Group : Organic Chemistry (WU), Bioanalytical chemistry (VU) and Toxicology (WFSR)
Project : Effect-driven identification of genotoxic polycyclic aromatic hydrocarbons (PAHs) in mineral oils
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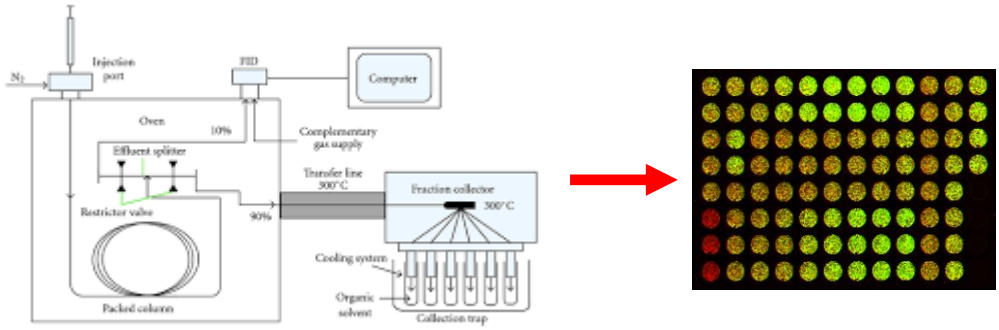
Introduction

Mineral oils are complex mixtures containing a large number of hydrocarbons which can be divided into two groups, MOSHs (mineral oil saturated hydrocarbons) and MOAHs (mineral oil aromatic hydrocarbons)[1]. These hydrocarbons can enter the food chain through contact with packaging materials, in particular material manufactured from recycled paper and cardboard, or through its use as lubricants in grinding processes. Both types of hydrocarbons have the attention of the European Commission (EC), European Food Safety Authority (EFSA)[2], Dutch government and organizations such as Foodwatch due to the supposed negative health effects. For example, MOSHs can accumulate and potentially cause damage in organs such as liver and lymph nodes and MOAHs may be genotoxic and carcinogenic. MOAHs basically are polycyclic aromatic hydrocarbons (PAHs) that can contain up to 50 carbon atoms. The 8 PAHs that the European Commission has designated as so-called indicator PAHs on the basis of their genotoxic/carcinogenic properties contain relatively few C atoms (C18-C22). There is an urgent need for getting more insight into the possible genotoxic effects of >C22 MOAHs (and possible differences in potencies) and for analytical methods that allow to measure and identify these hydrocarbons in a sufficiently sensitive and reliable manner.



Goals

The aim of this project is to set up a preparative GC method for collecting MOAH fractions from mineral oils and then testing the genotoxicity of these fractions in human liver cell lines with the γH2AX In Cell Western (ICW) assay (see Figure). By combining analytical chemical techniques with bioassays, we aim to identify those MOAHs that contribute most to the genotoxicity of mineral oils. The establishment of a preparative GC method for the separation of MOAHs will take place through a collaboration with Wageningen Food Safety Research (WFSR) and the Department of Biomolecular Analysis of the Vrije Universiteit (VU) in Amsterdam. WFSR is a research institute that is part of Wageningen University and Research and has many collaborative projects with ORC. At VU Amsterdam, the preparative separation of aromatic hydrocarbons will be set up step-by-step, initially with a mix of standards and later with more complex mixtures. The genotoxicity of the fractions will be tested at WFSR. Here, a number of human liver cell lines (HepaRG/HepG2) are exposed to the purified fractions and the γH2AX assay is used to determine whether DNA damage has occurred. The γH2AX ICW assay is an assay in which specific antibodies are used to determine whether double stranded DNA breaks occur [3]. The work will therefore be performed both in Wageningen and Amsterdam.

Techniques to be used

Preparative GC; Cell culture (HepaRG and HepG2 cell lines); cell viability (WST-1) and Genotoxicity (γH2AX) analysis.

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