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# Chemical mixtures: challenges for research and risk assessment?

Conference

10&11

DECEMBER 2013

Maison de la RATP - Espace du Centenaire
189, rue de Bercy
75012 Paris

Rencontres scientifiques de l'Anses

# Grouping of pesticide active substances for cumulative risk assessment

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10&11 DECEMBER 2013

# Background

Cumulative risk assessment: Requirement in Reg. (EC) No 396/2005 (MRL regulation)

→**Cumulative and synergistic effects** should be taken into account in dietary risk assessment **when methods** to assess such effects **are available**.

## Development of the methodology

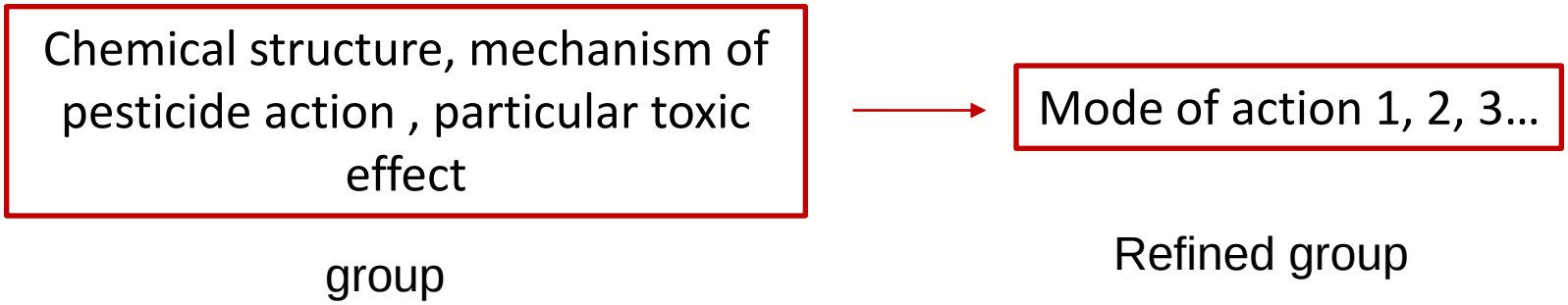
- Ongoing by EFSA
- Internal and external reports

# Background

EFSA, 2008; 2009: methodologies for a selected group of pesticides

Identification of pesticides to be included in cumulative assessment groups (CAG) based on their toxicological profile

→Tiered approach for hazard assessment




Outsourced project (Danish Technical University , 2012)

- Application of this procedure to organs or systems

Toxicolc ping
of pesti ive risk
assessn
- on live
- on the
- on rep

Supporting Publications
2013:EN-392

**EXTERNAL SCIENTIFIC REPORT**

**CFT/EFSA/PRAS/2012/07-CT 01, 02 and 03**

**"Toxicological data analysis to support grouping of pesticide active substances for cumulative risk assessment of effects on liver, on the nervous system and on reproduction and development"**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Lot 1:
Nervous system effects

Lot 2:
Liver (including biliary system and gallblader)

Rijksinstituut voor Volksgezondheid en Milieu
*Ministerie van Volksgezondheid, Welzijn en Sport*

**Consortium**

Lot 3:
Developmental and reproductive toxicity

anses
agence nationale de sécurité sanitaire alimentation, environnement, travail

# Objectives

Collection and analysis

- Determination of toxic effects of pesticides on reproduction and development in laboratory animals

➢ Consolidation DTU, 2012

- Active substances approved until 31 december 2009

➢ Pesticide active approved until 31 december 2011

→ 257 active substances

- Identification of Mechanism/mode of action for the effect

**Grouping** ( consideration by EFSA PPR Pannel)

# Methodology
# **Source of information**

## **Pesticides Draft Assessment Reports (DAR)**

- EU peer review documents
(addendum, PRAPeR documents …)
- Open literature, JMPR reports, CLH,
- Study reports when necessary

**Studies (summary)**

# Methodology
## **Screening of regulatory toxicity test**

- Short-term, Sub-chronic and chronic toxicity studies
- One and multi-generations reproduction studies
- Prenatal developmental toxicity studies
- Developmental neurotoxicity studies
- Mechanistic studies on reproductive effects

# Methodology:
# **determination of endpoints**

## Reproductive toxicity

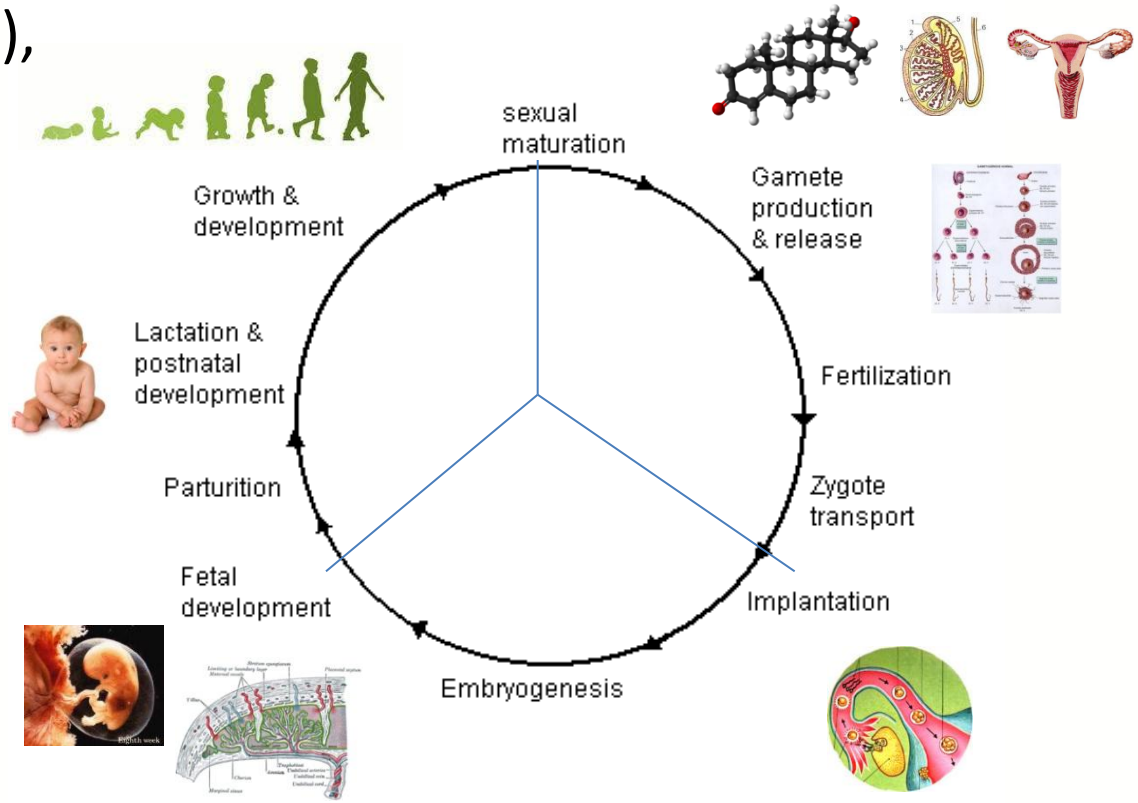
Sexual behaviour and fertility (males and females), parturition and lactation

## Developmental toxicity

Growth,
Structural or functional development
Mortality

## Tumor on reproductive organs

## The reproductive cycle



# Methodology:
# **determination of endpoints**

Developmental toxicity: mortality, structural abnormality, altered growth and functional impairment

| Prenatal |
| --- |
| Prenatal body weight changes |
| Delayed prenatal development |
| Post-implantation losses |
| Runts/stillbirth |
| **Malformations** |
| Skeletal malformations |
| Visceral variations |
| Skeletal variations |

| |
| --- |
| **Postnatal bw changes** |
| Delayed postnatal development |
| **Post-natal death** |
| Reduced litter size |
| ↑/↓ anogenital distance of ♂/♀ offsprings |
| Nipples retention in ♂/♀ offsprings |
| Preputial separation of offsprings |
| Delayed vaginal opening of offsprings |
| Clinical signs in offsprings |
| ↑/↓ weight of ♂/♀ offsprings reproductive organs |
| ↑/↓ weight of offsprings endocrine organs |
| ↑/↓ weight of offsprings other organs |
| Pathological changes of ♂/♀ offsprings reproductive organs |
| Pathological changes of offsprings endocrine organs |
| Pathological changes of offsprings other organs |
| Altered sperm in ♂ offsprings |
| **Learning and memory** |

# Methodology: **determination of endpoints**

Reproductive toxicity: Alterations of the female or male reproductive organs, the related endocrine system, or pregnancy outcomes.

| males | Females | Males/females |
|---|---|---|
| Altered male sexual behavior | Hormonal changes in females | Decreased fertility index |
| Hormonal changes in males | ↑/↓ weight of female reproductive organs | Postnatal death |
| ↑/↓ weight of male reproductive organs | Pathological changes of female reproductive organs | Decreased gestation index |
| **Pathological changes in male reproductive organs** | Altered maternal behaviour | Increased gestation length |
| | Oestrus cycle | Decreased mating index |
| | Increased placenta weight | Pre-implantation losses |
| | Lactation | Post-implantation losses |
| | | Reduced litter size |
| | | Sex ratio |

# Methodology:
# **determination of enpoints**

## Reproductive organ tumor induction

| Mammary gland tumors |
| --- |
| Ovarian tumors |
| Uterus tumors |
| Testis tumors |
| Prostate tumors |

# Methodology:
# **further considerations**

Need to distinguish:

- General maternal toxicity *vs.* developmental effect
- General toxicity *vs.* reproductive effect

→ **Included for each endpoint (indicator)**

# Database collection

## **Excel Database** (257 active substances, 5246 endpoints)

- Active substance (identification)
- Study type,
- Species, strain,
- Route, type of administration,

database

<u>For each endpoint:</u>

- NO(A)EL/LO(A)EL
- MoA/MeA (when available)
- Maternal/parental/general toxicity
- Year of regulatory evaluation

# Further steps
# **Identification of specific effects**

Defining **specific effects** for establishing CAG

- Not a local effect, not ambiguous, specific and adverse
- Relevance for human

Need to take into account (identify during the project):

- Same endpoint may be reported with different names, depending on the DAR and the experimental studies (runts/stunts/nanofoetuses…)
- General way may need further interpretation (e.g. anomalies *v.s.* malformations/variations
- Effects which occurs at high dose (general toxicity/maternal toxicity) or specific effects

# Identification of MoA

## Outcomes of the project

- Mainly based on open literature data
  - Interpretation, lack of experimental details?
  - Not peer-reviewed at EU-level
- Experimental mechanistic studies only available for a small number of pesticides
  - Limited during EU review process
  - Non-homogeneity of the data
- MoA/MeA are often very complex for reproductive toxicity
- Confirmation of the MoA necessary in many cases
- Relevance of MoA regarding human health

# Identification of MoA

## Examples of presume MoAs for developmental effects (DAR)

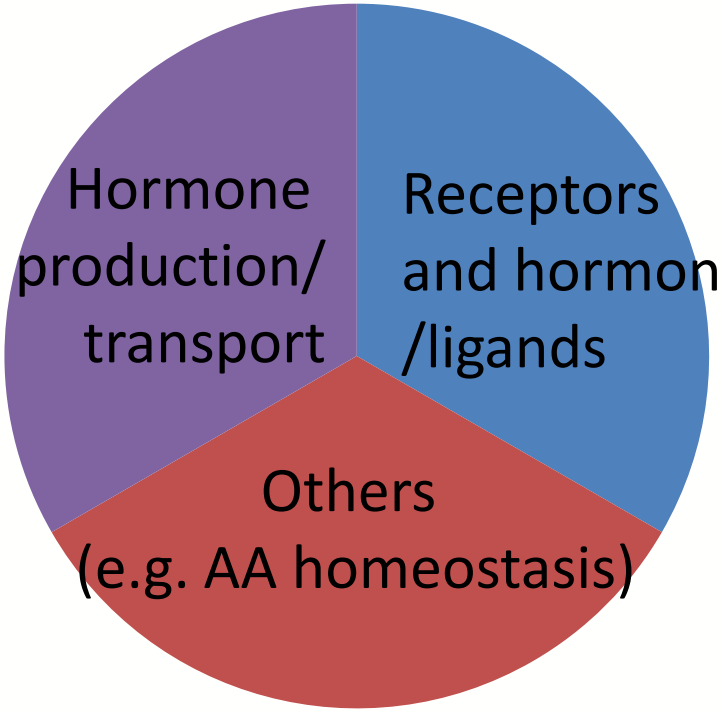
| MoA | Name of the pesticides active substances | Developmental effect |
|---|---|---|
| **Inhibition of embryonic CYP26 and/or blockade of Ikr potassium (HERG) channel** | Flusilazole<br>Epoxiconazole | Craniofacial or brain malformations (Menegola, 2006) |
| **Anaemia** | Flumioxazin | Ventricular septeal defects |
| **Inhibition of glutamine synthetase** | Glufosinate | **Post-implantation losses** |

# Identification of MoA

## Examples of presume MoAs for reproductive /tumors induction

Thyroid hormone homeostasis

Steroid hormone homeostasis




Androgen

Estrogen

# Further steps
# **Grouping of pesticides**

Mechanism of toxic action v.s. common toxic effects

More robust

Refined assessment

Available for only few compounds

Overestimation

More compounds

# Conclusions/perspectives

- Lot of ongoing work (EFSA opinion, 2013; new EFSA call)

- Need to develop in parallel exposure assessment (e.g. acropolis)

- Need to clearly define and harmonized some parameters :
  → Identification of pesticides to be included in CAG
  → Specific effects
  → Additivity/synergism
  → Susceptibility of foetuses

# Conclusions/perspectives

- Cumulative risk assessment is **challenging** and lot of work are ongoing (national, european and international)
- **No** methodology is currently internationally approved with **consensus**
- Currently difficult to apply a methodology in a **routine basis** for regulatory risk assessment
- Further application **non-dietary exposure** (ie. Operator, worker, bystander, resident exposure)

# References


- **EFSA (European Food Safety Authority), 2008.** Opinion of the Scientific Panel on Plant Protection products and their residues to evaluate the suitability of existing methodologies and, if appropriate, the identification of new approaches to assess cumulative and synergistic risks from pesticides to human health with a view to set MRLs for those pesticides in the frame of Regulation (EC) 396/2005. The EFSA Journal 2008, 704, 1-84.
- **EFSA (European Food Safety Authority), 2009.** Opinion on Risk Assessment for a selected group of pesticides from the triazole group to test possible methodologies to assess cumulative effects from exposure through food from these pesticides on human health. The EFSA Journal 2009, 7, 1167.
- **Danish Technical University (DTU), 2012.** Identification of Cumulative Assessment Groups of pesticides. External scientific report submitted to EFSA.
- **ANSES/ICPS/RIVM, 2013.** CFT/EFSA/PRAS/2012/07, 2013. Lot1 (RIVM), Lot2 (AOSACCO/ICPS), Lot3 (ANSES). Toxicological data analysis to support grouping of pesticide active substances for cumulative risk assessment of effects on liver, on the nervous system and on reproduction and development.
- **EFSA (European Food Safety Authority), 2013.** Scientific Opinion on the identification of pesticides to be included in cumulative assessment groups on the basis of their toxicological profile EFSA Journal 2013;11(7):3293. [131 pp.]. doi:10.2903/j.efsa.2013.3293.

# Thank you for your attention

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