FIRST SURVEILLANCE MONITORING RESULTS OF FEED AND FOOD SAMPLES FROM MARKETS IN KUWAIT FROM INTERNATIONAL ORIGIN FOR PCDD/PCDF/PCB-TEQ BY DR CALUX®

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Introduction

In the last few years animal feed and feed additives were the major sources of dioxin/dl-PCB contamination in the food chain and often caused severe crisis situation in the food chain[1-6]. The recurrent dioxin/PCB crisis situations raised a concern in Kuwait about their current dioxin/PCB levels from National and International feed/food products on their local market. In this study we present the first results of a surveillance monitoring program of several food (fish, milk, eggs, beef, lamb, chicken meat) and feed items (minerals, fish oil, animal fat oil, vegetable oil fat) of National and International origin from markets in Kuwait analysed by DR CALUX® technology for the total-PCDD/PCDF/dl-PCB-TEQ.

In 2009 the Kuwait Institute of Scientific Research has been trained at BDS in Amsterdam and established the DR CALUX in-house. In 2010 seventeen (17) fish samples, eighty (80) milk samples, seventeen (17) eggs samples, thirty-five (35) feeding stuff samples, fifty (50) beef meat samples, eighteen (18) lamb meat and ninety-nine (99) chicken meat samples have been analysed by the DR CALUX® technology by the team of KISR. Fifteen (15) samples have also been analysed by DR CALUX® technology in the laboratory of BDS as well as by chemical HRGC/HRMS analysis with a linear correlation of R^2 of 0.97. From these results two beef meat samples from Australia and New Zealand as well as one lamb shoulder from Kuwait were found to be positive. A re-sampling of these three samples in the local market could not again confirm positive results measured for PCDD/F/PCB-TEQ by DR CALUX®. The source of the contamination is still under investigation.

Methods and materials

Food and feed samples: All samples were analyzed at the Kuwait Institute of Scientific Research (KISR) according to most of the standard procedures of the DR CALUX® method from BDS[3,4].

DR CALUX® bioanalysis: The procedure for the BDS DR CALUX® bioassay is described in detail previously[3,4]. Briefly, H4IIE cells stably transfected with an AhR-controlled luciferase reporter gene construct, were cultured in α-MEM culture medium supplemented with 10% ($^{v}/_{v}$) FCS under standard conditions (37°C, 5% CO_2, 100% humidity). Cells were exposed in triplicate on 96-well microtiterplates containing the standard 2,3,7,8-TCDD calibration range, a DMSO blank Following a 24 hour incubation period, cells were lysed. A luciferine containing solution (Glow Mix) was added and the luminescence was measured using a luminometer (Berthold Centro XS3).

Results and Discussion

Kuwait is importing from many countries food and feed. In there here reported study the samples have been from twenty-three (23) different countries, such as Kuwait (n=117), UAE (n=34), Brazil (n=29) or Saudi Arabia (n=27).

Table 1 Number of food/feed samples from National and International origin from markets in Kuwait analysed for the Total-PCDD/PCDF/dl-PCB-TEQ by DR CALUX®.

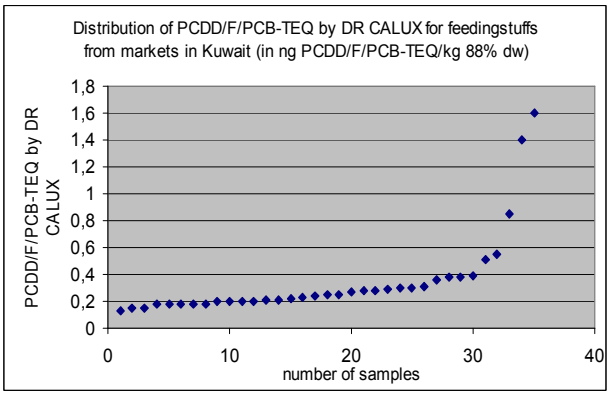
Country	N	Country	N
Australia	6	Ireland	2
Bahrain	2	Jordan	10
Belgium	4	Kuwait	117
Brazil	29	Malaysia	1
China	2	Netherlands	9
Dubai	1	New Zeeland	4
Egypt	4	Saudi Arabia	27
France	8	Singapore	1
Germany	4	Switzerland	1
India	2	Turkey	4
Iran	6	UAE	34
USA	7		

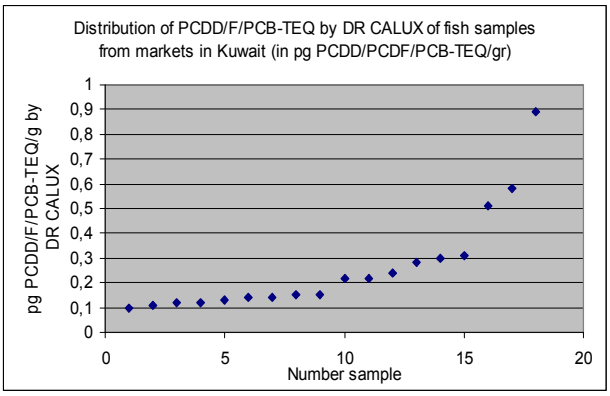
In Table 2 the number, mean value and range of concentration for PCDD/F/PCB-TEQ analysed by DR CALUX® is presented for each food and feed sample here reported: Seventeen (17) fish samples with a mean value of 2,2 pg PCDD/F/dl-PCB-TEQ/g, eighty (80) milk samples with a mean value of 2,3 PCDD/F/dl-PCB-TEQ/g, seventeen (17) eggs samples with a mean value of 2,2 PCDD/F/dl-PCB-TEQ/g, thirty-five (35) feeding stuff samples with a mean value of 0,35 PCDD/F/dl-PCB-TEQ/g, fifty (50) beef meat samples with a mean value of 1,9 PCDD/F/dl-PCB-TEQ/g, eighteen (18) lamb meat with a mean value of 1,9 PCDD/F/dl-PCB-TEQ/g and ninety-nine (99) chicken meat samples with a mean value of 1,7 PCDD/F/dl-PCB-TEQ/g have been analysed by the DR CALUX® technology by the team of KISR.

Table 2 Total-PCDD/F/dl-PCB-TEQ results from fish, milk, eggs, beef, lamb and chicken meat samples from markets in Kuwait tested by DR CALUX® technology (concentrations are in pg TEQ/g fat or fresh weight or ng TEQ/kg 88% dw).

Matrix tested	***PCDD/F/PCB-TEQ by DR CALUX***	***Matrix tested***	***PCDD/F/PCB-TEQ by DR CALUX***
1) Fish: N	18	*5) Beef Meat: N*	50
1) Fish: Mean	0.26	*5) Beef: Mean*	1.9
1) Fish: Range	0.1-0.89	*5) Beef: Range*	0.1-1.9
2) Milk: N	81	*6) Lamb Meat: N*	18
2) Milk: Mean	2.3	*6) Lamb: Mean*	1.9
2) Milk: Range	0.6-7.6	*6) Lamb: Range*	0.4-3.2
3) Eggs: N	17	*7) Chicken: N*	99
3) Eggs: Mean	2.2	*7) Chicken: Mean*	1.7
3) Eggs: Range	0.9-3.4	*7) Chicken: Range*	0.1-4.3
4) Feed: N	35		
4) Feed: Mean	0.35		
4) Feed: Range	0.1-1.6		

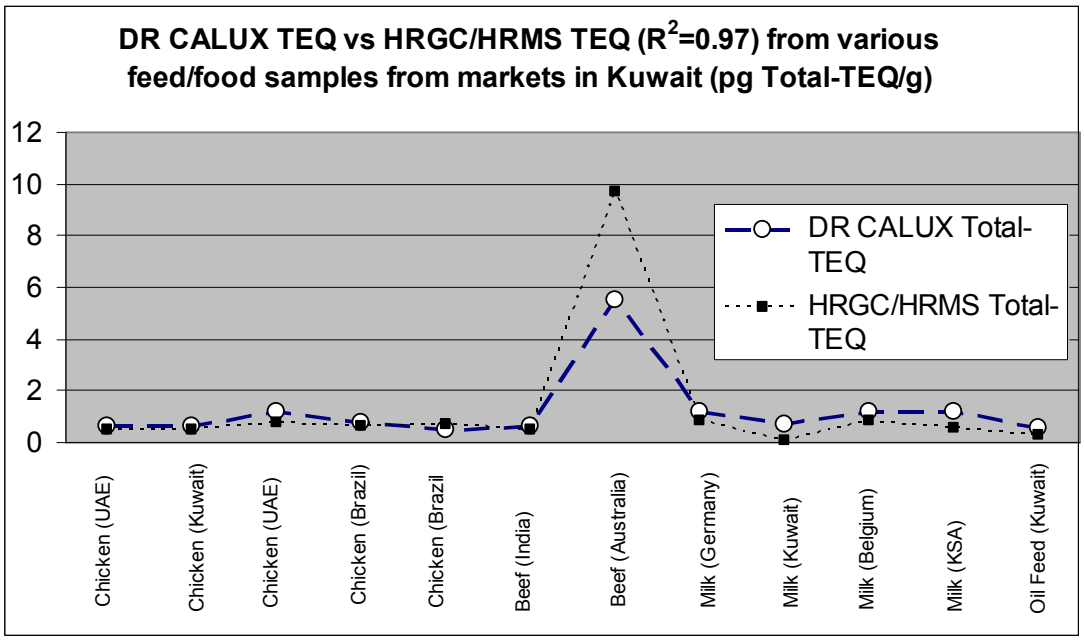
Graph 1 Distribution of the Total-PCDD/F/dl-PCB-TEQ results by DR CALUX® from some feed and fish samples from international origin from markets in Kuwait (concentrations are in pg TEQ/g fat or fresh weight or 88% dw).





Fifteen (15) samples have been also analysed by DR CALUX® technology in the laboratory of BDS as well as by chemical HRGC/HRMS analysis with a linear correlation of R^2 of 0.97. From these results two beef meat samples from Australia and New Zealand as well as one lamb shoulder from Kuwait were found to be positive for both DR CALUX and chemical HRGC/HRMS analysis. A re-sampling of these three samples in the local market could not again confirm positive results measured for PCDD/F/PCB-TEQ by DR CALUX®. The source of the contamination is still under investigation.

Graph 2 Comparison of the Total-PCDD/F/dl-PCB-TEQ results by DR CALUX® and HRGC/HRMS from several food/feed samples from international origin from markets in Kuwait (concentrations are in pg TEQ/g fat or fresh weight or 88% dw).



The above high peak of a beef sausage from Australia has been confirmed by both quantitative DR CALUX® (double measurement) and HRGC/HRMS (double measurement) as a non-compliant sample.

Conclusions

The demand for feed/food inspections is expect to rise not at least due to international export/import legislations with the ultimate goal to reduce these dioxins and dioxin-like PCBs significantly. More efforts will be promoted by the international food/feed-, public safety- and agriculture authorities to receive more data about the dioxins and dioxin-like PCBs in their countries. The results of this study shows that the DR CALUX® bioassay for screening of dioxin and dioxin-like PCBs in feed and food is an important tool to separate the bulk of unpolluted samples from the few percentage of the regulated limit exceeding samples. The here reported case of a national surveillance monitoring of feed/food samples of such a widespread international origin shows that screening of more samples also increases the chance to find the dioxin/PCB source and to prevent further damage to the feed and food chain.

References

1. Behnisch, P.A. (2005) in *Rapid Methods for biological and chemical contaminants in food and feed.* Wageningen Academic Publishers. pp. 303-318.
2. Behnisch, P.A., Besselink, H. and Brouwer, B. (2009). *Organohalogen Compounds*, 71: 966-969.
3. Besselink, H., Jonas, A., Pijnappels, M., Swinkels, A. and Brouwer, B. (2004). *Organohalogen Compounds*, 66:677-681.
4. Traag, W., Portier, L., Immerzeel, J., and Hoogenboom R. (2010). *Organohalogen Compounds*.
5. Schoeters, G., Goyvaerts, M.P., Ooms, D., and Van Cleuvenbergen, R. (2004). *Chemosphere*, 54, 1289-1297.
6. Behnisch, P.A., Besselink, H. and Brouwer, A. (2011). *DIOXIN 2011 Proceedings*.