyze the entire set of collected samples. In only three cheese samples were identified the PAH compounds. Specifically, were quantified the three PAHs with the lower molecular weight: NaP, MeNaP2 and MeNaP1. However, in these samples the contamination degree was very low with a highest total concentration of 30 µg/kg.

#### Conclusions

The experiment conducted in this study demonstrated that the proposed method allows an effective clean-up of the samples and at the same time achieve acceptable recoveries, repeatability and reproducibility for PAHs quantification in cheese. For this reason, the method could be successfully applied at different food matrices with high fat content.

#### References

 L.Duedahl-Olesen. 13 - Polycyclic aromatic hydrocarbons (PAHs) in foods -Persistent Organic Pollutants and Toxic Metals in Foods. Woodhead Publishing Series in Food Science, Technology and Nutrition 2013, pages 308-33.