Dear readers,

During the pandemic, many people have learned that following the rules of social distancing and hygiene, wearing masks, and taking the vaccination reduces their risk of catching and spreading SARS-Cov-2 and their risk of serious illness, but, even together, these measures cannot eliminate a residual risk of catching the virus. This issue's Food for Thought … contribution by Alexandra Maertens and colleagues discusses the uncertainty of toxicological tests and the role probabilistic risk assessment can play in dealing with this uncertainty. They argue that accepting and assessing the residual risk by objective evidence integration can enable better risk management decisions and support a transition of the field to new approach methodologies.

The guidance on Good Cell Culture Practice 2.0 by David Pamies et al. is an update of the original GCCP guidance of 2005, which now includes considerations on more complex cell culture systems such as 3D, microphysiological and stem cell-based models. The updated guidance, which was initially published as a draft for stakeholder discussion in 2020 and has received input from a broad range of experts and organizations, aims to improve the reproducibility and quality of *in vitro* research.

Renxiang Chen and colleagues report on the development of a highly automated high-throughput version of their cell-based genotoxicity assay to screen large numbers of chemicals for their ability to cause DNA damage, which can result in cancer. Their assay measures transcriptomic changes that reflect different cellular responses to genotoxic stress, thus also providing mechanistic information, and proves to be both highly sensitive and specific when challenged with 17 chemicals for which the mechanisms of action are known.

Recognizing that neuroactive chemicals can affect different phases of the response of zebrafish embryos to light, Julia Ortmann et al. have developed an analytical method that captures modulations of the response over all response phases and differentiates between neuroactive chemicals and secondary chemical toxicity. The method is intended for use in the ecotoxicological risk assessment of chemicals. Zebrafish embryos are considered an alternative to adult fish as they are not protected by European Directive 2010/63 until they start feeding independently.

Companies and regulatory programs that aim to reduce and replace animal use need to be able to track their success and account for the resources spent on the endeavor. Sue Marty et al. present considerations made by The Dow Chemical Company in installing animal use metrics for this purpose, starting with the definition of animal use and applicable studies, the establishment of baseline animal use, and metrics for animal savings based on the utility of NAM data. They encourage other organizations to adopt a similar approach.

Aiming to replace the mouse neutralization assay for determining the potency of type E botulinum antitoxins, Eran Diamant and colleagues introduce an *in vitro* cell line-based neutralization assay that measures the toxin's cleaved cellular target protein SNAP-25. They report a high correlation between the *in vivo* and *in vitro* methods across various antitoxin preparations including next-generation equine and rabbit antitoxins intended for therapeutic use.

Hazard and exposure data are lacking for many chemicals that are in use internationally, but testing capacities are limited. Marc Beal and colleagues introduce an approach to predict chemicals of concern that should be prioritized for further testing by filling data gaps in *in vitro* toxicity data from ToxCast using *in silico* methods. Upon comparing more than 1000 predicted bioactivity points of departure with existing *in vivo* toxicity data, they find that their predictions are conservative in 95% of cases and argue that this approach can be used to effectively prioritize chemicals for further assessment.

Marieke Hoonakker et al. optimize the CHO cell clustering test, an alternative to the *in vivo* histamine sensitization test used to determine the safety of acellular pertussis vaccine batches, by identifying a reliable and objective read-out parameter. While the test previously required manual assessment of cell clustering upon exposure to toxin, the nearest neighbor method applied here allows an automated assessment that displays a high sensitivity in detecting different pertussis toxin preparations as well as pertussis toxin spikes added to commercial vaccines.

The Open Letter addressed to two EU Commissioners as well as the European Chemicals Agency (ECHA) Management Board calls upon the Commission to select a new Executive Director of ECHA who combines the qualifications to steer Europe towards leadership in the field of chemicals management and implementation of innovative science including non-animal methodologies.

Four Meeting Reports inform on recent virtual and in-person meetings. The Corners include an introduction to the new Einstein 3R Center based in Berlin, Germany. Please consult the online Events page for information on this year's 3Rs-related webinars and conferences.

With best wishes for a good start into a successful and healthy 2022,

Sonja von Aulock
Editor-in-chief