"We may need the animals themselves as it were on the night; but the machines will do well at rehearsals."
Russell, 1959

Meeting Report

In silico Methods – Computational Alternatives to Animal Testing

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A seminar and interactive workshop on "*In silico* Methods – Computational Alternatives to Animal Testing" was held in Berlin, Germany, organized by Annemarie Lang, Frank Buttgereit and Andrea Volkamer at the Charité-Universitätsmedizin Berlin, on August 17-18, 2017. During the half-day seminar, the variety and applications of *in silico* methods as alternatives to animal testing were presented with room for scientific discussions with experts from academia, industry and the German federal ministry (Fig. 1). Talks on computational systems biology were followed by detailed information on predictive toxicology in order to display the diversity of *in silico* methods and the potential to embrace them in current approaches (Hartung and Hoffmann, 2009; Luechtefeld and Hartung, 2017). The following interactive one-day Design Thinking Workshop was aimed at experts, interested researchers and PhD-students interested in the use of *in silico* as alternative methods to promote the 3Rs (Fig. 2). Forty participants took part in the seminar while the workshop was restricted to sixteen participants.

Seminar Report – Update on current developments from experts

The seminar was opened by **Marlon Schneider** (Bf3R, BfR, Berlin) with an introduction on the 3Rs (Replacement, Reduction and Refinement of animal testing), their beginnings, impact and development over the years as well as the introduction of additional Rs such as Responsibility, Rehabilitation or Remember. Topics and strategies such as the use of "lower" animals, fewer animals, maximizing study outcome, animal sharing and welfare were discussed. Prof. Schneider closed his talk leading over to *in silico* methods including frameworks for high quality science, planning experiments and power analysis over databases, visualization tools and data analysis procedures, prediction models, and quantitative structure-activity relationships. **Susanna Röblitz** (ZIB, FU, Berlin) presented the area of computational systems biology starting with the definition of a model, which differs between experimental, conceptual and mathematical models. Mathematical models are built to formalize human interpretation. In accordance with the quote "*A model should be as simple as possible but not any simpler*" (Albert Einstein), it is important to achieve the right balance between simplicity and accuracy. Ordinary differential equations are used in systems biology, e.g., to describe kinetics. Since biological processes are non-linear, the models often exhibit non-obvious behavior. It is still a major challenge to estimate model parameters and to quantify the uncertainty of model predictions. Using an example (female menstrual cycle based on clinical data), she showed the

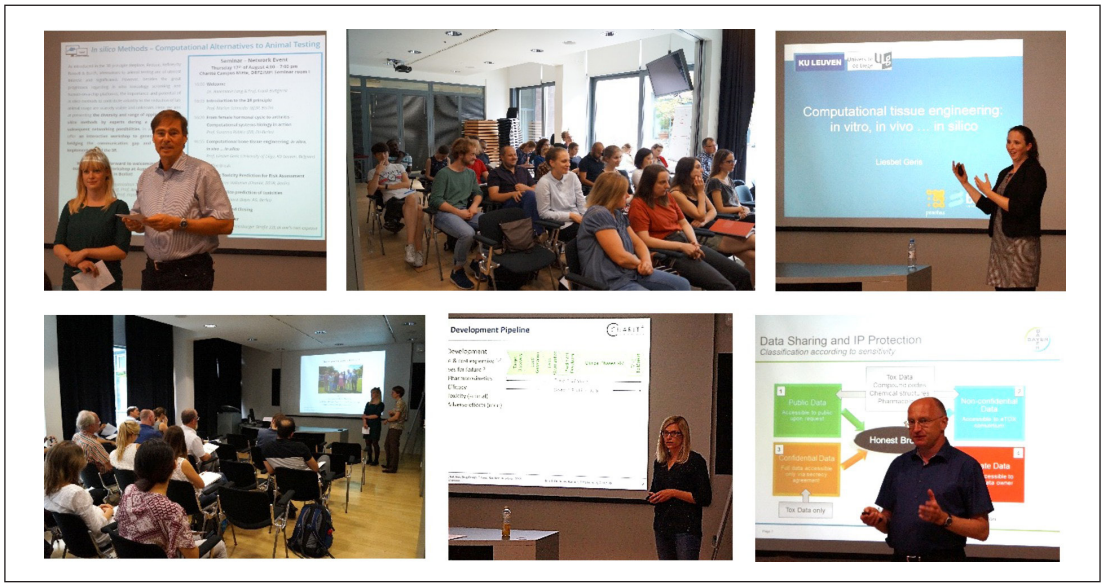

Fig. 1: Impressions of the seminar held in Berlin

power and complexity of these models needed to form virtual patient cohorts (Röblitz et al., 2013). **Liesbet Geris** (University of Liège, KU Leuven, Belgium) showed translational applications of *in silico* methods when combined with different aspects of tissue engineering. She presented the modelling of differentiation processes of mesenchymal stem cells or chondrocytes based on gene regulation by using Boolean models (Kerkhofs and Geris, 2015; Kerkhofs et al., 2016) and explained that prediction models can support the development or validation of new carriers for bone formation (Kerckhofs et al., 2016) or support the design and optimization of cell culture bioreactors (Guyot et al., 2014; Papantoniou et al., 2014). The ongoing work and importance of the Virtual Physiological Human Initiative[1] towards individualized physiology-based computer simulations that aims to revolutionize human medicine was also presented. **Andrea Volkamer** (Charité, BB3R, Berlin) introduced the focus area of predictive toxicology and its relevance for designing new, safe chemicals and drugs. The ever-growing amount of available compound toxicity data is being used to develop reliable methods for risk assessment aiming towards transforming toxicology into a predictive science and reducing animal testing. Beside the explanation of techniques such as read-across, structural alerts and QSAR/machine learning, she focused on molecular fingerprint-based screening that is used to identify structural similarities and ultimately predict toxicity, outlined on the Tox21 challenge[2] data. **Joerg Wichard** (Bayer AG, Berlin) presented the IMI eTOX ("Integrating bioinformatics and chemoinformatics approaches for the development of expert systems allowing the *in silico* prediction of toxicities") project that aimed at collecting high quality data from archives of different pharmaceutical companies within one database for data mining and *in silico* prediction. He pointed out the challenges of running this multi-stakeholder EU project (13 pharma companies + 7 academic institutions + 6 SMEs), including precompetitive data sharing and IP protection as well as the development of a controlled vocabulary and an ontology browser. Some internal examples of where the database/models helped to drive forward developments of a specific drug based on reported findings on similar compounds were shown. A demo database is open to the public and can be visited under http://etoxsys.com/. The seminar closed with a short discussion round and summary.

ReThink3R-Workshop – Implementation of *in silico* methods to promote the 3Rs

Design Thinking is a systematic approach to problem solving. It combines creative with analytical methods from different fields and helps exploit the intrinsic innovative potential of multidisciplinary teams. The Design Thinking process contributes to finding key questions, performing research with a focus on gaining empathy for the users, and designing a serious, realistic solution. Thus, important aspects of the process are interdisciplinary teamwork, "thinking outside the box", a flexible work environment, visualization and empathy.

The workshop, hosted by Annemarie Lang and Laura Behm, included sixteen participants, split into two teams each including experts, postdocs and PhD students. After an interactive warm-up and team check-in, the central challenge was introduced to the participants: "Design a strategy to implement *in silico* methods in basic research to promote the 3Rs". During the following understand-phase, the exploration of the complexity inherent to the challenge, including potential stakeholders/ target groups as well as possible problem areas, played a central role. Aspects that came up during the brain-storming phase were, e.g., a lack of knowledge and awareness of suitable *in silico* alternatives, a bias towards *in vitro* and *in vivo*, a lack of communication, time limitations, the definition of the end-user, reliability and reproduc-

[1] http://www.vph-institute.org/
[2] https://tripod.nih.gov/tox21/challenge/

Fig. 2: Impressions of the ReThink3R workshop

ibility. The observe-phase aimed at getting to know different perspectives and insights by interviewing different stakeholders and sharing the individual interview experience within the team. The participants prepared questions and went out in groups of two to interview people in the labs or on the campus. The synthesizing-phase revealed interesting aspects and problems. In essence, the participants identified i) a general discomfort of researchers to do animal testing, ii) a defensive attitude when the questions came to animal testing, iii) insecurity towards animal protection organizations, iv) a lack of knowledge and trust towards *in silico* methods and alternatives in general, v) limited willingness to talk about own scientific work, and vi) anger about burdens of animal experiment applications including the defense towards the 3Rs. Using different techniques, such as personas and problem statements (defining a user with his/her need in a certain context), the gathered information was synthesized and a concrete underlying problem was specified (define-phase). Within the ideation-phase the participants got to know a number of playful approaches that help to brainstorm in a creative and effective manner. This phase was completed by choosing one of the ideas and transforming it into a prototype. In the breakout groups, two very different innovative prototypes were created: i) a "Science Match" app, enabling scientists from basic research to connect with experts from the *in silico* field based on matching interests and ii) a data analyzer including the opportunity to analyze big data to identify new aspects and hypotheses that were not obvious before and to produce a paper out of the merged data sets. The following testing-phase enabled the participants to test their prototypes with the coaches and observe their reactions. Since Design Thinking is iterative, intermediate solutions are also potential starting points for alternative paths, including redefining the initial problem and going back to former steps such as the observe-phase. A small final presentation of the prototypes was followed by a feedback round concerning the workshop. The overall feedback of the participants was enthusiastic and motivated, although one more day would have been desirable to dig deeper into the context of the challenge and to transform the ideas into more realistic solutions. The workshop was a great experience for experts and young scientists since it allowed cooperation in a novel and creative environment and brought new insights into the field of *in silico* methods.

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