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Separation and Structural Elucidation of Xenobiotics in the Pico- to Nanogram Scale

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Abstract. Structural elucidation and quantification of complex mixtures of organic compounds is a real challenge when *i*) the substance amount approaches the pg to ng range, *ii*) a lot of closely related structures are present, and *iii*) when no special structure-attribute allows a specific detection. The main methodological interest of our group is the development of chromatographic techniques and selective detection mechanisms allowing an unequivocal identification, structure elucidation, and quantification of xenobiotics and biotoxins in the ultra-trace range.

Michael Oehme received his doctorate in 1977 at the ETH Zürich under the supervision of *W. Simon*. This was followed by a post-doc at the University of Oslo working with *W. Lund*. From 1979 until 1995, he was a scientist and later section leader at the Norwegian Institute for Atmospheric Research and Professor of organic analytic chemistry at the University of Tromsö. He was appointed Professor of organic analytical chemistry at the University of Basel in 1995. Additional details of his research interests are to be found at http://www.chemie.unibas.ch/Department/Broch/Broch-96/B96.html

We are particularly interested in on-line mass-spectrometric techniques since they are capable of elucidating complete or partial structures with 10^{-9} g or less of sample. Examples of methods are selective gas-phase reactions in the ion source or multiple mass spectrometry (MS^n) which generates sequences of daughter-ion spectra. They allow us to obtain structural information when soft ionization techniques, such as electrospray or atmospheric chemical ionzation (APCI), are used. So far, no general fragmentation rules or sequences typical for substructures have been established unlike the case for electron-ionization MS. Therefore, the understanding of such fragmentation processes and their relevance for structure elucidation is a major field of research for our group.

Besides method development to achieve the required selectivity, and selectivity for a given compound or compound class, the application to real-life problems is the final proof of the suitability of a method. Here, we are mostly interested in the detection of biotoxins, xenobiotics and undesired by-products in all kinds of matrices. A more detailed survey of our activities was given in *Chimia* last year [1]. Therefore, only a brief summary of these topics is given and a more detailed description of new research fields is presented.

Structural Elucidation and Quantification by LC-MS

Soft ionization techniques usually form only *quasi* molecular ions. Ion-trap mass spectrometry is able to generate sequentially daughter-ion spectra from the molecular ion and important fragments within 1 s. This approach gives much structurally relevant information. Recently, a fragmentation scheme for atmospherically relevant higher carbonyl compounds was established which enables complete structural elucidation of such substances as stable dinitrophenylhydrazones by direct HPLC-MS^n ($n = 3–4$) [2]. Furthermore, the first fully validated HPLC-APCI-MS method was developed for the identification and quantification of trichothecene toxins in cereals such as wheat. Both the class of trichothecenes, the number and character of functional groups as well as the basic skeleton can be identified. Detection limits are in the low ppb range. Another application of MS^n is the structural elucidation of bisphenol-A-diglycidyl ether oligomers and their reaction products with, for example, solvents and radical scavengers. They are found in canned food and originate from the epoxy polymer coating. The *Figure* shows the HPLC-MS chromatogram of a leachate from a corned-beef can and one of the identified structures.

Enantioselective High-Resolution Gas Chromatography (HRGC)

The determination of enantiomer ratios allows us to differentiate between the abiotic and biotic degradation of chiral organic xenobiotics in the environment and helps to trace transfer routes between environmental compartments. Chlorinated pesticides have been, or are, applied in

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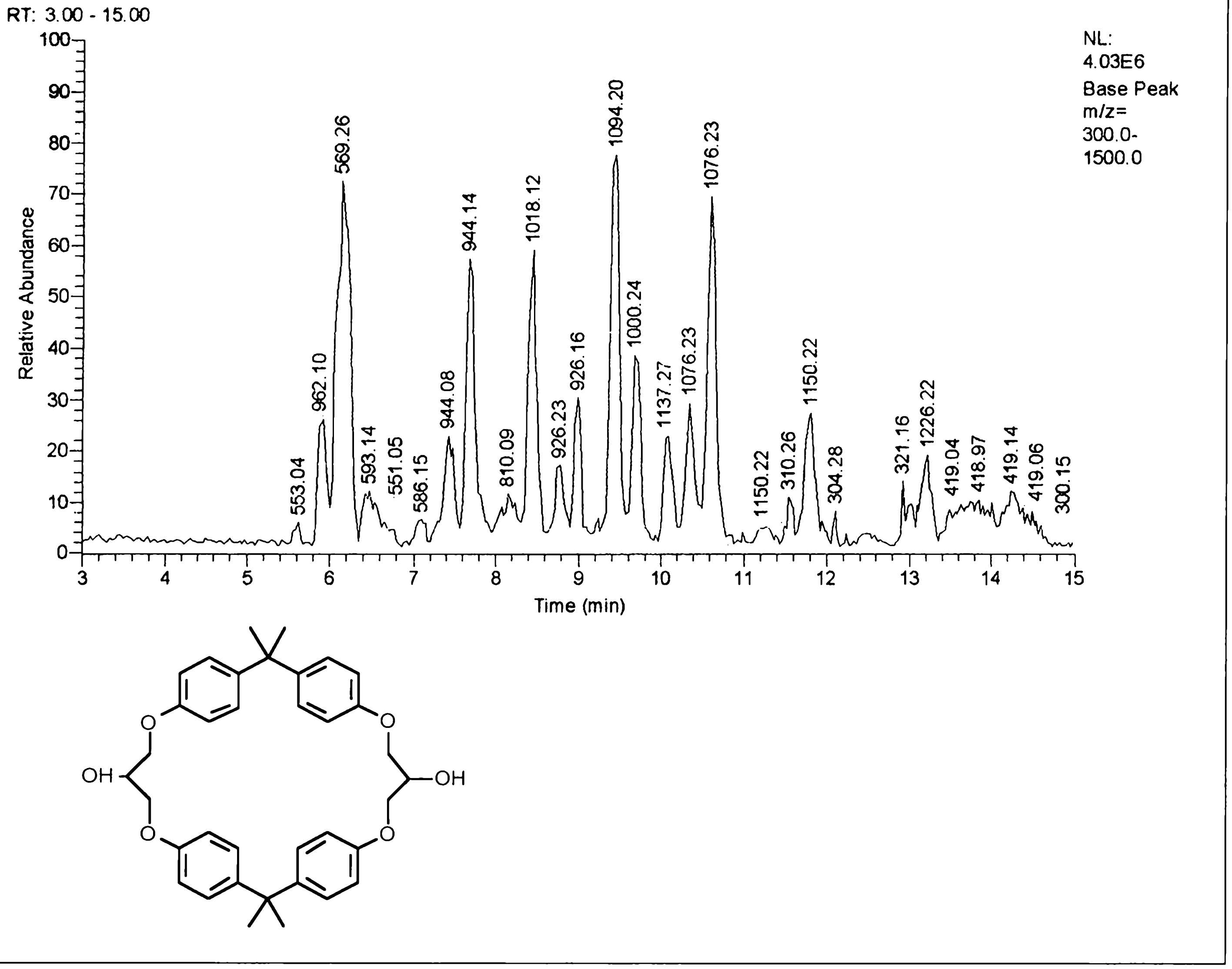

Figure. *HPLC-MS chromatogram of an extract of a corned-beef can coated with an epoxy layer.* A complex mixture of oligomers and their reaction products with solvents and additives is present. APCI in the positive-ion mode was employed. The mass of the [*M*+H] ion is given. The large signal at 6 min is the cyclodimer of bisphenol-A-diglycidyl ether, the structure of which is given.

their racemic forms, for example polychlorinated bornanes, *o,p'*-DDT, α-hexachlorocyclohexane and chlordanes. Their enantioselective determination requires chiral stationary phases. Modified cyclodextrins (CD) are well-suited for HRGC, but their properties have to be different from those of other important chiral substances (fragrances, flavors, *etc.*). Our group has developed such cyclodextrin based capillaries, and these have allowed us to detect the first gender-specific enantioselective metabolism of a pesticide in marine biota [1].

Even minor changes in the composition of the cyclodextrin have a great influence on the achieved enantioselectivity. Therefore, a method based on HPLC-MS was developed for the characterization of alkylated CD (mass ≥2500 u) [3] applied in HRGC. It showed that partially derivatized cyclodextrins containing some free OH-groups are more suited than completely alkylated structures [3].

Optimization of HRGC Separations Using Tandem Columns

Often, one cannot utilize the maximum separation power of HRGC due to clustering of signals in one part of the chromatogram while other ranges are nearly empty. Improvements can be made by time-consuming testing of other stationary phases or the use of two-dimensional techniques requiring complex instrumentation. Linking two columns together with different stationary-phase properties is the simplest and most cost-effective alternative. Since the length fraction of the single capillaries influences the overall separation properties, the optimization of a tandem system is normally very work-intensive. Therefore, a theory to model the overall achieved separation properties has been developed in our group. It is now applicable to temperature programming and allows us to predict the separation properties of any length combination from the characteristics of the single columns. Its first successful application was the optimization of the simultaneous isomer- and enantiomer-selective separation of a complex mixture of chlorinated bornanes (the pesticide which is most applied world-wide) [4].

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[1] M. Oehme, R. Baycan-Keller, M. Biniossek, H. Karlsson, S. Kölliker, L. Müller, D. Pekar, *Chimia* **1997**, *51*, 756.
[2] S. Kölliker, M. Oehme, *Anal. Chem.* **1998**, *70*, 1979.
[3] A. Jaus, M. Oehme, *Chromatographia* **1999**, in press.
[4] R. Baycan-Keller, M. Oehme, *J. Chromatogr. A* **1999**, in press.