FROM BAYREUTH TO CAIRNS: WHAT DID WE LEARN IN A QUARTER CENTURY? A PERSONAL PERSPECTIVE

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Introduction

At the time of the Dioxin conference in Bayreuth the personal experience in the dioxin field was not very old. After founding an independent private laboratory (ERGO Research) we started working in this field in 1984. We knew that the isotope dilution method was the method of choice. But the problem was at the early time of dioxin analysis: How to get the C^{13} standards. They were not available in Germany. Finally we received them from the United States not without of payment in advance.

Due to a chemical plant in Hamburg and resulting dumping activities for chemical residues at a big dump site in our city we started our "dioxin live" in analyzing highly contaminated residues. But, in the same year we started already to analyze biological samples from the area around the dump side. For many reasons, this type of samples attracts always our special attention.

Materials and methods

Samples

The main materials analyzed at the time of question are quite different and quite complex. We had to learn to analyze environmental samples like ambient, indoor and industrial effluent air samples, soil and sediment. But our main effort was quite early to analyze biological samples, especially human samples like human blood and human milk.

Instrumentation

Starting with only one HRGC/HRMS (VG 7035) the capacity was increase extending the instrumentation with an Autospec, followed by a MAT 95 and a Thermo DFS. Due to new developments we do work now with 13 instruments, mainly VG Autospec incl. 4 DFS. Table 1 shows the development sensitivity for measurement of TCDD by using various methods, especially HRGC/HRMS.

Figure 1: Increase of analytical sensitivity demonstrated for 2,3,7,8-TCDD

Year	Method	Detection limit (fg)
1967	GC/FID	500 000
1973	GC/MS (Balzers Quadrupol)	300 000
1976	HRGC/MS-SIM (LKB, Magnet)	200 000
1977	HRGC/MS-SIM (LKB, Magnet)	5000
1983	HRGC/HRMS (VG 70E)	150
1984	HRGC/MSD (HP 5970)	20
1986	HRGC/HRMS (VG 70S)	25
1989	HRGC/HRMS (VG AutoSpec)	10
2000	HRGC/HRMS (AutoSpec Ultima/ MAT 95)	2
2004	HRGC/HRMS (Thermo DFS)	1

QA/QC procedures are highly important in dioxin analysis. When starting to be active in this field in 1984, we did not know very much about "quality" of analytical data in dioxin analysis. I was lucky to be invited to learn a lot about QA/QC in at the CDC Atlanta in Don Patterson and Larry Needhams laboratory. Resulting from this

early introduction we were able to implement these QA/QC standards already as “normal” steps in our day to day analytical procedures.

Results and discussion

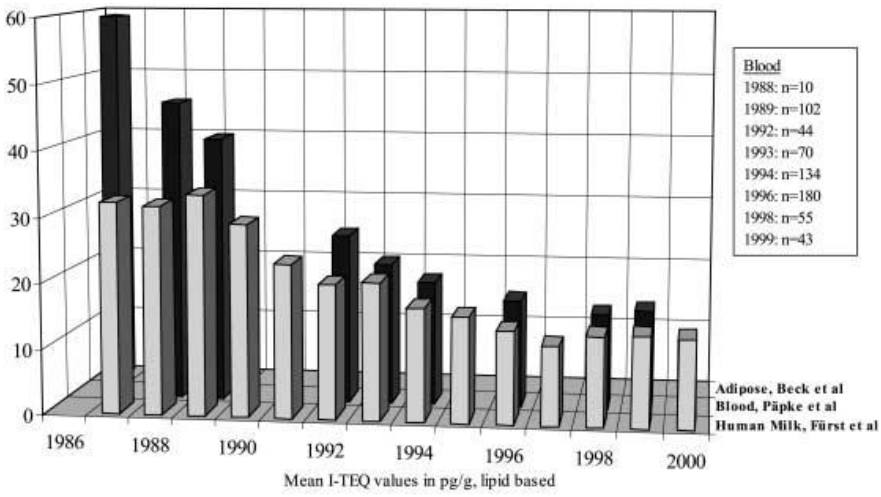
In the first years of dioxin measurements only PCDDs and PCDFs but no dioxin like PCBs were measured. We had to learn in the early 1980es from Christoffer Rappe (1) that analysis of human milk indicated a general background contamination for humans. These early results were confirmed by Peter Fürst (2) by analysis of milk samples from mothers living in Northrhein Westphalia.

After Don Patterson (3) important paper from 1988 on the correlation between serum and adipose tissue levels of 2,3,7,8-tetrachloro-*p*-dioxin we developed in our laboratory a method for measurements of all PCDDs/PCDFs in human whole blood.

First measurements for PCDD/PCDF-background contamination in adipose tissue were performed in Germany by Beck et al. (4). The mean value from 20 individual analyses was found at 59 pg I-TEQ/g lipid. First background data in blood for Germany originated from 10 individuals with no known exposure except food were found at a mean of 46 pg I-TEQ/g lipid by Paepke et al.(5).

A declining trend for PCDD/PCDFs in humans in the course of time was first observed for human milk in Germany by Fürst et al. (6). These declining data in milk and in blood (7) are given in Figure 1.

Figure 1: Time trend for PCDDs/PCDFs in humans from Germany, measured in adipose tissue, human milk and blood



In 1988 we began to analyze a number of blood samples originating from workers with potential occupational exposure. We could learn the difference of PCDD/PCDF - patterns in blood samples resulting from various occupational exposure situations like (8):

Metal reclamation
Pentachlrophenol production, Trichlorophenol production
Herbicide production
Chemical and municipal waste incineration

During investigation of occupationally exposed workers we analyzed a number of highly exposed individuals at several times. This offered the possibility to calculate the biological elimination for most congeners, published by Flesch- Janys et al., 1996 (9)

Long time collaboration with Arnold Schecter included comprehensive analysis from blood samples collected in a number of different countries word wide, inclusively Vietnam. (10). Vietnam became very important for me and our laboratory. On one side we started long lasting collaboration with some authorities in Vietnam especially

with Prof. Le Ke Son, Deputy Director General of Vietnam Environment Administration. On the other side we could support the Dioxin Laboratory in Hanoi (Head: Nguyen Hung Minh) by training the stuff in Hamburg.

Early collaboration with Jochen Müller (starting in 1990) and Caroline Gaus (starting in 1999) brought personal approaches and the entry to our Australian activities. It was of special interest to analyze samples originating from the Australian environment and to be present in a number of cases during collection of samples (11, 12)

We analyzed a number of highly contaminated blood samples in our laboratory. In some case values were found to be so high that is was nearly a problem to believe the results. But in case of repeated analyses you need to believe in your results. In 1998, during the Dioxin Conference in Stockholm, I asked some scientists for an ad hoc meeting to discuss results for 2,3,7,8-TCDD in a blood sample from a female worker from a textile institute in Vienna. The concentration found was the highest ever found for TCDD in a human sample at 144,000 pg/g lipid. (Geusau et al., 1999, (13)

Together with Jana Weiss and Ake Bergman we were able to organize a worldwide survey of PCDDs/PCDFs Polychlorinated and related contaminants in butter. Jana performed this investigation in the framework of her master thesis. In this study, 64 butter samples from 37 countries were analyzed to assess the global contamination of PCDDs/PCDFs, PCBs, and other contaminants. The highest PCDD/F and PCB concentrations were found in butter from Korea at an average of 1.97 pg TEQ /g lipid weight (14). Lowest concentrations were found in a sample from Russia/ Belogrod at 0.051 pg/g lipid.

One of the very special investigations in the last years was the analysis of Egyptian mummy tissues. Analyses of PCDD/PCDF patterns determined for tissue extracts of 10 Egyptian mummies supplied evidence that children in ancient Egypt may have been exposed to dioxins via food, where for young children nursing may have been the main source for the increased exposure to dioxins. (15)

During the last 10 to 15 years we were involved in most incidents of contaminated food or feed. Table 2 gives a selection of selection of a number of these special incidents.

Table 2: Accidental contamination of feed and foodstuffs (selection of some incidents)

Year	Incident	Reference
1998	Improper drying and mixing process of citrus pulp (Brasil)	
1999	Chicken feed contamination due to illegal disposal of PCB products (Belgium)	
1999	Caolinitic Clay	
2004	Elevated Dioxin and PCB levels in farmed salmon (Europe)	
2004	Potato by-products contaminated by use of dioxin-affected clay as seperation agent (NL)	
2005	Contamination of pig feed by use of polluted HCl in the production process (Belgium)	
2007	Contamination of guar gum from India with PCP and Dioxin	
2008	Mozzarella contamination in southern Italy	16
2011	Chicken and egg contamination in Germany (NRW)	17

Neugebauer et al. published on the results of the investigation in the Mozzarella case (16) and the actual incident of egg contamination (17). In both cases more than 1000 samples have been analyzed.

Acknowledgements and conclusion

I would like to express my special thanks to all my colleagues working in the different laboratories in Hamburg. Without their wonderful work my activities were not possible.
During my professional live I was very happy to have so many fruitful collaborations with scientists all over the world. Collaboration is one of the primary incitements for further developments. It is my strong recommendation to younger colleagues to search and find possibilities for collaboration with other groups.
Last but not least: Scientific publications. They are very important under several aspects:

- Providing of the "Scientific Community" with results from the own working group.
- Necessity to discuss the results of the project in question in detail.
- To go through the project from different sides, publications can stabilize a collaboration.
- Possibility to present the results at a conference and to defend/discuss potential week points of the paper or to come easily in contact to other working groups

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