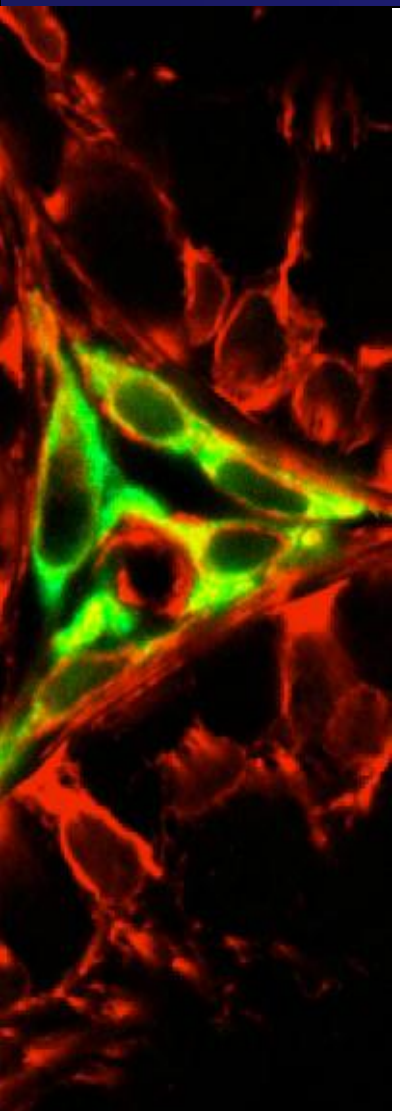
ED event European Commission



Endocrine disrupters – Identification and criteria for regulation

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European Commission 11-12 June 2012

Political/legal background

- Biocide regulation
- EU plant protection product regulation (EC 1107/2009)
  - Hazard-based cut-off criteria for endocrine disrupters
- REACH (EC 1907/2006)
  - Endocrine disrupters require authorisation
  - Authorisation not to be granted if risks are hard to manage

The issue

- Regulations do not provide criteria for assessment and decision about endocrine disrupting properties
- European Commission has to fill this gap
- **Principle**: uniform criteria across regulations

Three elements

What is an endocrine disrupter?

Definition (what is it you want to deal with?)

Tests (do you have the tools to identify an EDC?)

Criteria (how to translate test outcomes into regulatory decisions?)

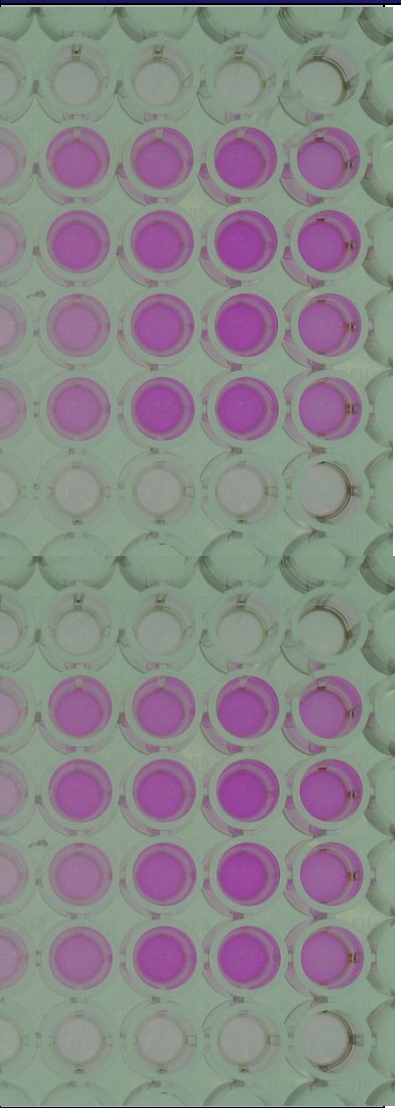
Definition

- WHO/IPCS definition
- “An endocrine disrupter is an exogenous substance or mixture that alters function(s) of the endocrine system and consequently causes adverse health effects in an intact organism, or its progeny, or (sub)populations.”

- Does not define the endocrine system

- **Adversity** – whole animal tests
- Endocrine **mode of action**

Identification of EDCs

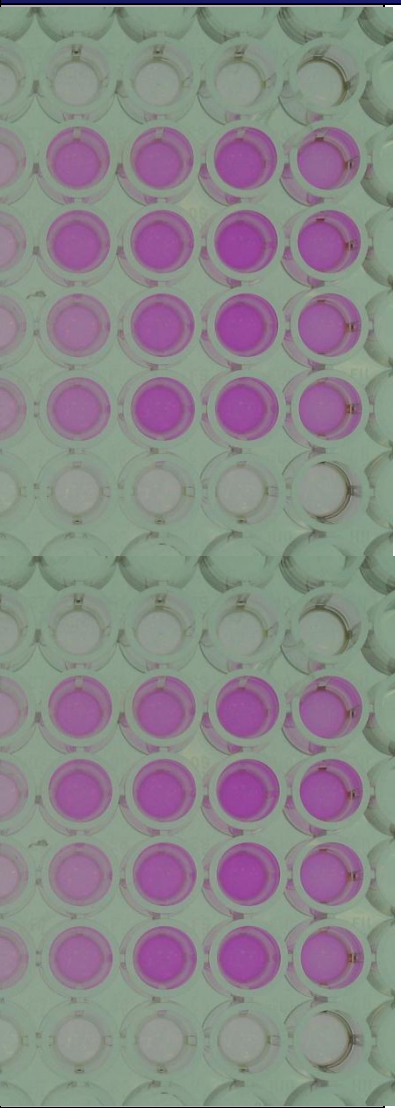


- Can ED effects be anticipated from existing toxicological data?
- For example pesticide dossiers?

Anticipation of ED activity by regulatory bodies

Active substance	In vitro activity	KEMI	UK PSD	UK HSE
Bitertanol	AR anta		yes	
Propanil	AR anta		yes	
Prochloraz	AR anta		yes	
Bifenox	AR anta			
Diuron	AR anta			
Propiconazole	AR anta		yes	
Fenarimol	AR anta, ER		yes	yes
Cyflutrin	AR anta, ER			
Pendimethalin	AR anta, ER			
Cyprodinil	AR anta			
Perimethanil	AR anta			
Fludioxonil	AR anta			

Tests for identifying ED properties

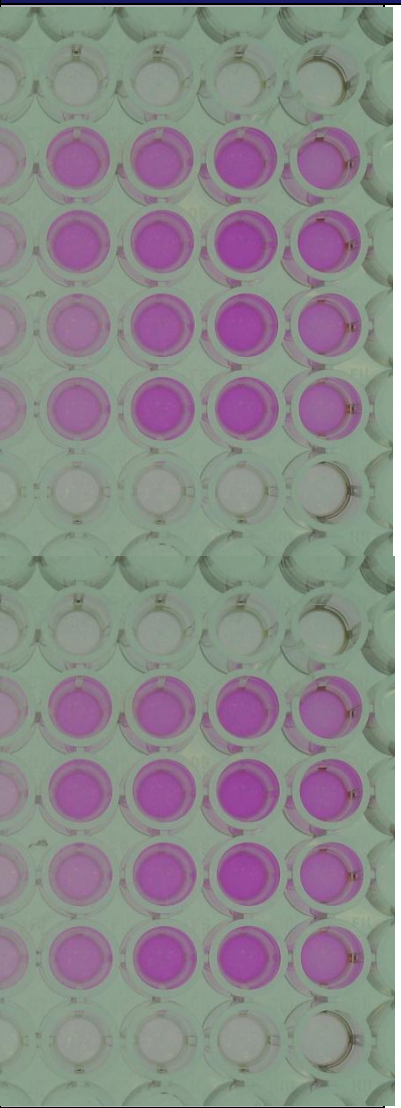


- Validated and internationally agreed test methods
- This **severely limits** the range of ED effects that can currently become subject to regulation

OECD framework EDC

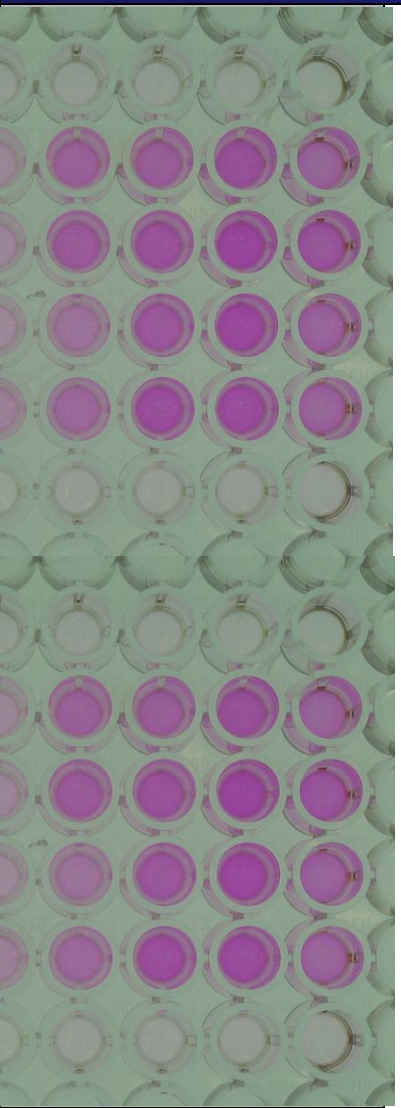
Level 1 Sorting & prioritization based upon existing information	- physical & chemical properties, e.g., MW, reactivity, volatility, biodegradability - human & environmental exposure, e.g., production volume, release, use patterns - hazard, e.g., available toxicological data	
Level 2 *In vitro* assays providing mechanistic data	- ER, AR, TR receptor binding affinity - Transcriptional activation - Aromatas and steroidogenesi *in vitro* - Aryl hydrocarbon receptor recognition/binding - QSAR	-High Through Put Prescreens - Thyroid function - Fish hepatocyt VTG assay - Others (as appropriate)
Level 3 *In vivo* assays providing data about single endocrine mechanisms and effects	- Uterotrophi assay (estrogenic related) - Hershberger assay (androgenic related) - Non -receptor mediated hormone function - Others (e.g. thyroid)	- Fish VTG vitellogenin) (estrogenic related)
Level 4 *In vivo* assays providing data about multiple endocrine mechanisms and effects	- enhanced OECD 407 (endpoints based on endocrine mechanisms) - male and female pubertal assays - adult intact male assay	- Fish gonadal histopathology assay - Frog metamorphosis assay
Level 5 *In vivo* assays providing data on effects from endocrine & other mechanisms	- 1-generation assay (TG415 enhanced) [1] - 2-generation assay (TG416 enhanced) [1] - reproductive screening test (TG421 enhanced) - combined 28 day/reproduction screening test (TG 422 enhanced) 1 Potential enhancements will be considered by VM63 mmm	- Partial and full life cycle assays in fish, birds, amphibians & invertebrates (developmental and reproduction)

Tests – general principles



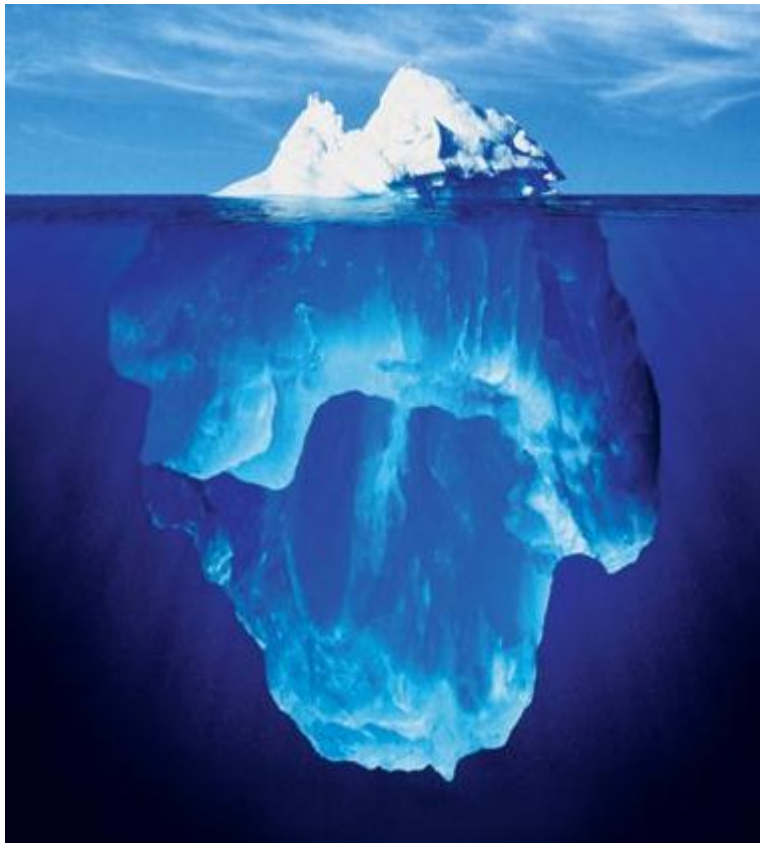
- Demonstrate **adverse** effects in **whole** organisms – *Level 5 OECD*
- Capture an **endocrine mechanism** – *Level 2 OECD*

Tests – effects currently not covered



- Carcinogenicity by endocrine modes of action
- Female reproductive health
- Metabolic syndrome, diabetes
- Any mode of action outside EAT

ED testing



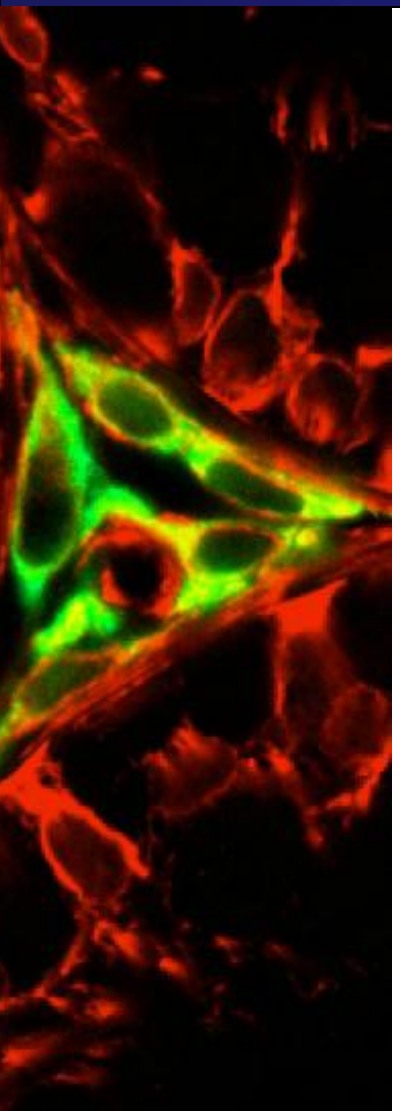
Current testing requirements

OECD Conceptual Framework

guidance is not yet drafted or those included in the Detailed Review Paper

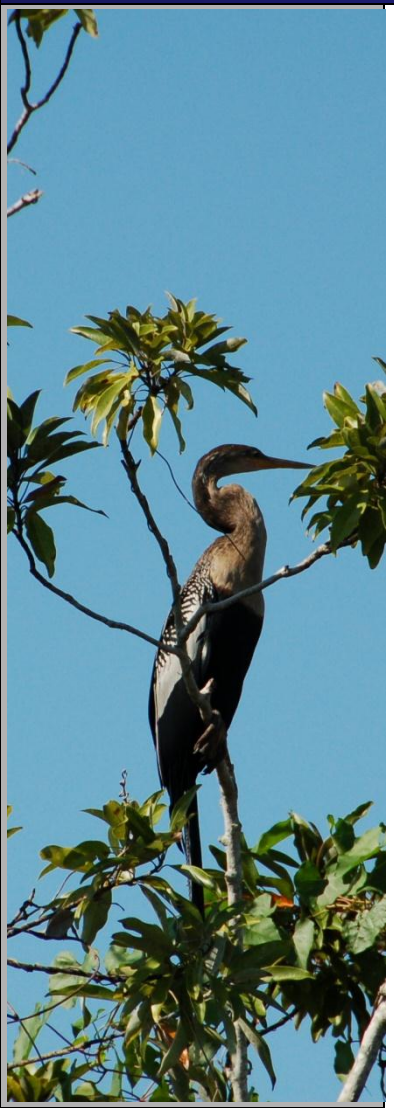
Other receptors /pathways

Tests: PPPR – Human toxicology



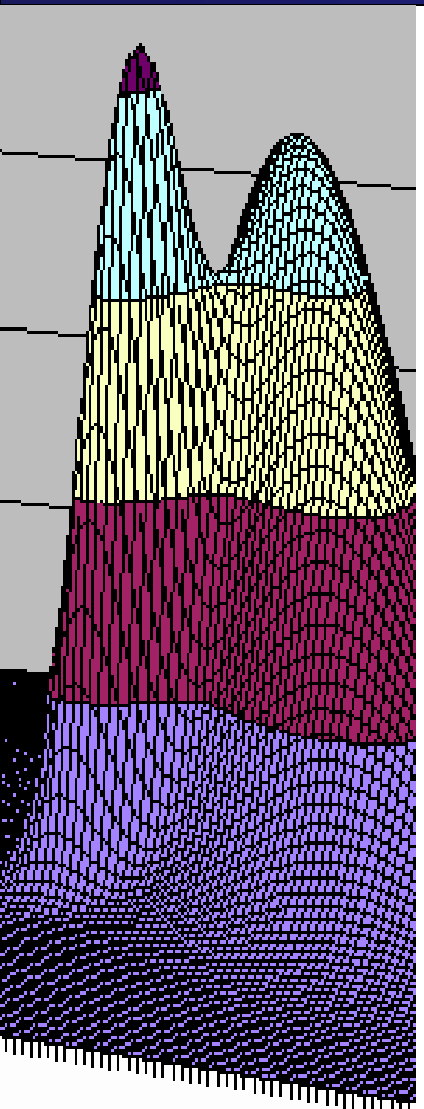
- **Update** Commission Regulations on requirements for active substances (544/2011) and products (545/2011)
- **Minimum requirements for EDC identification**, achievable immediately:
- Addition of endpoints relevant to ED in reproductive toxicity studies
- Two-generation repro (TG 416) or extended one-generation (draft TG 433)
- OECD Level 2 assays (to establish MoA)
- Merit of including OECD Level 3 and 4?

Tests: PPPR – Ecotoxicology



- **Problem:** Guidelines not worked out to sufficient detail at OECD Levels 4 or 5
- Chironomid life cycle (TG 233), daphnia reproduction (TG 211)
- No other OECD Level 5 guideline
- Level 4: Avian reproduction (TG 206), chironomid toxicity (TG 218-219), fish sexual development (draft TG 234)
- **Update** of information requirements PPPR: consider inclusion of all validated TG at Level 2-4

Tests: REACH



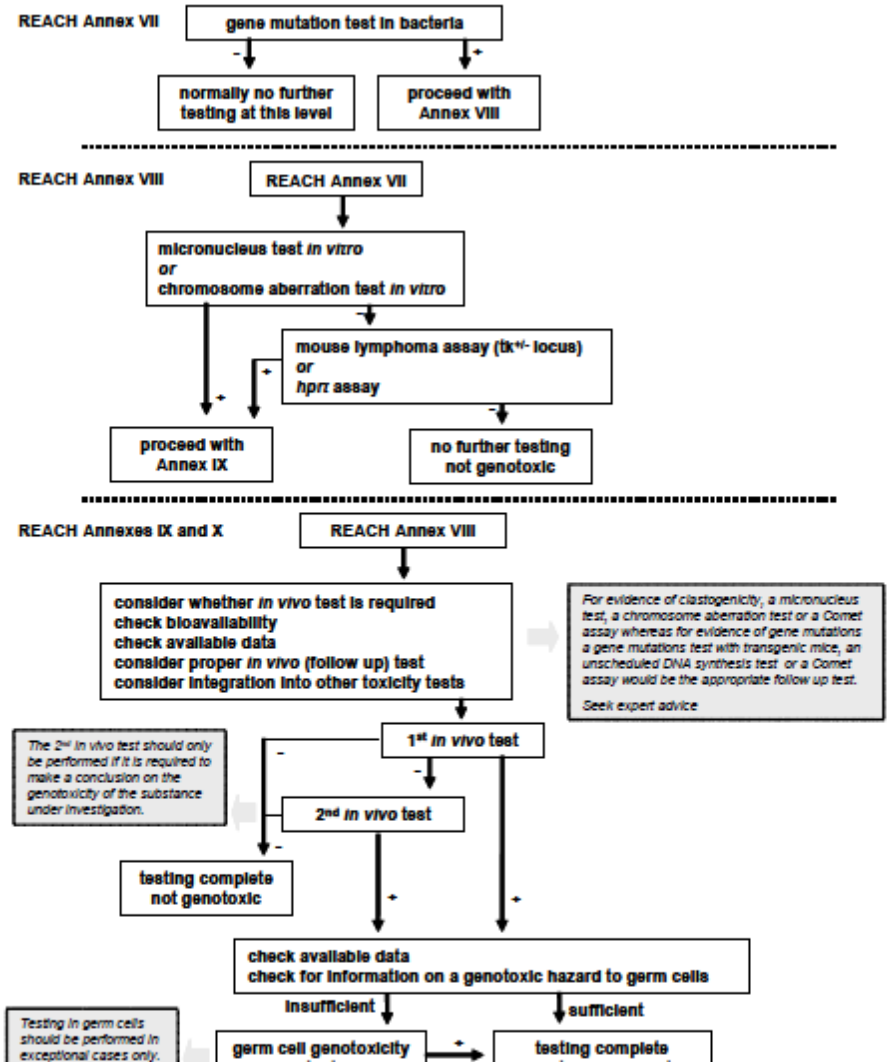
- Similar considerations apply
- Differentiation according to tonnage

- Lack of correlations between Level 2 and Level 4, 5 assays
- Difficult to decide on waiving of testing in case of positive results

Testing strategies and waiving of tests

Example: Mutagenicity testing in REACH

Equivalent schemes for EDCs not worked out

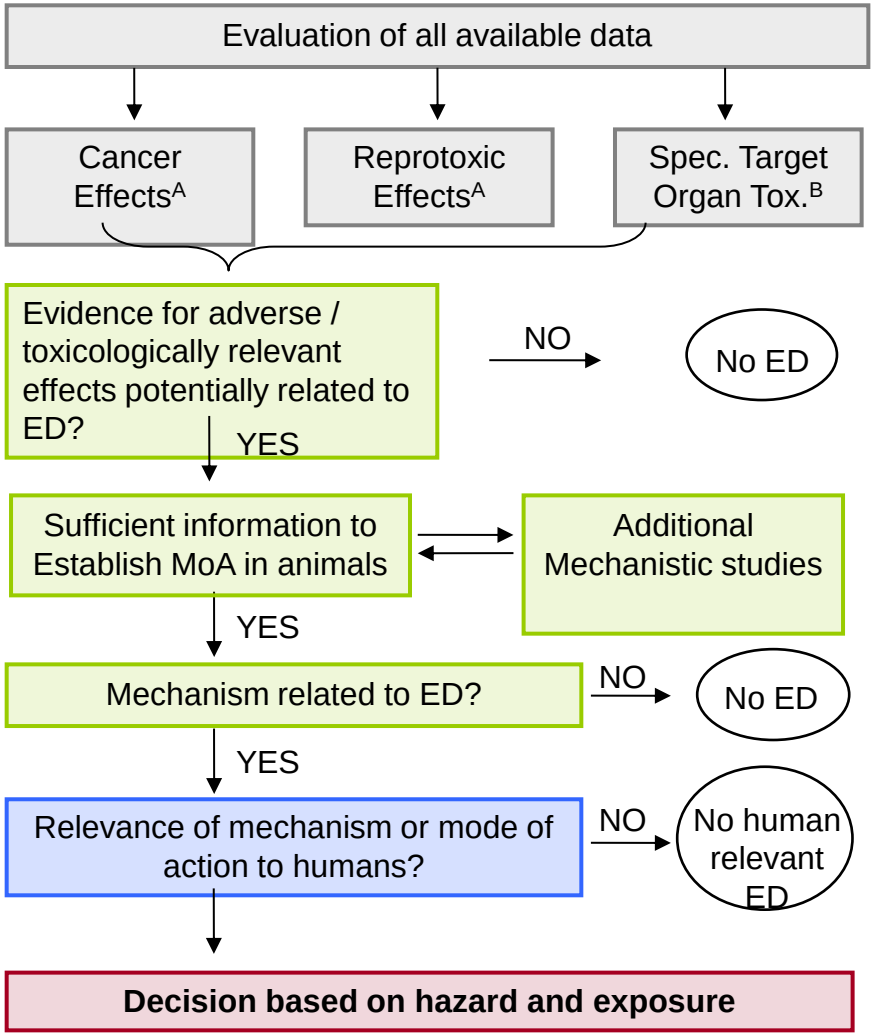


Criteria for EDCs: Initiatives in 2008-9

Focus: pesticides, human/ mammalian toxicity

- European Centre for Ecotoxicology and Toxicology of Chemicals (ECETOC)
  - Workshop
  - Technical Report 106
- German Federal Institute for Risk Assessment (BfR)
  - Workshop
  - Decision tree

BfR decision tree



BfR approach: option 2 – classification criteria

- Principles for hazard classification and labelling of Regulation (EC) 1272/2008 on classification, labelling and packaging
- adjust the basic classification criteria for substances causing **specific target organ toxicity** (STOT-ED)
- **Category 1**: ED in humans
  - 1 a: Epidemiological evidence
  - 1 b: Animal studies with human relevance, severe effects, low doses
- **Category 2**: ED in animals, with presumed relevance to humans

Criteria for regulatory decisions

- STOT-ED categories 1 a and 1 b fall under cut-off criterion: no approval
- STOT-ED category 2 shall **not** fall under cut-off criterion

Study type	STOT-ED 1 mg/kg/d	STOT-ED 2 mg/kg/d
28-day oral toxicity	≤ 30	≤ 300
90-day oral toxicity	≤ 10	≤ 100
Chronic toxicity	≤ 5	≤ 50

Known EDCs and cut-off values

90 day:
< 10 mg/kg d

Substance	NOAEL mg/kg bw/day	LOAEL mg/kg bw/day	Adverse effect(s) at LOAELs	LOAELs below 10 mg/kg bw/d?	LOAELs below 100 mg/kg bw/d?	References
DEHP	3 100 5	10 300	↓ AGD, ↑ Nipple retention, rat ↓ Testosterone GD 18, rat *Reproduktion (germ cell depletion, ↓ testis weight), developmental tox, rat*	Maybe?	Maybe?	Christiansen et al (2010) (27) Howdeshell et al 2008 (1) *Wolfe and Leyton, 2003 (*)EU RAR, EFSA*
DiNP	750 300 -	900 600 *750*	↓ AGD, rat ↑ Nipple retention, rat ↑ Nipple retention, rat	No	No	Boberg et al (2010) (28) Boberg et al (2010) (28) *Gray et al 2000 (2)* Exxon 1996 (*)
DnBP	- 250 50 10 100 -	250 500 250 50 300 (52) ***2***	↓ AGD, rat ↓ AGD, rat ↓ AGD, rat ↓ Testosterone GD 19, rat ↓ Testosterone GD 18, rat Embryotoxicity, rat *Germ cell development, mammary gland changes,*	No	No	Ema & Miyawaki 2001 (3) Jiang 2007 (4) Zhang 2004 (5) Lehmann et al 2004(6) Howdeshell et al 2008 (1) Wine et al 1997 (7) *Lee 2004 (8)*
1016 PCB77 *PCB126*			testosterone, rat			
(DDT) pp DDE		*10* 100	↑ Nipple retention, rat ↓ AGD, rat	Maybe	Yes	*You 1998)(18)* You 1998 (18)
Butylpara-ben		10 (100) 100 600 200	↓ sperm production, young rats (↓Testosterone, ↓ epidi-dymis weight) Sperm count Uterotrophic, rat Uterotrophic, rat (dry 200, wet 600)	No or ?	No or ?	Oishi 2001 (21) Kang et al 2002 (22) Hossaini et al 2000 (20) Rout-ledge 1998 (23)
Isobutyl-paraben	100	72 250	Uterotrophic, mouse Uterotrophic, rat	No	No	Darbre et al 2002 (24) Koda et al 2005 (25)

AGD = anogenital distance; GD = gestation day

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Known EDCs and cut-off values

90 day:
< 10 mg/kg d

Substance	NOAEL mg/kg bw/day	LOAEL mg/kg bw/day	Adverse effect(s) at LOAELs	LOAELs below 10 mg/kg bw/d?	LOAELs below 100 mg/kg bw/d?	References
			rat			
DiBP	**125** *100*	250 300	↓ AGD, ↑ Nipple retention, rat ↓ *Testosterone GD 18, rat*	No	No	Sallenfait et al 2008 (9) *Howdeshell et al 2008 (1)*
BBP	***50*** 167 100 100 185 182	250 250 500 300 375 910	↓ *AGD, rat* ↓ AGD (GD 21), rat ↓ AGD, rat ↓ Testosterone GD 18, rat Developmental toxicity, mice Developmental toxicity, rat	No	No	*Tyl et al 2004 (10)*Ema et al 2003 (11) Nagao et al 2000 (12) Howdeshell et al 2008 (1) Ema et al 1990 (19) Price et al 1990 (26)
Prochloraz	5 ***3,7***	10 13	↑ Nipple retention, rat *Reproductive toxicity, rat*	Maybe	Yes	Christiansen et al (2009)(29) *Cozens et al 1982 (*)*
Epoxicona-zol	***2,3***	23	Rat, 2-gen study, repro	No	Yes	Hellwig & Hildebrand 1992 (*)
Linuron	***0,8-1*** 10 25	 50	*Reproductive toxicity* Developmental, rabbit ↑ Nipple retention, rat	No	No	McKintyre et al 2000 (13)
Vinclozolin	- **5** *4* 4,9	5 10 - 	↑ Nipple retention, rat ↓ AGD, rat 2 gen, reproductive toxicity, rat Reproductive toxicity, rat	Yes	Yes	Hass et al 2007 (14) Hass et al 2007 (14) *Hellwig et al 1994, BASF (*)* Hellwig et al 1990, BASF (*)
Procymi-don	10 12,5 12,5 2,5	25 37,5 125 12,5	↑ Nipple retention, ↓ AGD, rat ↓ AGD, hypospadia, rat ↓ AGD, hypospadia, rat ↓ *AGD*, hypospadia, testis effekt, rotte	No	Yes	Hass et al 2007 (14) Wickramaratne et al 1998 (*) Hoberman et al 1992 (*) *EFSA scientific report 2009*
PCB's Arochlor 1254 Arochlor	- - - -	30 0,05 0,1 0,01	↓ AGD, ↓ Testosterone (↑ AGD, ↑ prostate weight, mice) ↓ AGD, ↓ organ weights, ↓	Yes	Yes	Lilienthal 2006 (15) Gupta 2000 (16) Faqi 1998 (17) Faqi 1998 (17)

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Potency as a decision criterion?

- Potency has a **context** (exposure in risk assessment)
- In isolation, potency-based trigger values are **arbitrary** – not a good basis for developing consensus
- Potency-based cut-offs do not take account of **susceptibility during critical windows** of exposure where potency may be less critical
- Violate the requirement for **consistency** across regulations – not suitable for substances of concern equivalent to CMR

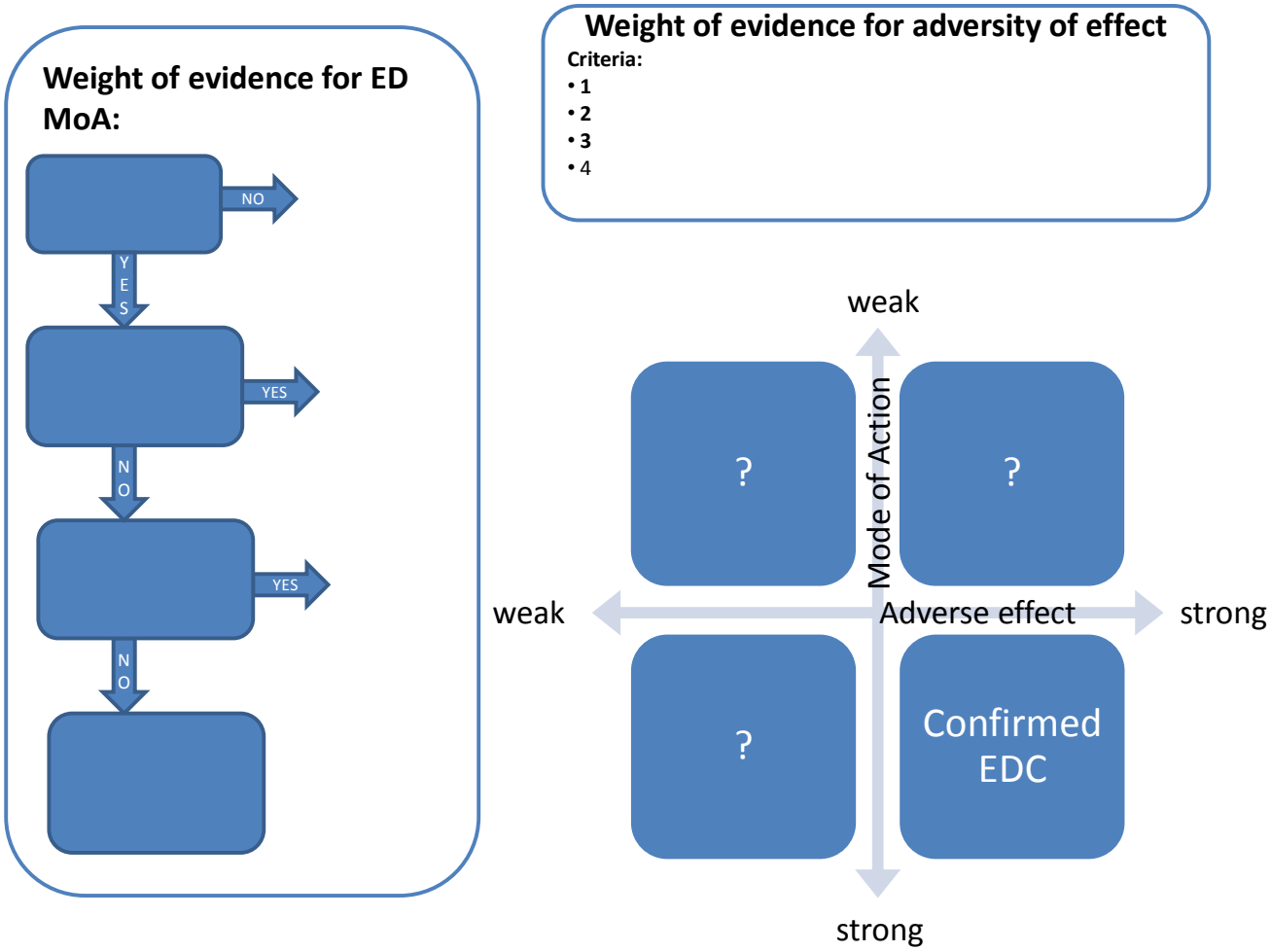
Proposed decision tree

- **Stage 1: Evaluation of evidence for ED properties**
- Adversity
- Mode of action

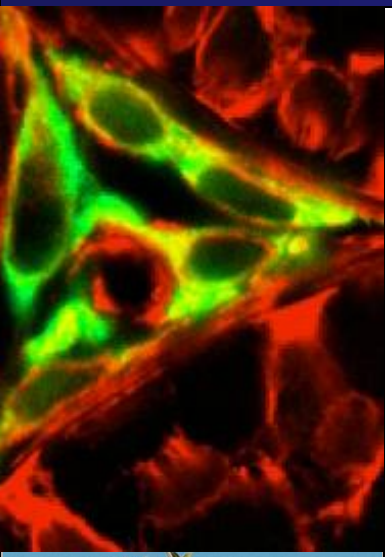
- **Filter**

Proposed decision tree

Adversity and MoA considered in parallel



Proposed decision tree



- **Stage 2: Evaluating human and wildlife relevance**
- Apply **weight of evidence approaches** (*to be worked out*)
- Assume relevance in the absence of appropriate scientific data
- **Filter**

Proposed decision tree

- **Stage 3: Toxicological evaluation**
- Potency
- Lead toxicity
- *Severity*
- Specificity
- *Irreversibility*
- **No criterion decisive**: no substance should leave the decision tree at this stage
- In line with weight of evidence approaches: consider **all the evidence**
- **Do not filter**

Proposed decision tree

- **Stage 4: Final decision, classification and categorisation**

- PPPR: cut-off
- REACH: authorisation required
- **Weight of evidence approaches to be worked out**
- **Case-by-case** decisions necessary

Recommendations

- Implementation of **test methods** as part of information requirements
- Further development of **guidance documents** for the interpretation of test data
- Develop **weight of evidence procedures** for criteria “adversity” and “mode of action” in an inclusive, but not mutually exclusive, way
- **Abandon** potency as a cut-off criterion
- Create regulatory categories that **stimulate the provision of data**

Thank you