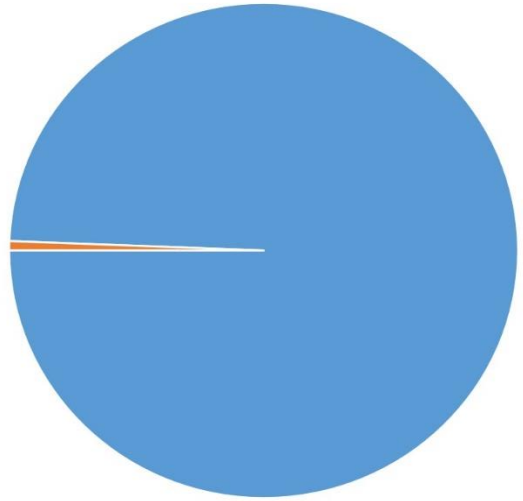
VU VRIJE UNIVERSITEIT AMSTERDAM

BDS BioDetection Systems

Bérénice COLLET

In vitro combined bioassay to assess endocrine disrupting chemical (EDCs) affecting the thyroid system.

Endocrine disruptors: an every-day treat



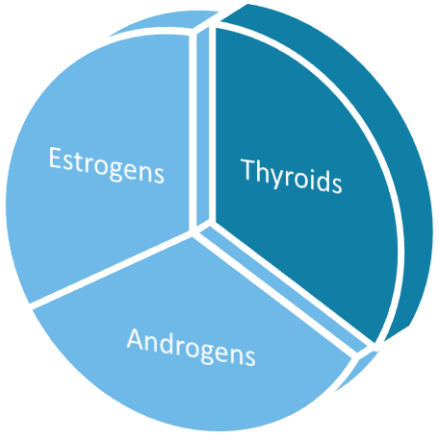
Develop new methods to **quickly** assess the potential disrupting properties of daily-life chemicals

BioDetection Systems

BioDetection Systems

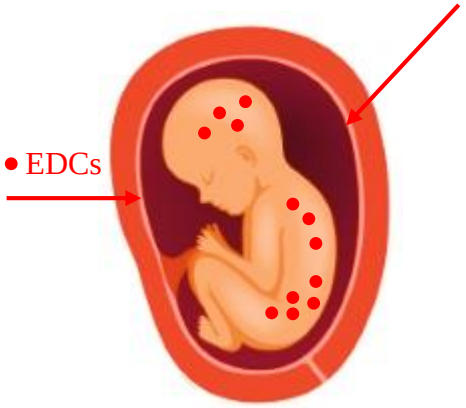
Innovative, sensitive and efficient in vitro test methods to protect the safety and the quality of our food, health and environment

Endocrine disruptors exposure leading to developmental and reproductive defects in newborns



Transthyretin-Thyroid Receptor β (TTR-TRβ) bioassay

Thyroid hormones: key to fetal development

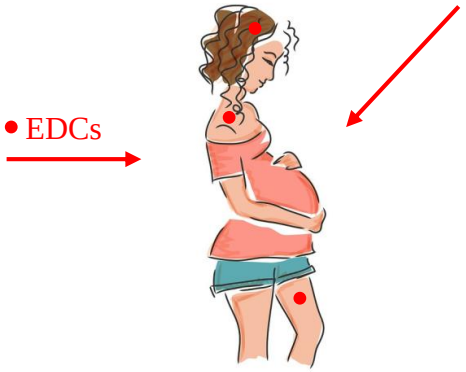


Brain, cardiovascular development

Proper growth

Premature delivery, low birth weight...

Impaired fertility



Fetus is heavily reliant on maternal thyroid hormones

Transporters to circulate in the serum

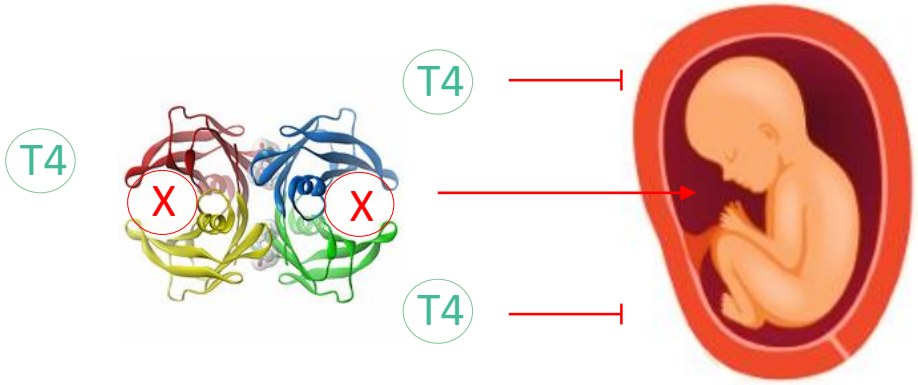
Two thyroid receptors (TR): α and β

Transthyretin (TTR): special delivery to the fetus

Thyroxine (T4) transporter

Found in serum and cerebrospinal fluid

Through the uterine-placenta wall and blood brain barrier (BBB)



↘ Thyroid hormones in the fetus

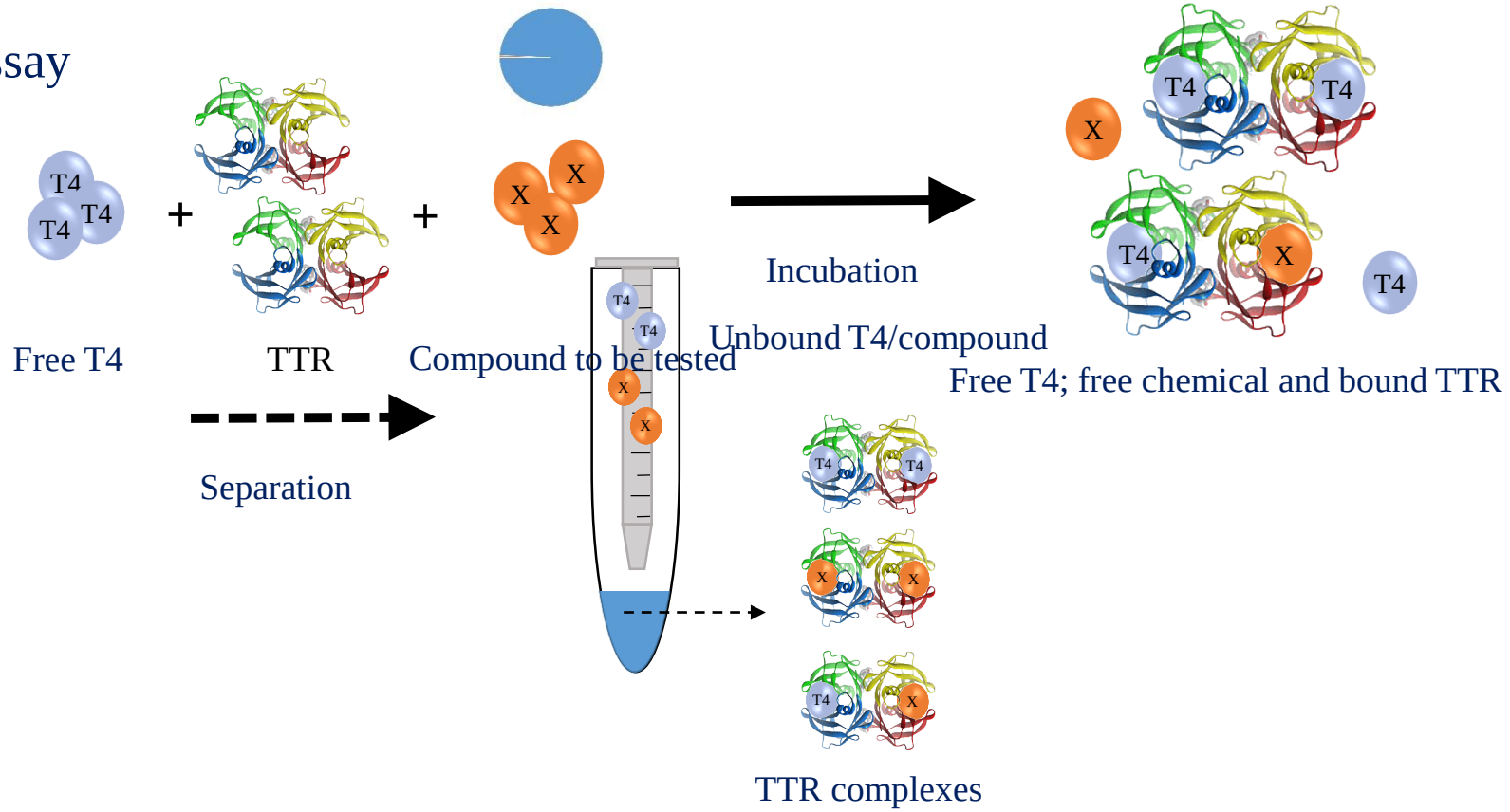
↗ Endocrine active compounds

Agonists

Antagonists

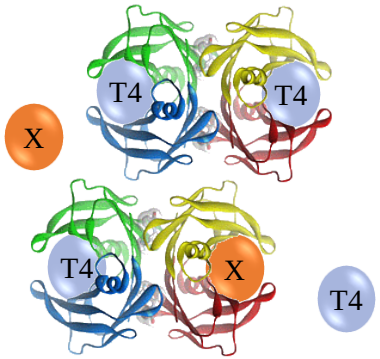
Transthyretin-TRβ bioassay

TTR-binding bioassay



Transthyretin-TRβ bioassay

TRβ-reporter gene bioassay



Separation

Analysis on TRβ CALUX

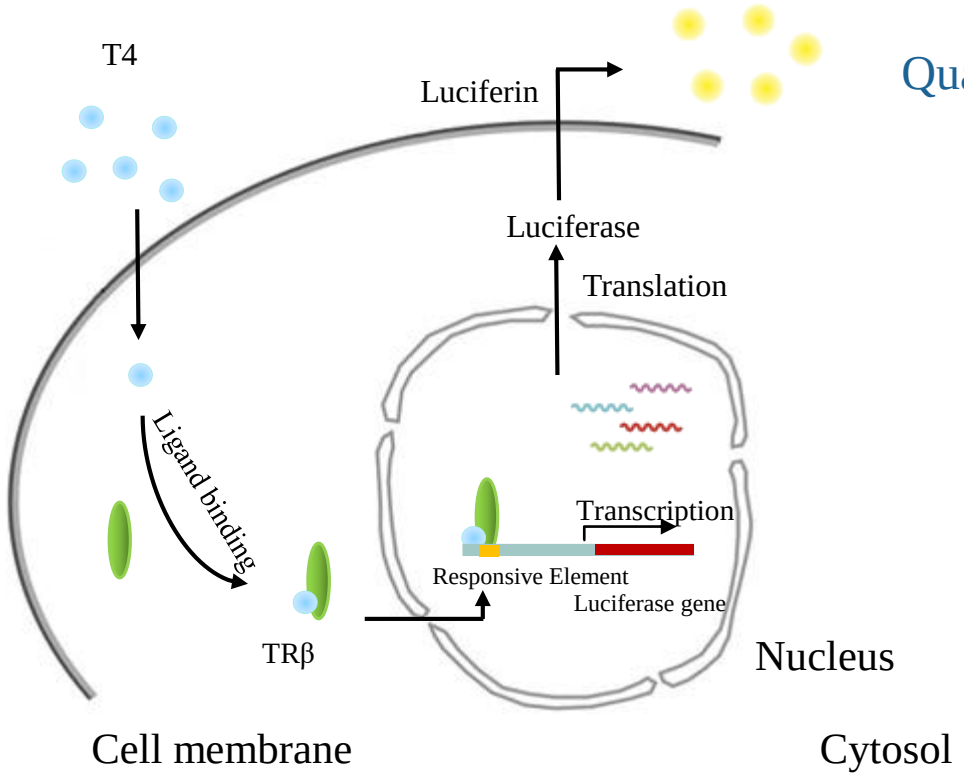
Unbound T4/c

U2OS cells expressing Thyroid Receptor β
Cotransfected with luciferase gene

TTR complexes

Reporter gene assay

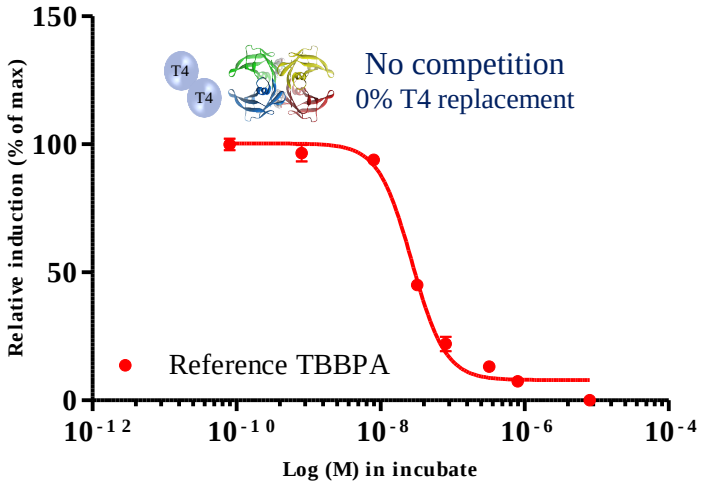
TRβ bioassay

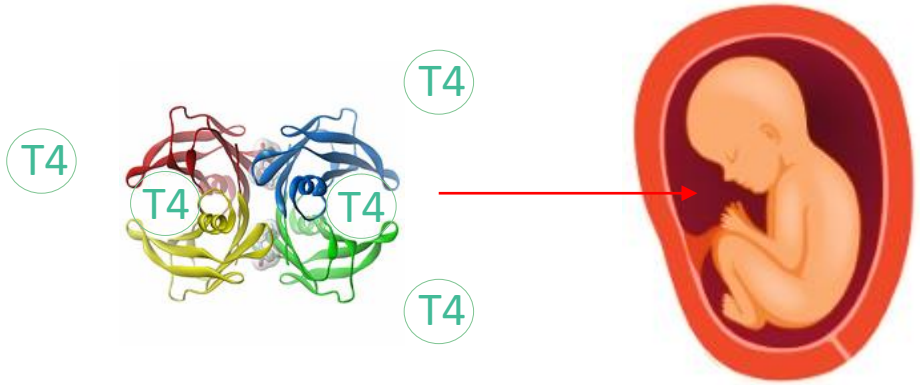


Quantifiable light production

Schematic representation of CALUX reporter assay

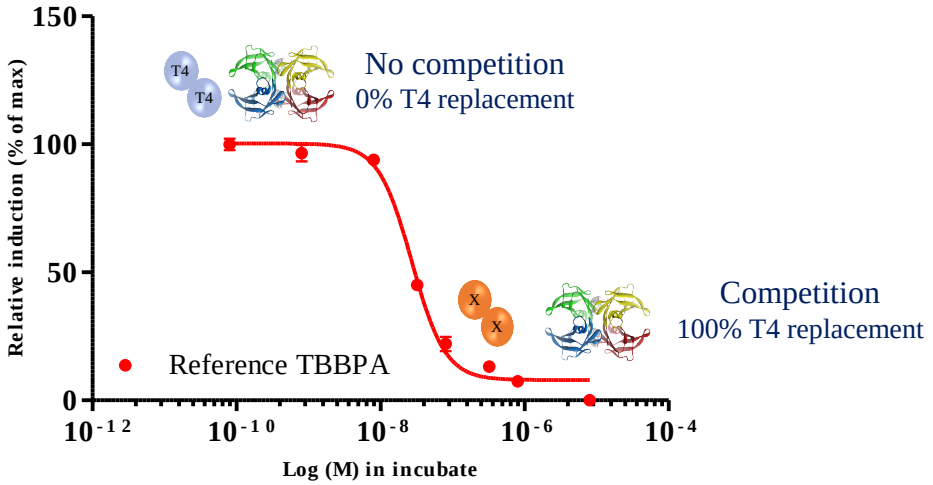
Analysis of the results

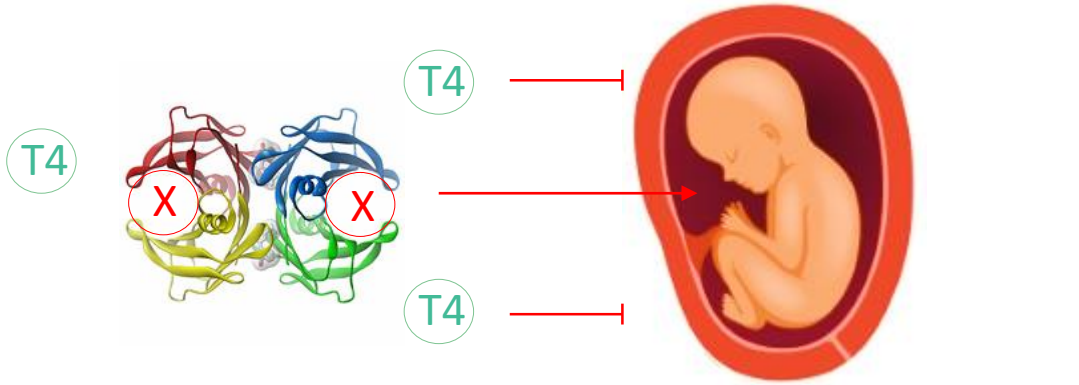




Normal thyroid hormones levels in the fetus

Analysis of the results





↘ Thyroid hormones in the fetus

↗ Endocrine active compound

What is the conclusion for an unknown chemical?

Data analysis: Agonist/Antagonist

1

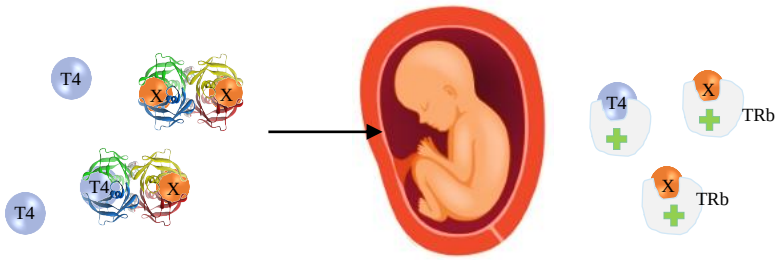
TRβ reporter gene assay: Agonist

2

TRβ reporter gene assay: Antagonist

3

TTR-TRβ assay: Competitor



↘ TRβ activation by T4 (competition)

↗ TRβ activation by compound binding

Data analysis: Agonist/Antagonist

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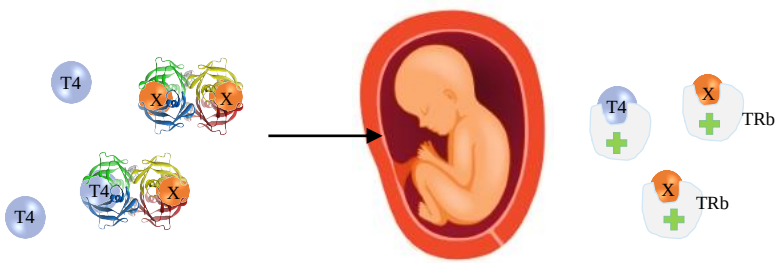
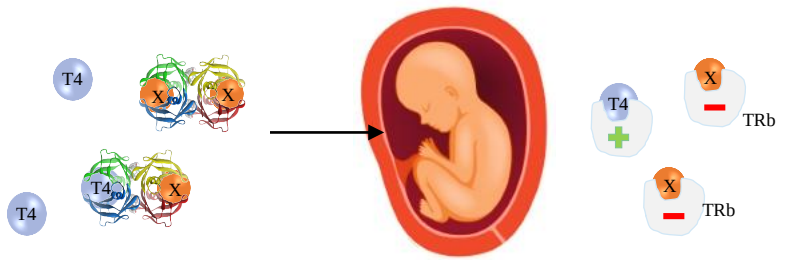




↘ TRβ activation by T4 (competition)

↗ TRβ activation by compound binding

↘↘ TRβ activation by T4 (competition)

Antagonist effect of chemical

Double hit

Validation 30 Compounds Triplicate Different technicians

1. 100% — TRβ reporter gene assay: Agonist
2. 84% — TRβ reporter gene assay: Antagonist
3. 100% — TTR-TRβ assay: Competitor

Competitive Binding of Poly- and Perfluorinated Thyr

Jana M. Weiss,* Patri

*Institute for Environmental Stu

Potential Mechanisms of Thyroid Disruption in Humans: Interaction of Organochlorine Compounds with Thyroid Receptor, Transthyretin, and Thyroid-binding Globulin

ition, and Organismal Biology; [3]Department of

Anti-thyroid hormone activity of bisphenol A, tetrabromobisphenol A and tetrachlorobisphenol A in an improved reporter gene assay

Hong Sun [a,b], Ou-Xi Shen [b], Xin-Ru Wang [b,*]

[a]Jiangsu Provincial Center for Disease Prevention and Control, Nanjing 2

Thyroid Hormone Action Is Disrupted by Bisphenol A as an Antagonist

Potent Competitive Interactions of Some Brominated Flame Retardants and Related Compounds with Human Transthyretin *in Vitro*

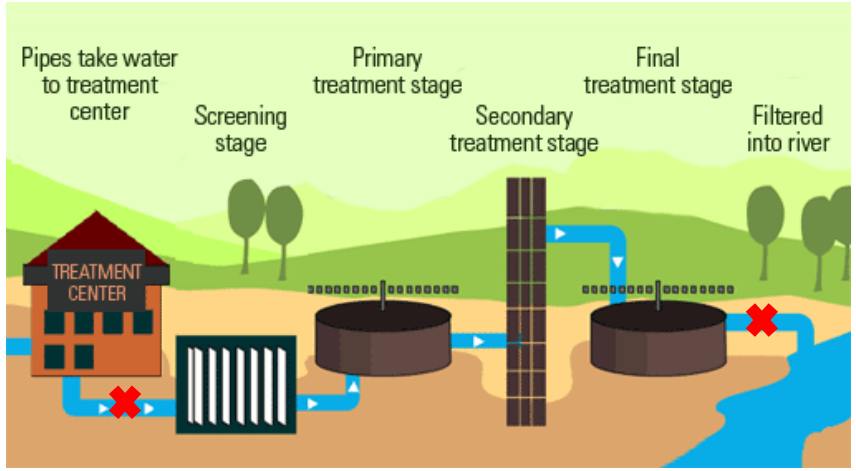
Ilonka A. T. M. Meerts,*[,1] Jelmer J. van Zanden,* Edwin A. C. Luijks,* Ingeborg van Leeuwen-Bol,* Göran Marsh,† Eva Jakobsson,† Åke Bergman,† and Abraham Brouwer*[,‡]

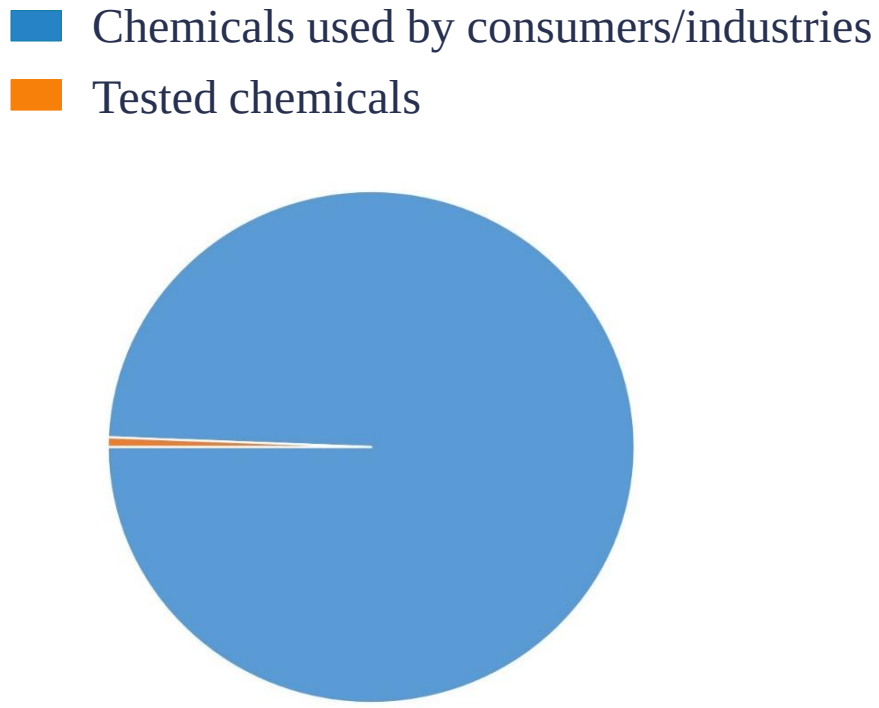
AMI, TAKASHI AKAMIZU, TAKESHI USUI, MISA SAIJO, AYA, AKIRA SHIMATSU, HIDESHI KUZUYA, AND

e, Graduate School of Medicine, Kyoto University (K.M., T.A., M.S., N.K., Y.H., Research Institute, Center for Endocrine and Metabolic Diseases, Kyoto National 612-8555, Japan

Conclusion: the TTR-TRβ bioassay

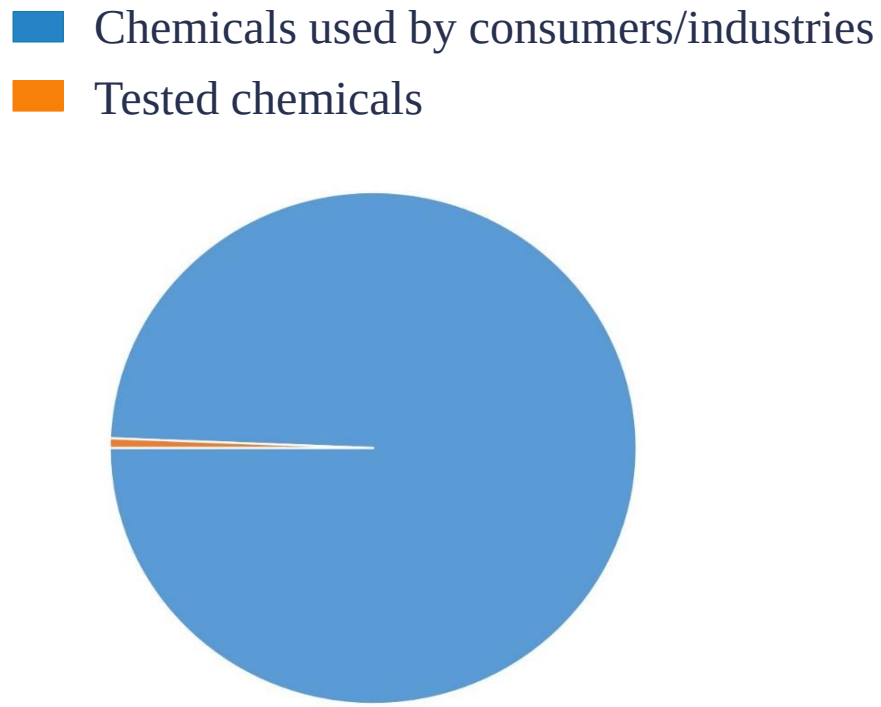
- Good alternative to currently available methods
- Generate data on yet untested chemicals
- Assess everyday life environment





Conclusion: the TTR-TRβ bioassay

- Good alternative to currently available methods
- Generate data on yet untested chemicals
- Assess everyday life environment
- Give better directives to population



Thank you for your attention

This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 722634.

Project coordinated by: Queen's University Belfast
www.qub.ac.uk

For further information:
www.protected.eu.com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Questions

If the chemical is an antagonist, will it influence the TTR-TRb bioassay?

Concentrations tested for the TTR-TRb are lower than concentrations usually required for getting an antagonistic response.

How is it better than another method?

No radioactivity

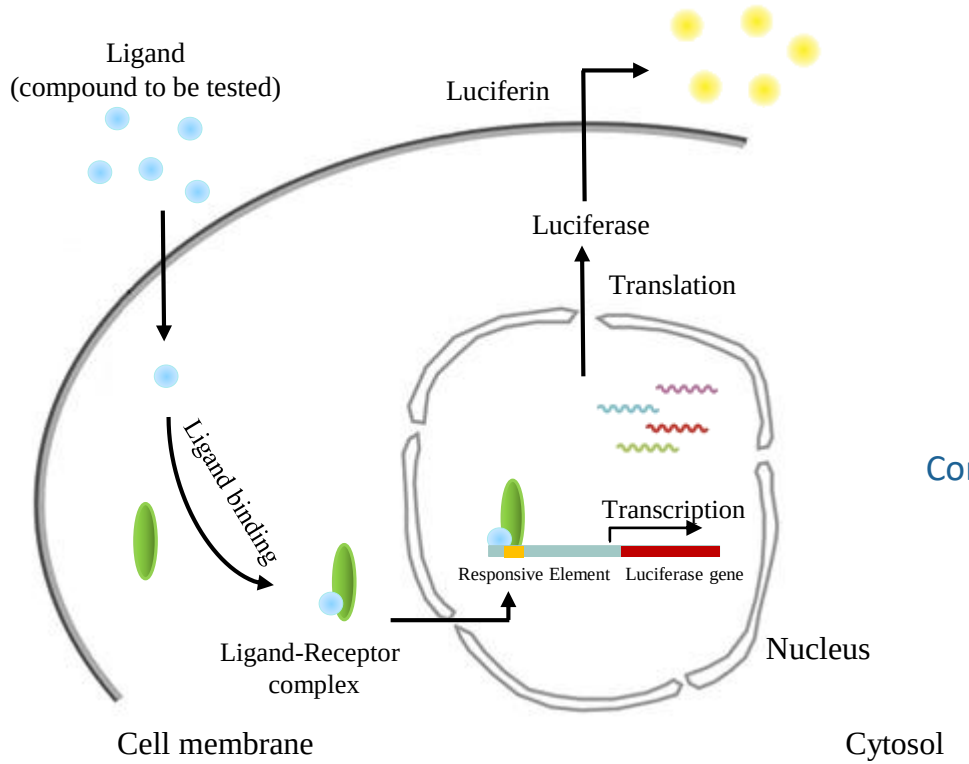
Three different information: agonist, antagonist, T4 competitor.

Can be performed alongside other CALUX bioassays (ER ; AR ; AhR…)

Why TRβ and not TRα?

TRβ is more likely to be involved in human pathophysiological changes if compared with TRα, suggesting a stronger role of this isoform in homeostasis

BioDetection Systems



Quantifiable light production

Compound's effects on receptor activity

Schematic representation of CALUX reporter assay