A Complex Mixture

Automated MOSH/MOAH determination with the aid of coupled LC-GC

Once only of academic interest, the combination of liquid chromatographic separation with subsequent separation in the gas-phase is proving to be suitable for routine use. In routine laboratory work, the method continues to present a major challenge, above all with regard to precision, repeatability and robustness. A sophisticated coupling of methods can offer decisive advantages, through a greater degree of automation. At the Institut Kirchhoff Berlin such a system is being used, and has been specifically developed for the determination of mineral oil contaminants in foods and packaging materials.

q&more spoke with the food chemist and laboratory manager, Erik Becker, about the application and efficiency of this technology in routine laboratory work.

q&more: What do you use coupled LC-GC for in your laboratory?

In the Institut Kirchhoff Berlin GmbH, this coupled technique is used for the analysis of mineral oil hydrocarbons in foods and cardboard packaging. The investigation is performed according to a special method, based on the original method by Dr. Grob of the Kantonales Labor Zurich (online coupled LC-GC-FID). The technological implementation of this coupled technique was made possible by the company Axel Semrau GmbH & Co. KG. The measurement of the mineral oil content in foodstuffs is challenging because it involves a complex mixture that must be quantified as the sum of all the components. An individual component analysis is, therefore, not possible, due to the immense number of compounds. Additionally, both mineral oil fractions

MOSH (mineral oil saturated hydrocarbons) and MOAH (mineral oil aromatic hydrocarbons) must be differentiated from the food's own hydrocarbon compounds and often also separated out from them. The underlying principle is the separation of these two fractions and the subsequent GC-FID detection.

What is coupled LC-GC?

With this two-component system, the HPLC serves as the automated online sample preparation stage, while the GC actually determines the appropriate mineral oil fractions. The core and decisive factor for successfully coupling these two instrument systems is the suitable transfer technology. For the LC-GC transfer, the so-called Retention-Gap technique is used by partially concurrent eluent evaporation, which is crucial for the determination of volatile hydrocarbons. Both fractions are then sent straight to the GC and analysed.

How is LC suitable for sample preparation for GC?

One combines the high separation efficiency of LC with the high separation performance and chromatography of GC. Compared with manual, multi-step reprocessing, automated sample preparation minimises analyte loss and possible carry-over. Also, the recovery is as a rule higher and more reproducible than with the offline-variants. The normal phase HPLC retains the interfering lipids and matrix components (separation of olefins and wax esters) and separates the MOSH from the MOAH fractions.

The online sample purification method is not sufficient for all foodstuffs. Analytical auxiliary techniques for complex matrices (such as tea, for example) are used, including pre-separation using active aluminium oxide for the separation of biogenic hydrocarbons (n-alkanes of predominant odd-numbered chain length, above all C_{23} to C_{35}) and the epoxidiation for the elimination of interfering olefins.

Automation of the LC-GC with a CTC-sampler

Does this affect the operating life of the entire analytical system?

Not in comparison with the established alternative processes (manual methods, offline LC-GC), because the corresponding sample preparation steps here are also integral parts of the method. The advantage of this coupled technology (in addition to what has already been mentioned) is in the automation and the resulting high sample throughput.

The method does not seem easily realisable. How is it in routine work?

The sample preparation (pre-fractioning of the mineral oils into MOSH and MOAH) makes it easy to control this complex equipment and the complicated interfaces. Following an appropriate "training phase" and technical assistance by the manufacturer, the coupled techniques can be performed as routine. A good example is Dr. Grob's working group, which has been successfully working with this system since the 1980s and has published a series of application examples.

How long have you been working with the system and how high is the sample throughput in your laboratory?

Although we have been carrying out the determination of mineral oil residues in foods for two years, we began with manual separation. The changeover to the online coupling three-quarters of a year ago was therefore considerably easier than a direct introduction into this complex topic and technology. The coupling links the sub-steps of this procedure, which were already established.

To date, we have tested more than 1,600 samples for residues of the mineral oil (MOSH/MOAH). Of these, around 200 samples have been cardboard packaging and around 1400 samples from foodstuffs. In approx. 30 % of the food samples, residues of mineral oils could be detected. The mineral oil content in the packaged foods reached values of up to 60 mg/kg. In almost all the positive findings, the values of the MOSH fraction lay over the JECFA-ADI derived reference value of 0.6 mg/kg and in addition contained the aromatic compound fraction (MOAH fraction). The recycled cartons all contained mineral oils with aromatic compounds.

Is the system used to its fullest capacity or could you process more samples?

A run lasts 30 minutes; both the fractions of a sample are, therefore, measured within an hour. After deducting the reference standards, blanks, quality control samples and other possible maintenance work, about 100 samples could be run per week. With our present sample throughput there is still some free capacity. The obvious ease on the personnel binding in the area of sample preparation is crucial.

What is your assessment regarding the reproducibility and sensitivity of the method?

We have validated this procedure for five different matrices as examples (rice, chocolate, tea, vegetable oil and cardboard packaging). The internal laboratory repro-ducibility, dependent on the matrix, lies between 10 % and 20 %, the recovery of the added spike is between 85 % and 105 %. The estimated expanded uncertainty gathered from the validation data is in the range of 30 % to 50 %. Comparative studies with other laboratories show a good consistency as a rule.

The method is sensitive enough for testing for MOSH in the majority of foods to the JECFA-ADI derived reference value of 0.6 mg/kg. The limiting factor for the detection limit is the capacity of the lipid separation column and the substances naturally occurring in the foodstuffs, such as waxes or terpenes, which must not be included in the quantification.

Is using the system economically viable for you?

We were able to start with the measurement of routine samples relatively soon after the installation and qualification of the LC-GC system and therefore begin the payback of the instrument. It is always difficult to predict how the demand will develop for a specialised analytical service, but we assume, based on the brisance (number of positive findings and the variety of the affected foodstuff groups, incl. the toxicological potential of the mineral oil residues) that this topic will occupy us for a while yet. At present, the demand for this analysis is in any case large.

The LC-GC system MOSH/MOAH seems to be a complex piece of equipment. How difficult was it to train the staff?

Because both the LC and the GC are known and are long-established chromatography techniques for us, the amount of necessary training for the staff was reasonable. However, due to the complexity of the system in the beginning, it was only possible to train academic members of staff. The concomitant applicative and technological support of the service technicians is absolutely crucial, especially during the initial period.

How do you assess the support of the manufacturer?

The rapid reaction time during device failures, the applicative competence and the possibility of an online error diagnosis and remedy by the firm Axel Semrau were basic conditions for the rapid commissioning of the system and its establishment in the routine analysis.

Do you see other application areas for the coupled LC-GC in your laboratory in the future?

Because the coupled LC-GC system is not a new technique, there are a whole series of application examples and innovative approaches for possible R&D projects, which are particularly interesting for analyses where the sample preparation is very labour intensive. We are very excited.

Erik Becker is a certified food chemist (degree in Bonn, practical year in Münster). Since 2002, he has been working at the Institut Kirchhoff GmbH with emphasis on the following points: Management of chemical analysis tests (2002 – 2007), Technical manager of the testing laboratory (2007 – 2009), Laboratory manager / scientific management (2009 – 2011). He has been the testing laboratory manager since 2011. His areas of work are the organisation and management of the analytical sections, customer support (focussing on scientific questions), the coordination and supervision of analytical development and validation projects, as well as the development of new quality assurance strategies and laboratory projects. He is involved in diverse committees and certification boards, such as the GA FETT (Joint Committee for the Analysis of Fats, Oils Fatty products, Related Products and Raw Materials), the DIN-Committee "Vitamins and Carotinoids", the §64 LFGB Working Group "Vitamin analysis" as well as control-testing expert.

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