

# Cluster workshop WP 2a & WP 2b

Coccidiostats & Antibiotics in Food and Feed

CONFIDENCE Open day (following Euroresidues VII)  
Egmond aan Zee, 16 May 2012



[www.confidence.eu](http://www.confidence.eu)



# What is CONFIDENCE?

---

- A **FP7 European project**: <http://www.confidence.eu>

## **CONTaminants in Food and Feed: Inexpensive DETection for Control of Exposure**

Grant agreement no.: 211326 – CP Collaborative Project

- Project **coordinator**: **Dr Jacob de Jong**, RIKILT, The Netherlands

Start: May 2008 – Duration: 56 months

- **Consortium**: **17** partners from **10** countries, representing **9** **research institutes**, **5** **universities**, **2** large **food** and **feed industries** and **1** **SME**



# Why has CONFIDENCE been set up?

---

- **RASFF** alerts: **monitoring** of **chemical contaminants** in **food** and **feed** very **relevant** in European **food safety**.
- **Consumers** placed chemical contaminants on **top of the “worry-scale”** of food-related risks.
- Often **expensive instrumental single-analyte methods** are being applied by regulatory and industrial laboratories.
- **urgent need** for replacement by validated screening tools which are **simple**, **inexpensive** and **rapid**, but also show **multiplex** capability by detecting as many contaminants in parallel as possible.



# CONFIDENCE overall objectives

---

- to provide **long-term solutions** to the **monitoring** of
  - ✓ persistent organic pollutants, perfluorinated compounds, pesticides
  - ✓ veterinary pharmaceuticals (coccidiostats, antibiotics)
  - ✓ heavy metals
  - ✓ biotoxins (alkaloids, marine toxins, mycotoxins)

in high-risk products such as **fish** and **fish feed**, cereal-based **food/feed** and **vegetables**.



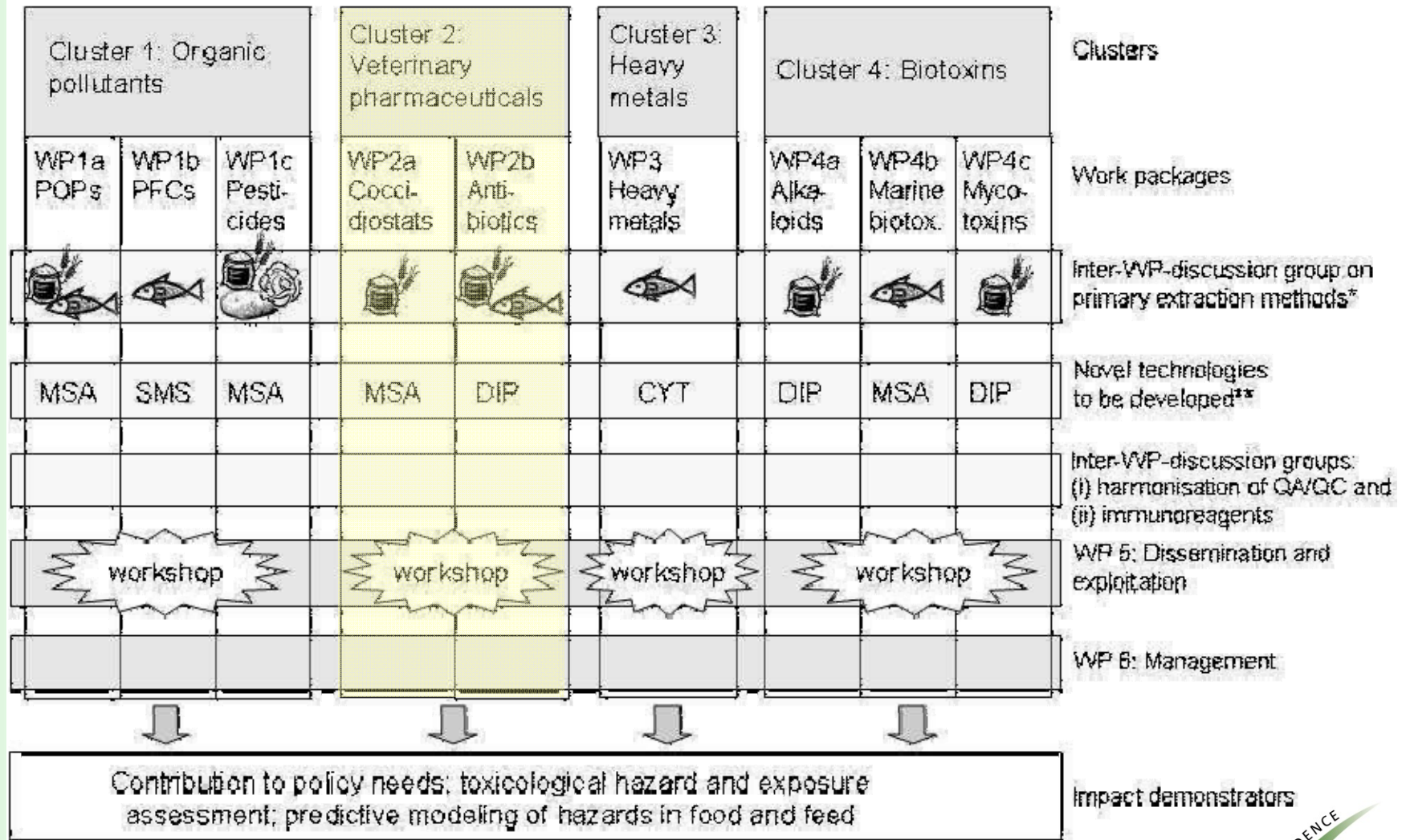
# CONFIDENCE overall objectives

---

- balanced mix of **novel multiplex technologies** including dipsticks, flow cytometry with functionalised beads, optical and electrochemical biosensors, cytosensors and metabolomics-like comprehensive profiling.
- **Validation** of the simplified methods, application in **impact demonstrators** that contribute to exposure, assessment and validation of hazard models.
- **Dissemination** to scientists and to relevant **stakeholders**



# How is CONFIDENCE organised?



\* = fish and fish feed,

= feed/cereals,

= potatoes/vegetables

\*\*MSA=multiplex screening assay, DIP= multi component dipstick assay, CYT=cytosensor, SMS = simplified MS method





# WELCOME TO THE CLUSTER 2

## WORKSHOP

### **WP 2a: Coccidiostats**

*Project leader: Dr Ursula Vincent (EC-JRC-IRMM, BE)*

*Deputy project leader: Dr Philippe Delahaut (CER groupe, BE)*

### **WP 2b: Antibiotics**

*Project leader: (Dr Sara Stead) – Mr Matthew Sharman (FERA, UK)*

*Deputy project leader: Dr Benoît Granier (Unisensor, BE)*



# CLUSTER 2 – DoW

## Rapid Methods for screening



### WP 2a

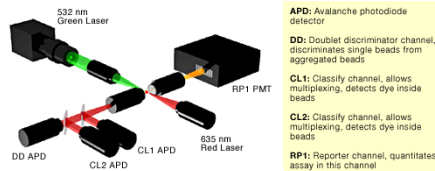
- **Analytes:** lasalocid A, monensin, salinomycin, narasin, and nicarbazin
- **Matrices:** eggs, laying hens feed
- **Technique:** Flow cytometry based immunoassay
- **Multiplex**
- **Development – optimisation**
- **Single-laboratory validation**
- **Inter-laboratory comparison**



### WP 2b

- **Analytes:** malachite green, tetracyclines, tylosin, chloramphenicol, quinolone and sulphonamide antibiotics
- **Matrices:** fish, feeds, urine, processed meat and honey
- **Technique:** Dipsticks
- **Single-component & Multiplex**
- **Development – optimisation**
- **Single-laboratory validation**
- **Inter-laboratory comparison**





# Objectives

## WP 2a

1. Validated **flow cytometry** based **multiplex immunoassay** for residues of lasalocid A, monensin, salinomycin, narasin and nicarbazine in **feed** and **eggs**

Policy support:

- **Screening method** addressing the needs of EC and EFSA to establish maximum tolerable levels in non target feeds
- **Cross-contamination** in non-targeted **feed** (laying hens feed)

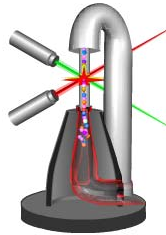
2. **Carry-over** study of lasalocid from **laying hens feed** to **eggs** aiming at contribution to a

## WP 2b

**Development, validation and impact demonstration of single-component and multiplex dipsticks to detect **malachite green, tetracyclines, tylosin, chloramphenicol, quinolone and sulfonamide** antibiotics in a range of matrices including fish, feeds, urine, processed meat and honey”**

# Scope - Need of sensitive methods!

## WP 2a



	LOD (DoW)		Max. Levels	
	Egg ( $\mu\text{g kg}^{-1}$ )	Feed ( $\text{mg kg}^{-1}$ )	Egg ( $\mu\text{g kg}^{-1}$ )	Feed ( $\text{mg kg}^{-1}$ )
Monensin	75	1.25	2*	1.25
Salinomycin	75	0.7	3	0.70
Narasin	75	0.7	2	0.70
Lasalocid	75	1.25	150	1.25
Diclazuril	-	-	2	0.01
Nicarbazin	100	0.5	100	1.25

\* in other food stuff of animal origin

Maximum content of coccidiostats in eggs (ML's) ( $\mu\text{g kg}^{-1}$  wet weight)

Maximum levels of unavoidable carry-over of coccidiostats in non-target feed ( $\text{mg of active substance/kg feed}$ )



# Scope - Need of sensitive methods!

WP 2b

**ZERO TOLERANCE LEVEL**

**MRPL**

**MRL where applicable**



## Status WP 2a



➤ All immunochemical reagents (antibodies and conjugates) produced and characterized



➤ All test materials (eggs & feed) produced – homogeneity OK



➤ Protocol and prototype test of the multiplex assay for coccidiostats in buffer – Done

➤ Protocol and prototype test kit for coccidiostats in feed and eggs including simplified sample preparation procedures – Done



➤ Single-laboratory validation – Performed

➤ Performance characteristics of the new method vs. existing ELISA, HPLC and LC-MS/MS methods – Performed

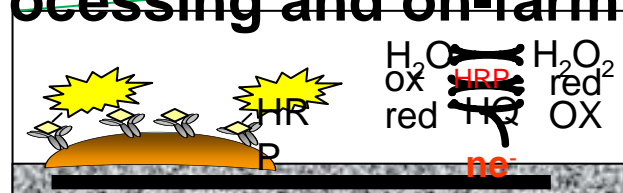
➤ Small-scale interlab study – Launched

➤ Carry-over study of lasalocid from laying hens feed to eggs using the FCIA – Started



## Status WP 2b

- All immunochemical reagents (antibodies and conjugates) produced and characterized
- All test materials produced – homogeneity OK
- Development / optimisation of (multiplex) dipsticks – Done
- Assay optimisation and comparison to current technology – Performed
- Single-laboratory validation (2002/657/EC) – Performed
- Interlab study – Done (data treatment on-going)
- Impact demonstrators: regional survey of malachite green/LMG in salmon and antibiotics in honey and contribution to validation of a predictive model for the fate of the tetracycline hazard from farm to fork including the effect of food processing and on-farm demonstration – Started



CSIC



Thanks for your attention





➤ <http://www.confidence.eu>

