COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

Introduction to the discussion paper for the development of methods for potency estimation

Background

Food Standards Agency requirement for potency estimation

1. The FSA have previously put forward a business case for potency estimation to aid in risk assessment. When responding to food incidents[1], there are regularly chemicals, particularly novel foods and sports/dietary supplements such as selective androgen receptor modulators (SARMs) where certain ingredients have very little, or no, toxicological information. For certain novel ingredients, a lot of which tend to be from plants and have a history of medical use in certain parts of the world, again, there is very little toxicological information and sometimes it is not possible to give any risk advice to our FSA Policy colleagues.

2. The possible toxicological values for the chemical can potentially be estimated by in silico models from chemicals with a similar structure or in the same group. A method or approach which could provide a means of estimating the potency of these chemicals could improve the accuracy of the information and confidence in the risk assessment. An *i*n vitro/in silico approach that can provide information on the relative potencies would provide essential information for toxicity prediction, where information is only available on 1 or 2 compounds from the group. This will allow the FSA to identify the level of risk from a given chemical and give greater confidence in risk assessments that individual compounds can be assessed, not just assuming that all compounds have the same toxicological potency.

3. This will be fundamental in scenarios in risk assessment whereby we have limited to no information available on the toxicity of a chemical.

4. In 2009, The Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment (COT) held a workshop on 21st century toxicology[2]. The workshop addressed the United States (US) National Academy report called Toxicity Testing in the 21st Century: A Vision and a Strategy[3]. The report called for accelerated development and adoption of human cell *in vitro* and *in silico* methods for the prediction of hazards, the determination of mechanistic information, and the integration of data.

[1] https://www.food.gov.uk/business-guidance/food-incidents-product-withdrawals-and-recalls
[2] https://cot.food.gov.uk/cotmtgs/cotmtsem/cotwrkshop11feb09
[3] https://www.nap.edu/catalog/11970/toxicity-testing-in-the-21st-century-a-vision-and-a

5. The National Academy report set out a 10-20 year strategy in which the goal would be to develop and validate toxicological protocols that move away from testing in animals through use of in vitro and computer-based assessments of toxicity and mechanisms. The aim was to enable predictions of human in vivo responses to chemicals in a high-throughput and cost-effective manner, with less use of experimental animals.

6. As we are half way through the vision and strategy (10 years) it would be apt to review the current methodologies available and how they might be applied in case studies as well as applied in risk assessment.

7. We wanted to introduce the subject for the forthcoming COT paper in December which will discuss the development/technologies of the current methodologies for prediction models i.e. potency estimation including the intertwined in vitro/in vivo/in silico and some case studies.

8. Furthermore, this is a foresight into our planned combined workshop with physiologically based pharmacokinetic (PBPK) modelling[4] on 11th March 2020 in which we are going to invite experts/organisations to speak/trial out in-house case studies/have roundtable discussions on the topic.

9. The output of the discussion paper in December and workshop/project would be to discuss an approach/method which could be used to provide a relative potency of Toxic Equivalent Factors (TEFs) for any chemical.

Current methodologies

Chemical Landscape

10. Advances in molecular biology, biotechnology, and other fields are paving the way for major improvements in how scientists evaluate the health risks posed by potentially toxic chemicals. These advances could make toxicity testing quicker, less expensive, and more directly relevant to human exposures. They could also reduce the need for animal testing by substituting more laboratory tests based on human cells.

11. One of the major state-of-the-art methods is called potency estimation via a collective multidisciplinary approach. Potency is a measure of the chemical activity expressed in terms of the amount required to produce an effect of given intensity. Potency estimates can be used to directly compare chemical profiles and prioritize compounds for confirmation studies or employed as input data for prediction modelling and association mapping.

12. The combined advances in discovery and clinical sciences, data science and technology [5] has resulted in toxicity testing reaching a pivotal transformation point

[4] Physiologically based pharmacokinetic (PBPK): a mathematical modelling technique for predicting the absorption, distribution, metabolism and excretion (ADME) of synthetic or natural chemical substances in humans and other animal species. PBPK modelling is used in pharmaceutical research and drug development, and in health risk assessment for cosmetics or general chemicals.

[5] http://www3.weforum.org/docs/WEF__Shaping_the_Future_of_Health_Council_Report.pdf

with the advances in the technology and science sector taking advantage of the 4th industrial revolution (4IR)[6].

Toxicology Testing in the 21st Century (Tox21)

13. The phrase '21st century toxicology' (Tox-21c) (Hartung, 2010) refers to 'the transformation underway in the tools and approaches used to evaluate chemical substances for possible effects on human health'[7]. Tox-21c focuses on toxicity pathways (Bhattacharya et al., 2011) mechanisms, modes of action, and adverse outcome pathways (AOP) (Tollefsen ***et al.,*** 2014) in humans.

14. Another related concept is the 3Rs (Hartung, 2010) which was proposed 50 years ago in the publication of Russell and Burch (1959)[8]:

Replace: Methods which avoid or replace the use of animals
Reduce: Methods which minimise the number of animals used per experiment
Refine: Methods which minimise the number of animals used per experiment

15. The principles of the 3Rs is providing a framework for performing more humane animal research.

16. Several strategies have been proposed to implement Tox-21c. In 2004, the National Toxicology Program (NTP) published its report "A National Toxicology Program for the 21st century", which aims 'to support the evolution of toxicology from a predominantly observational science at the level of disease specific models to a predominantly predictive science focused upon a broad inclusion of target-specific, mechanism-based, biological observations'.[9]

17. In 2007, the National Research Council (NRC) published another report "Toxicity Testing in the 21st Century: a Vision and a Strategy", which proposed using computational methods *i*.e. in silico methods to decrease the number of tested animals, make toxicity testing more relevant to humans by using human cells, and make toxicity testing cheaper and faster.[10] This might also facilitate toxicological assessment of combined exposure to multiple chemicals, which has been an area of increasing interest in recent years.

18. Another one of the goals of the Tox21 collaboration is to establish *in vitro* signatures of in vivo human and rodent toxicity (*i.e.* in vitro to in vivo extrapolation[11]) which include cytotoxicity, cellular pathway assays and computer modelling. Some examples include adverse outcome pathways[12] cardiotoxicity[13], skin sensitisation[14] and organ on a chip (Maschmeyer et al., 2015).

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[6] The Fourth Industrial Revolution (4IR) is the fourth major industrial era since the initial Industrial Revolution of the 18th century. It is characterized by a fusion of technologies that is blurring the lines between the physical, digital and biological spheres, collectively referred to as cyber-physical systems
[7] National Research Council, 2007. Toxicity testing in the 21st century: a vision and a strategy. National Academies Press.
[8] https://www.nc3rs.org.uk/the-3rs
[9] https://ntp.niehs.nih.gov/ntp/about_ntp/ntpvision/ntproadmap_508.pdf
[10] https://www.nap.edu/read/11970/chapter/1#iii
[11] https://ntp.niehs.nih.gov/whatwestudy/niceatm/test-method-evaluations/comptox/ct-ivive/ivive.html
[12] https://ntp.niehs.nih.gov/whatwestudy/niceatm/test-method-evaluations/comptox/ct-aop/aop.html?utm_source=direct&utm_medium=prod&utm_campaign=ntpgolinks&utm_term=niceatm-aop
[13] https://ntp.niehs.nih.gov/whatwestudy/niceatm/test-method-evaluations/cardio/index.html
[14] https://ntp.niehs.nih.gov/whatwestudy/niceatm/test-method-evaluations/immunotoxicity/nonanimal/index.html

In silico

19. In silico toxicity modelling is the use of computational resources (i.e. methods, algorithms, software, data) to organise, analyse, model simulate, visualize or predict toxicity of chemicals (Deeb et al., 2012; Valerio, 2009). Computational methods aim to complement in vitro/in vivo toxicity testing to potentially minimize the need for animal testing, reduce the cost and time of toxicity tests and improve toxicity prediction.

20. However, in silico techniques are used in a wide variety of scenarios within and between industries including, but not limited to, screening, prioritisation, classification and labelling, risk assessment, and product development. As an example, within the pharmaceutical industry, knowledge-based systems and Quantitative Structure Activity Relationships (QSAR)s are used to predict mutagenicity of impurities as part of the International Council for Harmonisation (ICH) Harmonised Guideline M7 scheme[15] (Amberg *et al*., 2018).

21. In the cosmetics industry, in silico techniques are used as part of an ab initio[16] approach to assess the overall impact of a chemical. The assessment typically includes information on mechanisms of action, exposure, and uses case scenarios, as well as the more traditional and accepted use for toxicity prediction (Berggren et al., 2015).

22. Currently, in silico tools are gaining importance in the identification of non-intentionally added substances (NIAS) specifically in food contact materials (FCM). Recent publications describe the use of so-called explorative methods, an untargeted analytical strategy to estimate the concentration and chemical structure of NIAS (Pieker et al., 2018). However, a comprehensive analysis of all compounds found via exploration is not realistic and therefore a risk prioritization is required to identify the compounds that most likely have adverse health effects. As a result, the most promising application of in silico methods is its use in priority setting upon screening of a large number of compounds (Peters et al., 2019).

Integrated Approaches to Testing and Assessment

23. Integrated approaches to testing and assessment (IATAs) provide a means by which all relevant and reliable existing information about a chemical can be used to answer a defined hazard characterization question. Information considered can include toxicity data, exposure routes, use cases, and production volumes. This information is used to characterize outcomes that can inform regulatory decision-making.

24. The drawbacks of traditional toxicity testing approaches using laboratory animals may be overcome by the use of human cell-based, biochemical, and/or computational methods to predict chemical toxicity. Due to the complexity of toxicity mechanisms, data from several methods usually need to be considered in

[15] http://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Multidisciplinary/M7/M7_R1_Addendum_Step_4_31Mar2017.pdf

[16] *Ab initio*: a Latin term meaning "from the beginning"

combination to adequately predict toxic effects. IATAs provide a means by which these data can be considered in combination. When necessary, IATAs can guide generation of new data, preferably using non-animal approaches, to inform regulatory decision-making[17].

Application of the models

25. The models reviewed in the discussion paper in December and at the workshop in March 2020, will include real world scenario case studies which could potentially improve risk assessments of certain (groups of) chemicals at the FSA. They can also be applied across government departments for risk assessment in other chemical scenarios in health and the environment.

26. Ultimately, the in silico toxicity data modelling could be synergised with other projects such as the chemicals life cycle assessment[18].

Conclusions

27. Potency is a measure of the chemical activity expressed in terms of the amount required to produce an effect of given intensity. Potency estimates can be used to directly compare chemical profiles and prioritize compounds for confirmation studies, or employed as input data for prediction modelling and association mapping.

28. In silico toxicology encompasses a wide variety of computational tools, databases for storing data about chemicals, their toxicity, and chemical properties; software for generating molecular descriptors; simulation tools for systems biology and molecular dynamics; modelling methods for toxicity prediction; modelling tools such as statistical packages and software for generating prediction models; expert systems that include pre-built models in web servers or standalone applications for predicting toxicity; and visualization tools.

29. The output of this paper as well as the upcoming paper/workshop/project would be an approach and method which could be used to provide a relative potency of TEF for any chemical using *in* vitro/in vivo and in silico methodologies.

30. Relative potencies or TEFs would enable more informed risk assessments to be undertaken where the potential toxicity can be more accurately estimated from the difference from the chemical for which there are toxicity data. This will be more accurate and would provide more confidence in the level of safety defined by the risk assessment.

Questions for the COT

i) Are there any specific areas that Members think should be/would like covered in the December paper?

[17] https://ntp.niehs.nih.gov/whatwestudy/niceatm/integrated-testing-strategies/index.html
[18] https://ec.europa.eu/health/scientific_committees/environmental_risks/docs/scher_o_155.pdf

ii) Any possible experts/speakers that we may want to consider inviting to the workshop?

iii) Any other comments?

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Abbreviations

AOP	adverse outcome pathways
COT	The Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment
FCM	food contact materials
ICH	International Council for Harmonisation
IATA	Integrated approaches to testing and assessment
NTP	National Toxicology Program
NRC	National Research Council
NIAS	non-intentionally added substances
PBPK	physiologically based pharmacokinetic
SARMS	selective androgen receptor modulators
TEF	toxic equivalent factors
US	United States
QSAR	Quantitative Structure Activity Relationships

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