Questions and Answers from the CRACK IT Challenge 46 FET4Thyroid Challenge Launch Webinar

Questions related to eligibility and the application process are covered in the Guide for Participants.

1. **Is there a need to measure thyroid hormones more accurately or is the requirement for more mechanistic readouts?**

   Measuring thyroid hormone levels is not a must. This might be technically challenging due to the small size of eleutheroembryos and therefore not easily implementable in routine screening tests with these organisms (need to pool large numbers of organisms). The requirement is more about mechanistic readouts using a limited number of organisms per test condition (see existing OECD Test Guidelines (TGs) as examples). The reporter system we are looking for should enable improved mechanistic understanding of thyroid mediated effects.

2. **Are Sponsors interested in sodium-iodide symporter (NIS) expression or function or both?**

   NIS function would be highest priority, but expression can be included. This also applies to other targets within the thyroid system (*e.g.*, thyroperoxidase (TPO)).

3. **It is necessary to use a fluorescence-based readout in the approach?**

   To stay in line with technologies already included in existing OECD TGs and facilitate technology transfer, fluorescence methods are preferred. 'Omics approaches that can also be used to evaluate responses of the endocrine system are not in scope as they require more methodological development.

4. **Will the transgenic reporter be provided or is it the applicants responsibility to ensure availability?**

   The reporters should be provided by the applicant and the applicant should ensure that appropriate licenses are in place that do not restrict their use that align with the OECD policy on intellectual property.

5. **Are Sponsors able to conduct chemical analytics of test chemicals?**

   Sponsors are only able to advise on analytical approaches and methods.

6. **Is the Challenge requiring an assay that is both high-content (addressing all thyroid-related mechanisms, multiple targets) and targeted (fluorescence)?**

   Several thyroid-dependent mechanisms can trigger the response of fluorescence reporters. See OECD TG 248 as an example. The assay(s) should be able to identify mechanisms that are not (consistently) detected in the XETA (*e.g.*, inhibition of NIS and TPO) but also other thyroid-related mechanisms, such thyroid hormone plasmatic transport, interaction with the thyroid hormone receptor, thyroid hormone metabolisms, through fluorescence.

It is encouraged that applicants discuss their proposed approach with the Challenge Sponsors. If you would like to be introduced to the Sponsors to discuss your proposed approach before the submission deadline, please contact the CRACK IT team at crackitenquiries@nc3rs.org.uk