Thyroid Tox - Development of an *in silico* model to predict thyroid receptor mediated human endocrine disruption

Overall aim

1. The aim of this Challenge is to develop quantitative structure-activity relationship (QSAR) and molecular docking tools that reliably predict thyroid receptor mediated human endocrine disruption. The tool must meet the Organisation for Economic Cooperation and Development (OECD) principles for QSAR validation to demonstrate the statistical and mechanistic reliability of the model for use in a regulatory context.

Duration

2. Up to 18 months.

Budget

3. Up to £100k.

Sponsors

4. Challenge Sponsors define the Challenges in collaboration with the NC3Rs to set out the business case and 3Rs benefits, with a view to using the product developed. Sponsors are required to provide in-kind contributions to help solve the Challenge.

5. The Sponsors for this Challenge are Shell and Syngenta.

Background

6. There is increasing scientific, political, and public interest in the impact of endocrine effects of chemicals on human health and environmental species. Regulatory authorities are continuing to develop improved frameworks and models to predict and assess the effects of endocrine disrupting chemicals (EDCs). For example, the European Chemicals Agency (ECHA) have developed assessment criteria for identifying and classifying chemicals as EDCs that are primarily based on hazard identification [1] and in the US, the Environmental Protection Agency (EPA) are using a risk-based testing approach for EDC identification that is comprised of *in vitro* and *in vivo* tests [2].

7. EDCs may exhibit their toxicity by interacting with endocrine-related receptors such as the oestrogen, androgen, and thyroid receptors [3]. The current of endocrine testing and chemical safety regulations require *in vitro* testing followed by extensive *in vivo* testing in multiple animal species. *In vitro* high throughput screens that elucidate the potential interaction of EDCs with oestrogen receptors have successfully been used for EDC identification [4]. However, *in vitro* thyroid receptor assays are limited or lack full validation [4].

8. Apical endpoints related to endocrine disruption are primarily assessed using *in vivo* reproductive and developmental toxicity studies (e.g. hormone changes and/or histopathological changes are assessed in these tests). These endpoints are used as part of the assessment of potential thyroid receptor-mediated endocrine disruption, but it is not possible to examine the direct interaction or mechanism of action of the test chemical on the two subclasses of thyroid receptor – alpha and beta. Most of the understanding of chemical interactions with the thyroid receptors is based on knowledge derived from the pharmaceutical and agrochemical sectors, and there is limited information on the direct interaction of petrochemicals and high production volume chemicals (chemicals that are manufactured at exceptionally high volumes and are subject to specific regulation [5, 6]) on the thyroid receptor.

9. QSARs are a key tool in the growing field of new approach methodologies (NAMs) that aim to better evaluate potential risks of chemicals on human health and the environment and reduce the reliance on animal models [7]. QSARs have been shown to accurately predict human health toxicities [8] and unlike current *in vivo* studies, can provide information on potential toxicities in a rapid and cost-efficient manner with specific information on the potential binding of chemicals to receptors prior to *in vitro* assays. Molecular docking enables the prediction and/or assessment of the interaction of a substance with a specific receptor. In combination, a QSAR and a molecular docking model will provide specific information on the interaction of a test chemical with the thyroid alpha and beta receptors, addressing an unmet need in endocrine disruption testing.

10. A QSAR and molecular docking model that can accurately predict the interaction and potential binding of petrochemicals and high volume chemicals with the thyroid alpha and beta receptors will:

    - Permit the early identification of potential thyroid-related toxicities in chemical candidate selection without *in vivo* studies.
    - Contribute to the scientific justification, along other NAMs to potentially waive studies and/or reduce animal group size of those taken forward to chemical registration.
    - Improve the predictive capacity of required *in vivo* models.
    - Decrease development cost and time to market for products containing these chemicals.

    The aim of this Challenge is to develop an *in silico* model for predicting endocrine disruption that occurs via the thyroid receptor. It is expected that the Challenge will result in a tool that accurately and reliably predicts the interaction of a chemical with the human thyroid alpha and beta receptors using a combined

QSAR and molecular docking approach.

3Rs benefits

11. If successful, this Challenge will deliver an *in silico* tool to replace animals used for thyroid-related endpoints in the OECD Test Guidelines (TGs) for reproductive and developmental toxicity studies. The four OECD TGs considered most informative for endocrine disruption are:

    - The prenatal developmental toxicity study (OECD TG 414) [9].
    - The reproduction/developmental toxicity screening test and combined repeated dose toxicity study (OECD TG 421/422) [10].
    - The extended one-generation reproductive toxicity study (OECD TG 443) [11].
    - The two-generation reproduction toxicity study (OECD TG 416) [12].

12. Collectively, these four OECD studies utilise more than 2,500 animals per chemical for regulatory registration and may take over 18 months to conduct. The reproductive and developmental toxicity studies are invasive, time-consuming, and expensive to conduct and can cause animal welfare concerns. In addition, these toxicity studies may not identify if the candidate chemical is acting directly at the thyroid alpha and beta receptors. Removal of the thyroid apical endpoints with a detailed mechanistic tool could reduce groups sizes by 10 to 15%, which would reduce the animals used by 250 to 375 per chemical tested per company.

Key deliverables

13. The aim of this Challenge is to develop an *in silico* model that includes: (1) a QSAR model and (2) a molecular docking model, that provides reliable predictions for petrochemicals and high production volume chemicals that potentially lead to endocrine disruption through interactions with the human thyroid alpha and beta receptors.

    Essential

    - Mining of existing public and private data sources for information on thyroid receptors (e.g. ECHA, EPA Toxcast), and the development of a data repository on chemicals that interact with the thyroid receptors. This data repository will be essential for developing a training-set of chemicals for the QSAR and molecular docking models.
    - Clear demonstration of the methodology for the prediction of thyroid alpha and beta receptor interaction.

- Demonstration that the QSAR and molecular docking models can reliably predict the potential endocrine disruption-derived toxicity and interaction with the thyroid receptor of chemicals using a validation set of chemicals with known thyroid effects.
- The models should meet all the OECD principles for QSAR validation [13].
- The models should be delivered as a user-friendly tool.
- The developed tool should be made widely available across all relevant industries and the predictions must be transparent to the user.

Desirable

- Initial dissemination of the use and application of the QSAR and molecular docking tools at scientific meetings.
- Publication of the QSAR and molecular docking tools in a peer-reviewed journal.
- Evidence of the future applicability of the model approach for other non-nuclear hormone receptors involved in endocrine biology.

Sponsor in-kind contributions

14. The Sponsors will provide:
    - Expertise in toxicology and human health QSAR development and QSAR testing.
    - Sharing of data from *in vivo* reproductive and developmental studies (e.g. clinical chemistry data, histopathology data).
15. The NC3Rs will facilitate access to expert input on use of the US EPA ToxCast database [4].

References

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10. OECD (2016) *Test No. 422: Combined Repeated Dose Toxicity Study with the Reproduction/Developmental Toxicity Screening Test, OECD Guidelines for the Testing of Chemicals,* Section 4. doi: 10.1787/9789264264403-en

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13. OECD (2007) Principles for QSAR validation.

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