

6th BioDetectors 2012

"DIOXINS/PCBs, ENDOCRINE DISRUPTERS (EDC), OBESOGENS AND EMERGING POLLUTANTS"

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Milk and eggs screening for dioxin/PCBs in Piedmont region by DR CALUX

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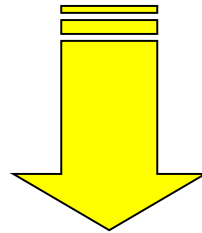
The Istituto Zooprofilattico Sperimentale of Piemonte, Liguria and Valle d'Aosta (IZSPLV) is a public veterinary Institute which conducts prevention, control and research activities in the main areas of animal health and welfare, food safety, and environmental protection



Why the IZSPLV decided in 2010 to acquire a screening test for the dioxin/PCB detection in food & feed?

BY REDUCTION OF

- TIME
- COSTS



BIOMONITORING OF RISK AREAS IN THE PIEDMONT REGION

LARGE NUMBER ANALYSES IN CASE OF ACCIDENTS



RESEARCH PROJECT by DR CALUX (1)

“To analyze milk coming from Piedmont farms, by screening method for dioxin and PCBs detection ”

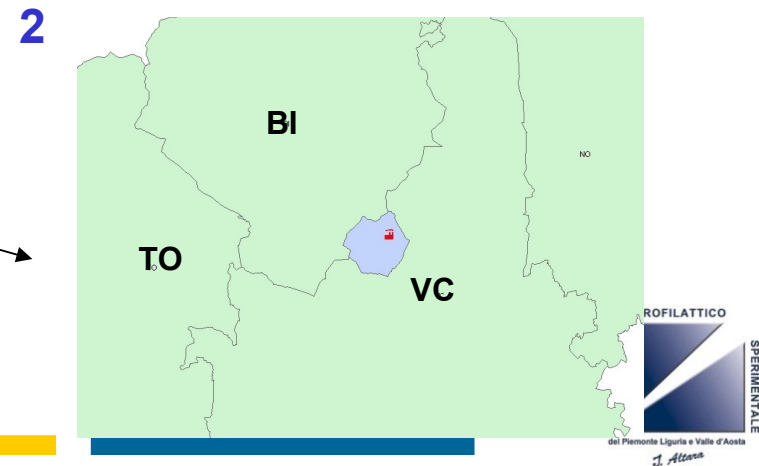
- ✓ Scientific project manager: dr. Maria Caramelli (SC Neuroscienze);
- ✓ co-founding by IZSPLVA + Fondazione CRT (Cassa di Risparmio di Torino);
- ✓ Aim of the **starting project**:
 - 1) to evaluate the BDS DR CALUX[®] method as a screening tool for monitoring dioxin/PCBs in food, particularly in cow milk coming from Piedmont region;
 - 2) to evaluate if this method is really useful for our labs;
 - 3) to evaluate the ICC value of the BDS DR CALUX[®] vs the HRGC/HRMS

BACKGROUND

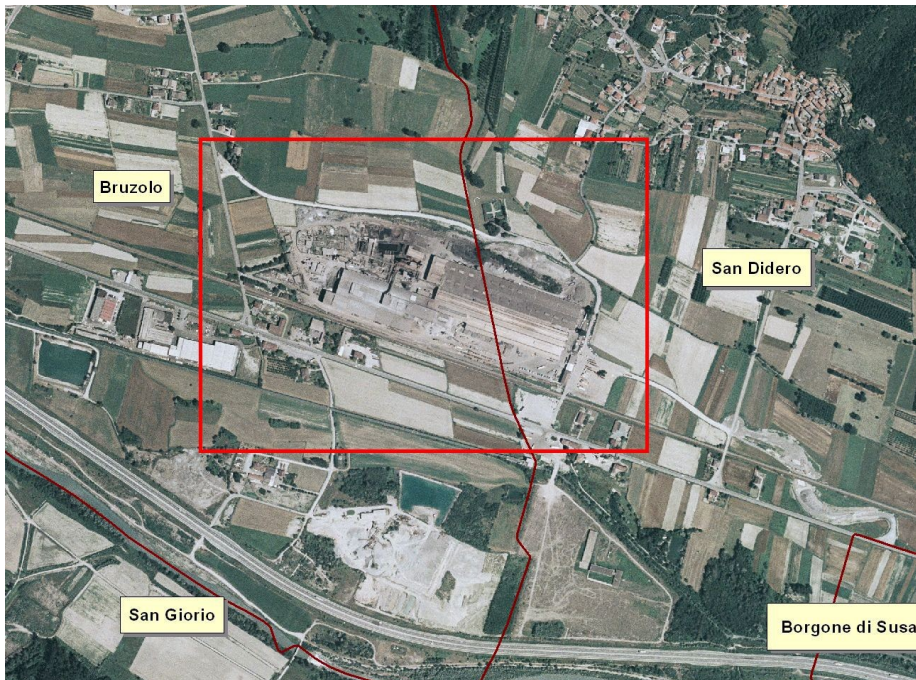
2 risk areas in Piedmont region:

1) The area of the lowest Val di Susa
- **a second fusion steelworks**

2) The area of Carisio between Vercelli and Biella (east of Piedmont region)
- **a second fusion aluminium factory**



BACKGROUND – Val di Susa



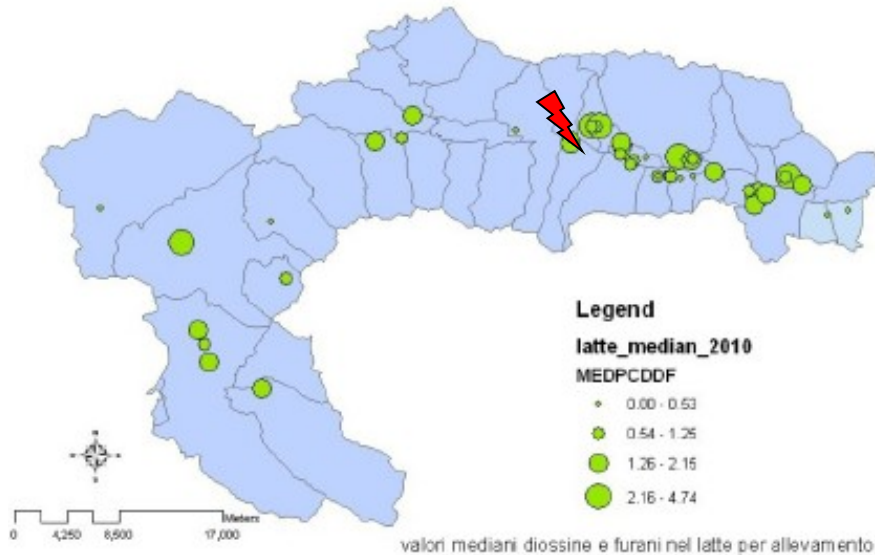
Emissioni diffuse provenienti dall'acciaieria.



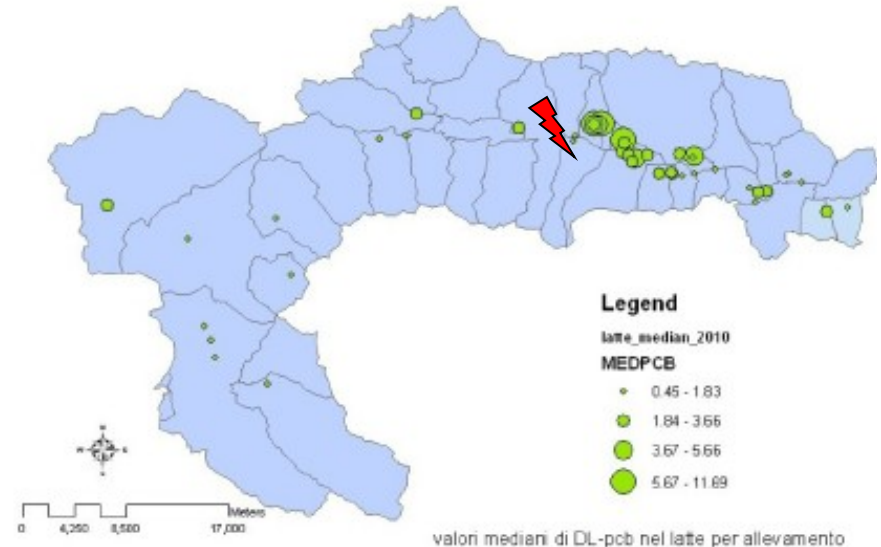
- population concerns due to uncontrolled emissions
 - first analyses in air and soil in 2003-2004
- 2004 – 2011 biomonitoring of raw cow milk, eggs and meat

GEOGRAPHICAL DISTRIBUTION (2004-2010)

PCDD/F



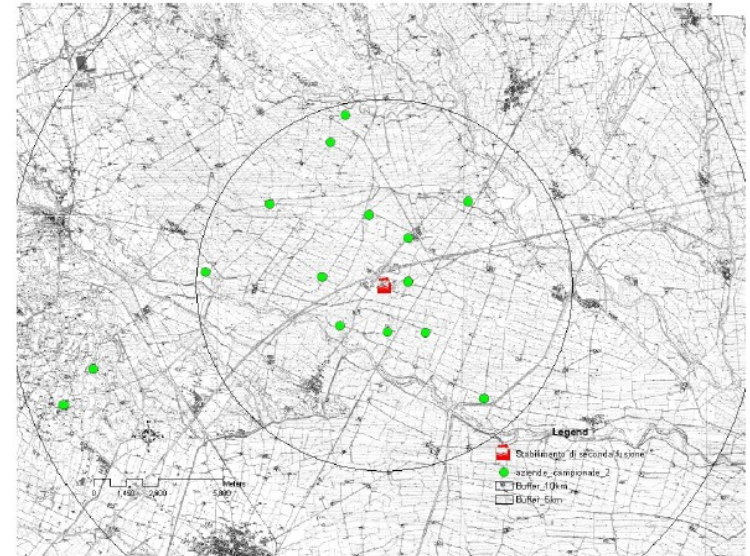
DL-PCB



median values in milk per farm

BACKGROUND

2) The area of Carisio: a second fusion aluminium factory



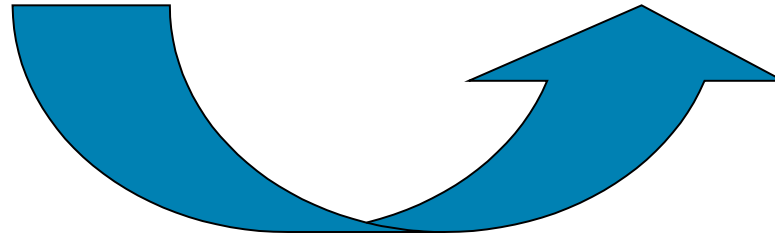
- **2004-2007 the ARPA (Agency for the environmental survey) identifies important dioxin like emissions**
 - **no bovine farms in the nearest area**
 - **biomonitoring on biological eggs**

MAT&MET - RESULTS



30 MILK SAMPLES FORM THE SUPERMARKETS

ALL COMPLIANT



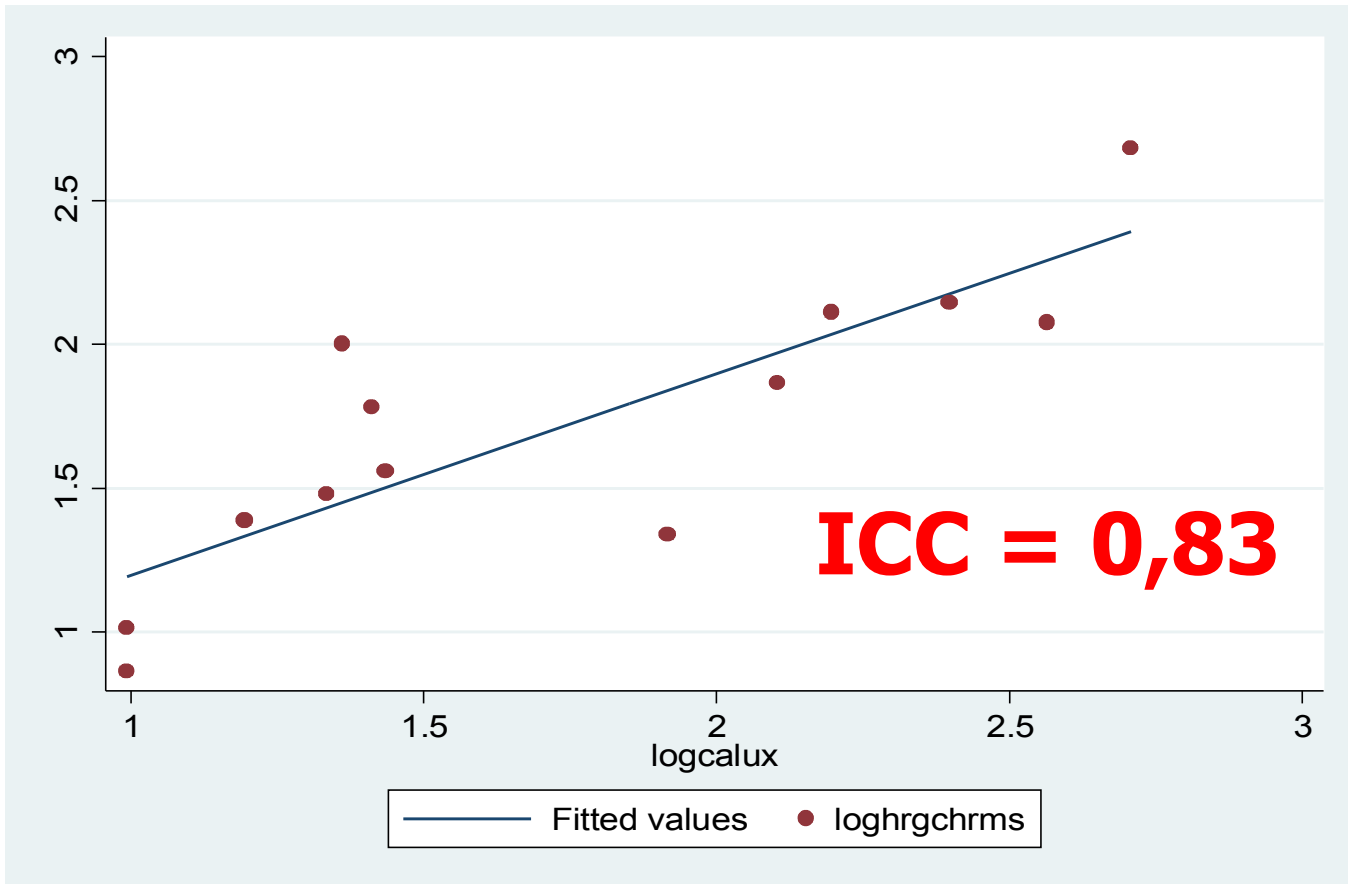
13 ROW MILK COMING FROM A CONTAMINATED AREA OF THE PIEDMONT REGION, PREVIOUSLY EXAMINED BY HRGC/HRMS



MAT&MET - RESULTS

ID	RISULTATO HRGC/HRMS pg/g grasso	RISULTATO DR-CALUX® pg/g grasso	DEV. ST. (±) DR- CALUX®
6-Val di Susa	8,5	11	0,31
13-Val di Susa	2,7	2,7	0,11
14-Val di Susa	4	3,3	0,18
15-Val di Susa	7,4	3,9	0,077
20-Val di Susa	3,8	6,8	0,22
21-Val di Susa	2,4	2,7	0,099
23-Val di Susa	7,9	13	0,77
31-Val di Susa	14,61	15	0,46
35-Val di Susa	5,93	4,1	0,21
40-Val di Susa	4,75	4,2	0,38
41-Val di Susa	6,45	8,2	0,23
46-Val di Susa	8,26	9	0,65
52-Val di Susa	4,39	3,8	0,17

**Scatter plot of the logarithm values obtained by Calux
versus the ones obtained by HRGC/HRMS,
In comparison with the fitted values**



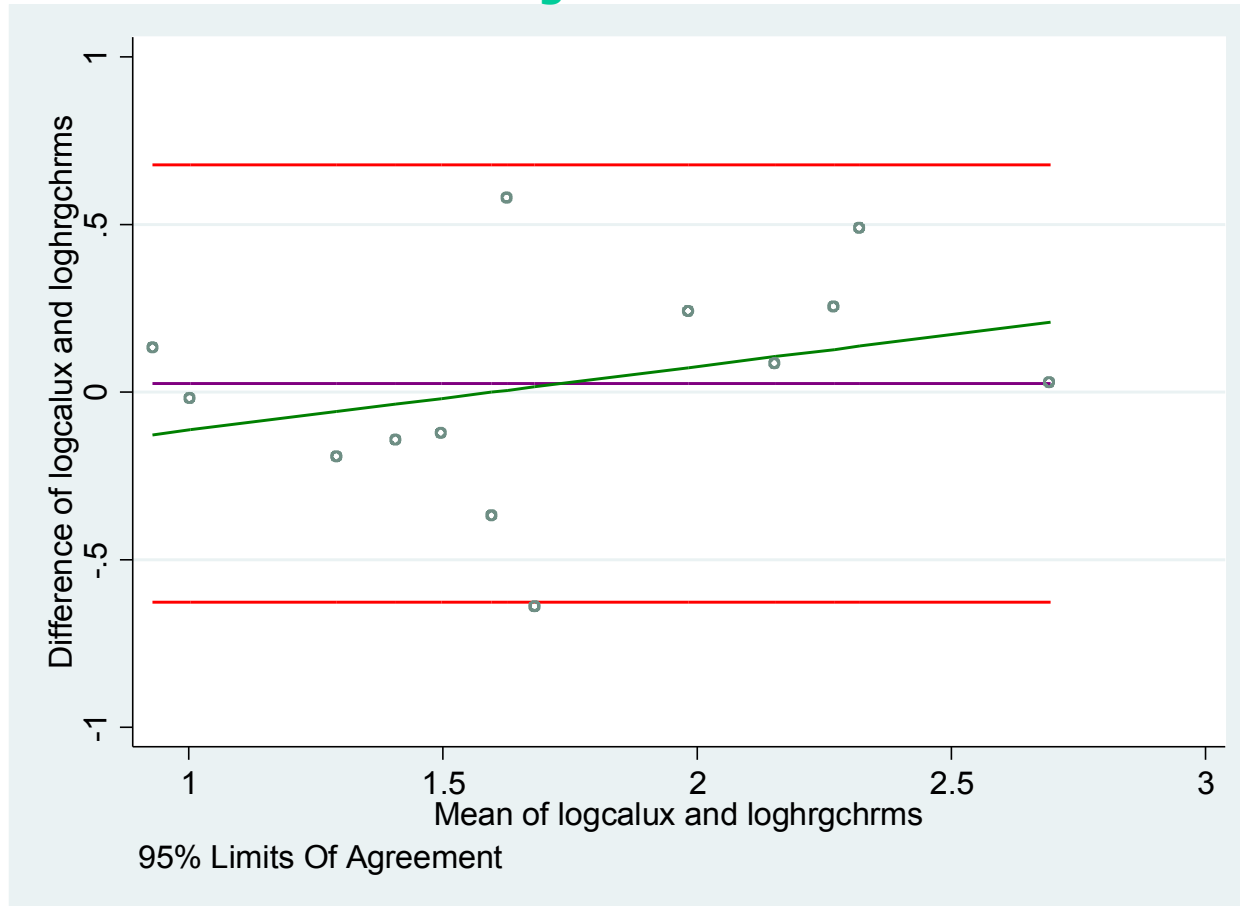


Bland Altman plot of milk log values:

it represents differences for coupled values in comparison with a mean value

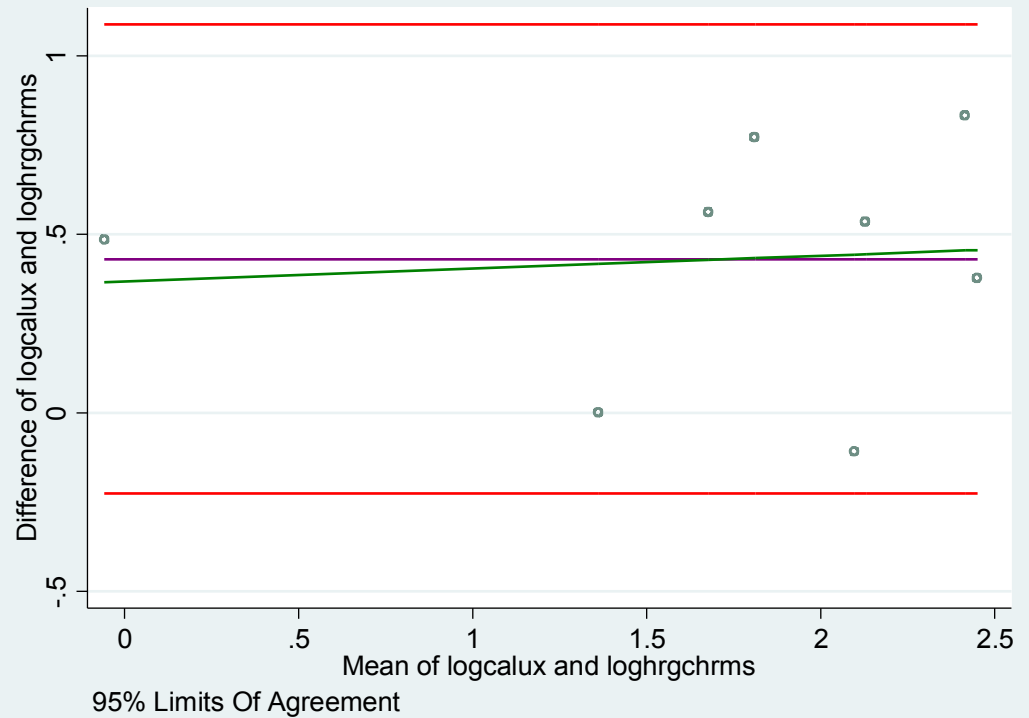
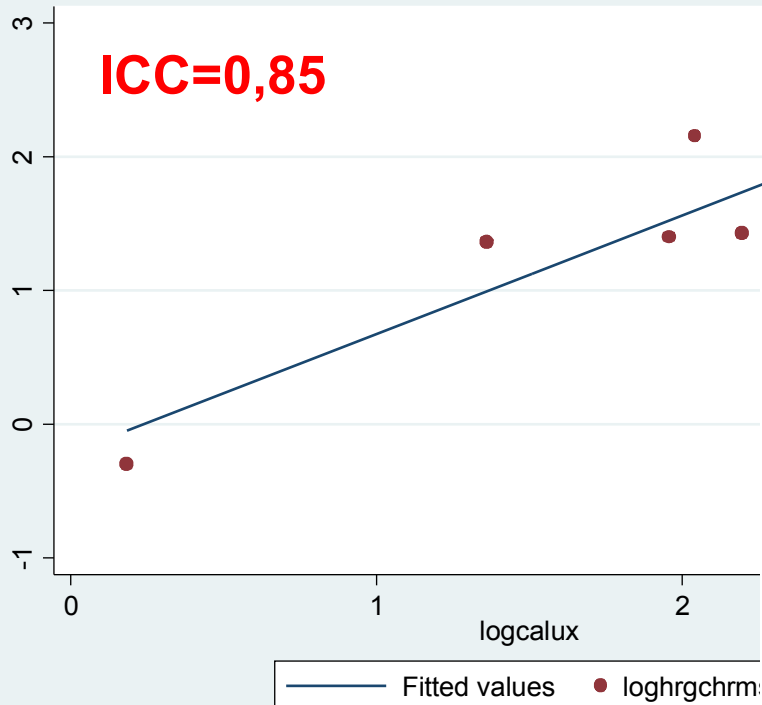
Violet line: the ideal condition in which the variance between the two groups is similar

Green line: the regression line from the data



... and the eggs?

ID	HRGC/HRMS sum PCDD/F+dl- PCB	DR-CALUX® sum PCDD/F+dl-PCB	Dev. st. DR- CALUX®
U7	62	27	0,14
U8	3,9	3,9	0,25
U9	8,6	7,7	0,23
U10	6,45	11	1,40
U12	9,6	14	0,53
U13	4,17	9	0,10
U14	4,05	7,09	0,09
U15	7,4	17	0,21
U16	0,74	1,2	0,09



CONCLUSIONS

OBSERVED VARIABILITY

mainly depends by the variance between the subjects (milk or egg samples),
but not by the variance into the subjects



There is no significant difference between the methods
affecting the obtained values of the analyzed samples

Intraclass Correlation Coefficient (ICC)

shows a good agreement between the Calux and HRGC/HRMS values

Anyways, due to the particular characteristics of the examined samples (mostly dl-PCB)

we have to:

- 1) Evaluate the results once divided by group of contaminants (PCDD/F and dl-PCB) – alumina columns
- 2) Re-test the method increasing the Nr. of the samples especially with compliant samples
- 3) Re-evaluate the data on BEQ basis

WORK IN PROGRESS ... RESEARCH



"To develop a nanotech device based on nanostructured functionalized surfaces for the detection of dioxins/PCBs contamination in food and feed"

Founding: Italian Ministry of Health

Matrices examined by DR-CALUX: milk, eggs, meat, vegetables, feed

"To develop screening biomolecular techniques for the detection of the dioxin and dioxin like contaminants exposure in cows"

Founding: European/Regional Founding Program (POR-FESR 2007-2013)

Matrices examined by DR-CALUX: bovine blood serum; bovine milk

WORK IN PROGRESS ... BIO - MONITORING



1) BIOMONITORING of the SIN PIEVE VERGONTE – Remediation Site of National Interest

To analyze by Calux and HRGC/MS 30 samples of ovi-caprine milk coming from farms located in a range of 10 km from the SIN -> AIM: to verify the level of contamination in the biota

2) BIOMONITORING of the AREA of the WASTE-TO ENERGY PLANT under construction

To analyze at least 20 milk and eggs samples coming from farms near the area -> AIM: to verify the basal condition of that area, before it become operational

3) BIOMONITORING of two RISK AREAS VAL SUSA and CARISIO 2012

To analyze at least 60 samples (milk and/or eggs) coming from farms in the range of the two risk areas -> AIM: to verify the evolution of the situation

Acknowledgements

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Thank you for your attention