An integrated approach to studying in vitro, clinical and epidemiological effects of endocrine disruptor exposure prenatally and in early infancy

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OBesogenic Endocrine disrupting chemicals: Linking prenatal eXposure to the development of obesity later in life

- European Commission FP7 funded research project
- Project duration: May 2009 – December 2013
- 7 institutes in 5 countries (NL, BE, FR, NO, SL)
OBELIX hypothesis

- perinatal exposure to endocrine disrupting chemicals (EDCs) plays a role in the development of obesity later in life

**Early exposure**

- EDCs?
- Change in birth weight?
- Change in early growth and BMI?
- Changes in hormone and lipid metabolism?
- Mechanism: altered DNA methylation?

**Late phenotype**

Change in adipocyte differentiation?
OBELIX approach

Epidemiology

- Mother/child cohorts:
  - Belgium
  - The Netherlands
  - Norway
  - Slovakia

Toxicology

- Animal studies
- In vitro models

Mechanistic analysis

Risk Assessment
OBELIX chemical classes and model compounds

EPI- STUDIES

Brominated flame retardants
- BDE 47

Organochlorine pesticides
- DDE, HCB

EPI- AND ANIMAL STUDIES

Non-dioxin like PCBs
- PCB 153

Perfluorinated compounds
- PFOA

IN VITRO STUDIES

Dioxin-like compounds
- TCDD

Phthalates
- DEHP

ANIMAL STUDIES

Bisphenol A
## Perinatal exposure assessment

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<td>metabolites</td>
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*Number of samples

**Pink:** available before OBELIX  
**Green:** analysed within OBELIX
OBELIX-ENRIIECO cohorts: prenatal PCB 153 exposure

Govarts et al, 2012, EHP

*OBELIX cohorts
Human studies: design

EXPOSURE

PCB
DDE
TEQs
PFOA

PRENATAL

POSTNATAL

PERINATAL

OUTCOME

Birth weight

2 y

Growth 0-2 y
BMI 2 y

7 y

BMI 7 y
leptin
adiponectin
insulin

Samples

courtesy of Nina Iszatt, NIPH
Animal studies: design

Perinatal dietary exposure

Mouse strain: C57BL/6J * FVB

body weight
fat pad weight
histopathology
food consumption
physical activity
serum lipids, insulin
leptin, adiponectin, glucagon
glucose tol. test
internal dose
DNA methylation
PCB 153: postnatal exposure

- Significant decrease in growth 0-24 months*
  - Interquartile increase 183 ng/g lipid: change in z-score of -10
  - approx. 200 g lighter
  - n=2487, 7 cohorts

- No association with BMI at 7 yrs
  - n=2307, 9 cohorts

- Significant negative association with serum leptin at 7 yrs#
  \( \beta = -0.24, 95\% \text{ CI } -0.45, -0.03 \)
  - n=221, 1 cohort
  - BMI also significantly lower in this cohort

PFOA: perinatal exposure

Growth curves F1 after perinatal 300 µg PFOA/kg bw/day

Van Esterik et al., 2015, Archives of Tox, in press
Mechanistic studies in OBELIX

Liana Bastos Sales

Jorke Kamstra

Sylvie Remy
3T3-L1 cell line

Day 0
Pre-adipocyte
Adipogenic cocktail
Change in DNA methylation?

Day 8
Mature Adipocyte

Effects of EDCs on adipocyte differentiation

Bastos Sales et al, 2013

* p<0.05
Effects of EDCs on adipocyte differentiation

Bastos Sales et al, 2013

* p<0.05
Effects of EDCs on global DNA methylation

Bastos Sales et al, 2013

*\(p<0.05\)
Demethylation of Pparg2 promoter by BDE 47

Region analyzed

Pparg2 promoter

Kamstra et al. 2014, Env Sci Technol
OBELIX hypothesis

- perinatal exposure to endocrine disrupting chemicals (EDCs) plays a role in the development of obesity later in life

Perinatal EDC exposure affects growth and metabolic pathways later in life
  - effects may lead to changes in adiposity and/or body weight

  • Evidence for a “heavier” phenotype
    → DDE, dioxin-like compounds, BPA (male mice)

  • Evidence for a “leaner” phenotype
    → PCB 153, PFOA, BPA (female mice)
OBELIX summary

- Current EDC levels pose a risk for metabolic disruption
- Gender specificity
- Divergent effects of pre- and postnatal exposure
- Mechanisms: generated new hypotheses

- Interactions with other risk factors?
- Long term consequences of changes in growth?
- Effects of mixtures?
Future research

- Cohort follow-up in EU DENAMIC project (perinatal exposure and neurodevelopment)

- My lab: development of zebrafish model of obesity
  - Multigenerational epigenetic studies, effects of altered circadian rhythm on adipogenesis

Adipocytes in living 15 d old zebrafish
THANKS TO THE

VU University Amsterdam
Timo Hamers
Thijs van Boxtel
Liana Bastos Sales
Jorke Kamstra
Peter Cenijn
Marja Lamoree
Marijke de Cock
Margot van de Bor

RIVM Netherlands
Leo van der Ven
Martijn Dollé
Joantine van Esterik

Slovak Medical University
Tomas Trnovec
Lubica Palkovicova
Dana Jureckova
Kinga Lancz

VITO Belgium
Greet Schoeters
Karolien Bloemen
Eva Govarts
Patrick de Boever
Britt Wens
Sylvie Remy

INRA – Met@Risk FR
Phillipe Verger
Max Feinberg

Ecobaby NL
Janna Koppe

Norwegian Inst for Public Health
Merete Eggesbo
Nina Iszatt

Scientific Advisors
Matt Longnecker
Rebecca Simmons
Reinhard Stoger
Marc-André Verner

www.theobelixproject.org

NWO

European Commission

THANKS TO THE BELIX TEAM