



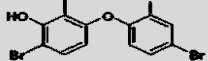
# Biorecognition-based multiplex detection of chemicals in food and feed

A. Meimaridou<sup>1</sup>, W. Haasnoot<sup>1</sup>, M.E. Bienenmann-Ploum<sup>1</sup>, M.W.F. Nielen<sup>1,2</sup>

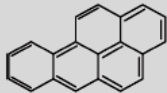
## Objectives

The development of Luminex flow cytometry-based screening assays for the detection of several small compounds:

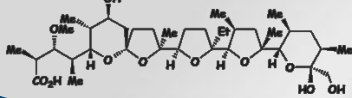
- potential thyroid disrupting compounds such as hydroxylated metabolites of polybrominated diphenylethers (OH-PBDEs) in fish, fish feed.



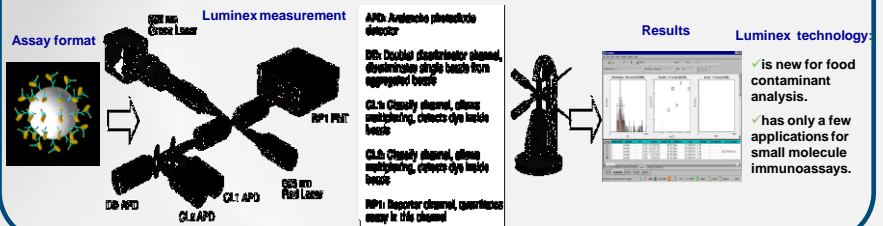
- polycyclic aromatic hydrocarbons (PAHs) in cereals.



- priority coccidiostats such as lasalocid, monensin, narasin, nicarbazin and salinomycin in laying hens feed.



## Luminex (flow cytometry) principle



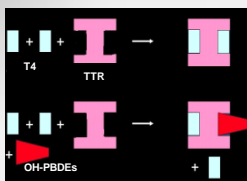
## Challenges of the project

- Microsphere coupling chemistries (for the design of the different assays using either transport proteins or antibodies).
- The specificity of the biochemicals in multiplex assays for assay performances and their inter-assay interactions under the influence of different coupling strategies.
- Generic sample preparation procedures for the multiplex assays.

## LUMINEX BASED ASSAY'S FORMAT

### OH-PBDEs

#### THYROID DISRUPTING MECHANISM



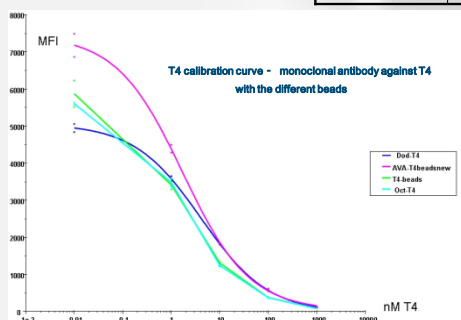
#### LUMINEX BASED ASSAY



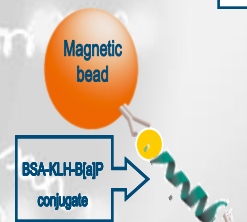
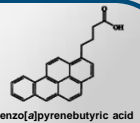
- Thyroxine (T4) immobilization on the bead surface via spacer.
- Addition of transthyretin (TTR).
- Bound TTR detection with monoclonal antibody against TTR.
- Addition of the secondary antibody conjugated with Phycoerythrin (PE).
- Identification and quantification by Luminex.

Spacers	Structure
5-Aminovaleric acid	
8-amino octanoic acid	
12-amino dodecanoic acid	
carboxypolyethylene glycol(e) amine	

#### T4-beads



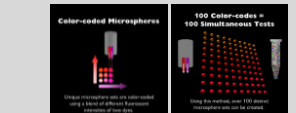
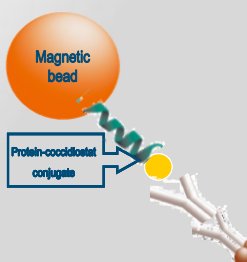
### PAHs



- Immobilization of the monoclonal antibody against benzo[a]pyrene (B[a]P).
  - Binding of the benzo[a]pyrene conjugate\*.
  - Addition of the anti-protein PE.
  - Identification and quantification by Luminex.
- \* this conjugate can be benzo[a]pyrene linked with BSA (bovine serum albumin) or KLH (Keyhole limpet hemocyanin)

### COCCIDIOSTATS

#### 6-MULTIPLEX LUMINEX BASED ASSAY



- Immobilization of different coccidiostats conjugates.
- Binding of the antibody against coccidiostats.
- Addition of secondary antibody conjugated with (PE).
- Identification and quantification by Luminex.

