Improving drug safety assessment

Side effects of drugs are studied extensively. However, the predictive power of in vitro and in vivo tests is often limited. Several projects, presented in a DMD special session, are addressing this problem.

From the simple aspirin to the most sophisticated prescription medicine, all drugs come with side effects. Many are minor, some are just an inconvenience, but a few are serious indeed. Not surprisingly, much effort is being put into safety assessments before a drug (or a chemical) reaches the market. Most pharmaceutical companies are still using animal models to predict the toxicological effects of drugs in humans, particularly as this is required by regulatory agencies. This situation is far from ideal. Apart from the ethical issues involved, rodents and other animals are not always a good model of drug side effects in humans.

Interplay

In vitro tests, especially those based on human derived cell lines or tissue samples, could perform better in this regard. But they present their own problems, finds Rob Stierum, head of applied systems toxicology at the Netherlands Organisation for Applied Scientific Research (TNO). 'One of the problems is the extrapolation of in vitro data to the human situation', he points out. 'In humans there is a multicellular and complex organisation which is affected by the ultimate concentration of the drug in the target organ. If you study cells in tissue culture you only have one cell type to study.' As an example, Stierum names cholestasis, a common side effect in which the bile flow from the liver to the duodenum becomes obstructed. This side effect is an interplay between many different cell types. 'Using only one liver cell line is not very effective', says Stierum. 'You can use liver slices, but still, dose-dependent extrapolation to the human situation is difficult.'

To address this issue, TNO is taking the lead in the development of the ASAT knowledge base: Assuring Safety without Animal Testing. 'We try to combine in vitro toxicological data, in particular toxicogenomics data, with toxicokinetic data', says Stierum. 'We can then test the effect of a drug on a range of different molecules simultaneously, in cells.' However, this approach does not show at which dose level an effect is likely to occur in humans. To address this, scientists derive toxicokinetic data from clinical trials in humans or from toxicokinetic models, taking into account compound properties and properties of on- or off-site targets to predict the blood and target organ concentrations of that drug. 'Using the ASAT knowledge base, combining toxicogenomics with toxicokinetics, we are now starting to see interesting patterns', says Stierum. 'The database allows us, using in vitro and modelling approaches only, to determine which dosage would cause adverse effects in humans.'

'Using only one liver cell line is not very effective'

The ASAT knowledge base started 4 years ago with the collection of toxicogenomics data from in vitro studies. For the past 2 years, kinetic data has also been included. Several pharmaceutical companies, universities and research institutes contribute to the database, including the University of Groningen, Radboud University Nijmegen, Maastricht University, and the National Institute for Public Health and the Environment (RIVM). 'ZonMw has been instrumental in providing support to the development of the database', explains Stierum, 'and we are now trying to attract additional funding from other organisations. Our ambition is to include more compound classes and more toxicological endpoints.'

DILI

In a worst case scenario, drugs can cause serious liver injury, potentially leading to liver failure. This is called drug induced liver injury, or DILI, and is a major health problem with broad implications. 'DILI is still poorly predictable from early drug development studies. It is normally only seen in clinical trials', says Suzanna Huppelschoten, PhD student at the Leiden Academic Centre for Drug

Research (LACDR) of Leiden University. 'Basically the problem is: you can look at the toxicological profile of a certain drug, but if you use an assay that is essentially different from that of another company, you don't actually know anything yet. There is just too much variation between in vitro tests and systems.'

To address these problems the MIP-DILI project was started in early 2012: Mechanism-based Integrated systems for the Prediction of Drug Induced Liver Injury. The project is run by a consortium of 26 participants from the pharmaceutical industry, academic institutions and some small companies, funded by the EU's Innovative Medicines Initiative (IMI) and the European Federation of Pharmaceutical Industries (EFPIA). 'One of the goals is to develop a collection of tests that can predict DILI in vitro, in vivo and also in humans', tells Huppelschoten. 'Another endpoint is an improved panel of in vitro 'best practice assays' for predicting DILI in the human population.'

Translation

Huppelschoten's own research fits nicely within the project, she explains. 'I am using in vitro methods to study the role of innate immune signalling. For this purpose we use the hepatic cell line HepG2. These cells are metabolically not very active, so when a drug is creating toxic metabolites, you may miss certain toxicity in your assay. To overcome this problem we work together with some small companies and the University of Freiburg. We try to make a model of HepG2 cells and to find differences between HepG2 cells and primary human hepatocytes to improve that model. We hope that this will improve translation from HepG2 cells to human liver cells.'

Acceptance

The data generated in this manner can later be added to the MIP-DILI database. The MIP-DILI project will run until the end of this year. However, there is talk of continuing the project and even expanding it or applying for a new project. 'But for now, the database already includes the results of many years of research', says Huppelschoten. 'Still more papers will be published. More companies can add data to the database, now and in the future. And they can extract data whenever they need it. So MIP-DILI will be a beautiful legacy database for future research.'

ASAT and MIP-DILI will not remain the only initiatives, stresses Rob Stierum at TNO. He thinks it's likely that drug safety assessment will be improved in the near future using methods currently developed, some of which are already applied. 'An example is the use of disease network biology and toxicoinformatics for improved early in silico prediction of side effects associated with drug targets', he says, 'which we do at TNO.'

'The database includes many years of research'

He emphasises that the application of these technologies during drug development requires corporate investments. These, and their added value, will always be balanced against the costs and effectiveness of more conventional models, in particular during late preclinical development. Stierum: 'Ultimate acceptance by regulatory agencies will also be key to further promote the development and broader application of new methods.' ●