News

EU: Comments on REACH test proposals reduce animal testing

The EU chemicals regulation REACH requires that chemicals on the European market be tested for safety. The required tests depend on the volume of chemical imported or produced per year. Companies may only use animal tests to fulfil data requirements as a last resort and must submit proposals for animal tests to ECHA for approval. The proposals are then made available for public comment for 45 days before a decision is reached.

Toxicologists working with the animal protection organizations European Coalition to End Animal Experiments (ECEAE) and Doctors against Animal Experiments Germany (*Ärzte gegen Tierversuche*) have been analyzing such animal testing proposals and identifying scientific or legal reasons for rejection of individual proposals. By March 2017 they had commented on 540 proposals (35%).

The animal protection organizations recently analyzed the success of the project and report that 50 animal testing proposals were rejected on account of the comments they made. Successful arguments included demonstrating that sufficient toxicological data was already available or pointing out that the test was not legally required as the chemical was only imported or produced in low quantities. In other cases, they supported companies objecting against certain testing requirements and were successful in at least 4 cases. The organizations estimate that their intervention activity has prevented experiments on 60,000 animals in the last five years.

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EU: Validation study for *in vitro* methods to detect thyroid disruptors to start

The Joint Research Centre's European Reference Laboratory – European Centre for the Validation of Alternative Methods (EURL ECVAM) has identified 17 *in vitro* methods as candidates for a validation study that will be carried out in collaboration with the European Union Network of Laboratories for the Validation of Alternative Methods (EU-NETVAL). Methods that perform well could eventually be used in a regulatory context for the identification of endocrine disruptors. The launch of the validation study was announced on July 4 following an expert workshop on setting priorities for further development and validation of test methods for evaluating endocrine disruption held by DG Environment and the JRC in May/June.

Endocrine disruptors are chemicals that interfere with the hormonal system of humans or other organisms. Apart from effects on the action of sex hormones, chemicals can also influence other elements of the hormonal system including the thyroid. The thyroid regulates metabolism and vital body functions including breathing, heart rate, central and peripheral nervous systems, body weight, muscle strength, menstrual cycles, body temperature and cholesterol levels through the action of thyroid hormone. Thyroid disruptors may cause adverse health effects by influencing the function of the thyroid or the action of thyroid hormone.

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EU: REACH Guidance on repeated dose toxicity updated

ECHA has published an update of Section R.7.5 on repeated dose toxicity in Chapter R.7a of the *Guidance on Information Requirements and Chemical Safety Assessment* taking into account revised OECD test guidelines and updated recommendations on the use of non-testing methods. The recommended testing and assessment strategy for repeated dose toxicity has been refined accordingly.

Additionally, Section R.7.3 has also been updated to take into account the recent change in REACH Annex VII for skin sensitization regarding the appropriateness of *in vivo* skin sensitization studies carried out or initiated before the date of entry into force of this revised annex.

Chapter R.7a, Version 6: http://bit.ly/2tVKygj

Adapted from ECHA Weekly
July 19, 2017

GER: Berlin Animal Protection-Research Prize awarded

Two teams share the 4th Berlin prize for animal protection in research:

Dr Philipp Mergenthaler and Dr Harald Stachscheid from the Center for Stroke Research Berlin were recognized for establishing a high-content platform for 2- and 3-dimensional models of the human brain using induced pluripotent stem cells. The platform is intended to investigate causes and possible therapies of stroke instead of performing animal experiments.

A cooperation between the University of Potsdam, the Robert-Koch Institute and the Fraunhofer-Institute for Cell Therapy and Immunology Berlin-Brandenburg mentored by Prof. Frank Bier was recognized for developing the *in vitro* platform "FluType" for the detection of influenza subtypes. This assay can

replace hemagglutination inhibition tests that use blood from infected ferrets. The new method enables faster influenza monitoring, which allows the faster definition of vaccination recommendations without animal experiments.

The prize of 25,000€, sponsored by the State of Berlin and the Association of Research-based Pharmaceutical Companies (vfa), was awarded on October 12 in Berlin.

Adapted from http://www.invitrojobs.com

INT: OECD issues guidance on AOP development, IATA for eye damage and irritation

The Organisation for Economic Co-operation and Development (OECD) recently issued two guidance documents relevant to alternative methods development:

- "Revised Guidance Document on Developing and Assessing Adverse Outcome Pathways (Series on Testing & Assessment No. 184)" is available at http://bit.ly/2hNGWEb.
- "Guidance Document on an Integrated Approach on Testing and Assessment (IATA) for Serious Eye Damage and Eye Irritation (Series on Testing & Assessment No. 263)" is available at http://bit.ly/2ywpldt.

A full list of OECD guidance documents for chemical safety testing is available at http://bit.ly/2yt8si1.

The overall work plan for the Test Guidelines Programme as of August 2017 has been posted online at http://bit.ly/2grtUj5

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INT: OECD releases 3R relevant Test Guidelines

On October 9, the OECD released further new, updated, corrected or deleted Test Guidelines accepted internationally as standard methods for safety testing.

The release contains:

- Test Guidelines that use fewer animals to determine acute inhalation and acute dermal toxicity (TG 433 and TG 402)
- A Test Guideline on *in vitro* methods for skin sensitisation, updated with an additional assay (TG 442E)
- Corrected Test Guidelines for Eye Hazard Potential to include a new reference to a Guidance Document on integrated Approaches to Testing and Assessment (TG 405, TG 437, TG 438, TG 460, TG 491 and TG 492)
- The cancellation of TG 415 on the One-Generation Reproductive Toxicity Study (dating from 1983) because it is no longer used and better alternatives are available to address current regulatory needs.

Adapted from OECD.org

ITA: First course on *in vitro* toxicology in veterinary science master program

The course "Toxicology and *in vitro* models" is a combined theoretical and practical course at the University of Milan offered during the second year of the master degree in Veterinary Biotechnology Science that responds to the need to train students in alternatives to animal experiments. The purpose of the course is to provide tools and information on *in vitro* tests and models for toxicological studies, with particular attention paid to the emerging replacement techniques. The theoretical part is focused on the principles and applications of *in vitro* as well as *in silico* methodologies in different areas of toxicological science, and their future perspectives. The major part of the course is practical work in the laboratory, including *in vitro* cytotoxicity assays, toxicity and permeability assays using an *in vitro* epithelial barrier, *in vitro* models for endocrine disruptor activity, and *in vitro* models for xenobiotic metabolism. The lab activity is environmentally friendly, as material is re-used where possible. The didactic material, including photographic material of the lab activity, is up-loaded to a dedicated on-line platform (http://www.ariel.unimi.it) for the students. A feedback questionnaire showed great appreciation of the course by the students, especially of the extensive practical laboratory activity supporting the theoretical learning.

Thanks to Marco Albonico and Cristina Cortinovis for their support in the organization of the practical activity and to Emilio Benfenati, Lena Buzanska, Thomas Hartung, Helena Kandarova, Silvia Letasiova, Marisa Meloni and Yula Sambuy for their contributions. A special thanks to the students of the 2nd year of Veterinary Biotechnology Science, Università degli Studi di Milano, AA 2015/2016, 2016/2017.

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JPN: JSAAE's International Research Grants accepting applications

The Japanese Society for Alternatives to Animal Experiments (JSAAE) announced that applications are being accepted for the 11th Mandom International Research Grants on Alternative to Animal Experiments. Eligible applicants are researchers around the world, mainly focusing on Asia, who are members of the JSAAE.

Deadline: January 31, 2018
More information:
http://www.asas.or.jp/jsaae/eng/info/11thMandom.html

NLD: FCS-free online platform launched

In vitro research (cell and tissue culture) is generally recognized as a replacement method for animal experiments. However, animals are still indirectly used for this type of research. The serum that is commonly used to grow cells (fetal calf serum, or FCS), is harvested from living bovine fetuses taken from pregnant cows during slaughter. The calves should therefore be regarded as experimental animals. Many scientists are unaware of this fact. On August 2, the 3Rs-Centre Utrecht Life Sciences and Animal Free Research UK launched a new website that allows researchers to identify FCS-free media: fcs-free.org

"As long as people use fetal calf serum, *in vitro* research is not animal free", says Dr. Jan van der Valk, coordinator of the 3Rs-Centre Utrecht Life Sciences (ULS). FCS is a common supplement to animal cell culture media in which cells are grown in the lab (*in vitro*). However, FCS is harvested, with a high chance of suffering, from unborn calves. Furthermore, since it is a natural product, the composition of the commercially available FCS varies from batch to batch, which impedes the reproducibility of results. These moral and scientific concerns demonstrate the urgency to switch to an FCS-free medium.

Identifying and using serum-free media not only replaces animals for research, but also improves the scientific quality of *in vitro* methods. The website fcs-free.org allows scientists to identify FCS-free media for specific cell types. Furthermore, the website serves as a platform to exchange information on the quality and applicability of each product. The FCS-free database provides an overview of commercially available serum-free media for cell and tissue culture, as well as medium compositions obtained from scientific literature. The database is offered by the 3Rs-Centre Utrecht Life Sciences (ULS) in collaboration with Animal Free Research UK.

The FCS-free database is part of the 3Rs database program, initiated by the 3Rs-Centre ULS to make 3Rs information (on Replacement, Reduction and Refinement of animal experiments) available for free. The program does not receive any subsidies and its future relies on donations and gifts. In order to guarantee a sustainable future for the websites and increase their impact, the 3Rs database program is inviting partners who are willing to support its activities. The 3Rs-Centre ULS is part of the faculty of Veterinary Medicine at Utrecht University.

FCS-free platform: https://fcs-free.org
3Rs database program: https://www.uu.nl/en/3rsdatabases

Recent related publication:
van der Valk, J., Bieback, K., Buta, C. et al. (2017). Fetal Bovine Serum (FBS): Past – present – future. *ALTEX*, in press. doi:10.14573/altex.1705101

Adapted from University of Utrecht press release August 2, 2017

NOR: Norecopa bestows 3Rs award

The 2017 Norecopa prize for outstanding efforts to advance the 3Rs in connection with animal research was awarded to Adam Lillicrap, research manager at the Norwegian Institute for Water Research (NIVA) on June 7.

Dr Lillicrap was the independent advisor for the validation management group for the Fish Embryo Toxicity (FET) test guideline and has been actively promoting the use the FET test as a replacement/refinement to acute fish toxicity testing through his involvement within numerous regulatory frameworks. He was also responsible for the revision of the OECD fish bioaccumulation test incorporating a minimized approach to reduce the number of animals from ca.110 to 20 fish/test. Adam Lillicrap is the project leader for the new draft ISO standard for an *in vitro* cytotoxicity assay (submitted in 2016).

Norecopa places special emphasis on advances in research and development which benefit Norwegian conditions. The prize, which consists of NOK 30,000 and a diploma, has been awarded annually since 2010.

Adapted from Norecopa Newsletter 3-2017

NOR: PREPARE guidelines for planning animal research and testing

A number of guidelines and checklists for reporting animal experiments have been produced over the years. Despite this, scientists still experience problems with reproducibility of animal experiments, and translatability of the results to humans. This is undoubtedly due in part to the fact that there are many other factors, in addition to those reported in the scientific papers, which can influence the outcome of experiments. These factors can also affect the animals' welfare, and the health and safety of all those taking part (both animals and humans).

The Norwegian 3R centre Norecopa (https://norecopa.no) has taken the initiative to produce a set of planning guidelines for scientists and facilities, called PREPARE (*Planning Research and Experimental Procedures on Animals: Recommendations for Excellence*). PREPARE covers all stages of quality assurance from the earliest stages of planning an animal experiment.

The PREPARE guidelines are divided into 15 topics which cover everything from facility management to the individual procedures which make up an experiment. A 2-page checklist is provided to help scientists through this process. This checklist has, so far, been translated into 13 languages. The guidelines are linked to a comprehensive website with hundreds of links to specific guidelines on each topic, so that those needing more detailed information can access this. These links include working party reports, literature references and other guidance. The PREPARE website also includes suggestions for a contract between the animal facility and the research group, to help the di-

vision of duties and costs between these. This is important to avoid situations where scientists are unable to publish their results because they lack data which should have been recorded during the experiments. Such failures are a waste of human resources, money and animal lives.

PREPARE is designed to complement reporting guidelines such as ARRIVE. The PREPARE guidelines should be used by scientists from day 1 when planning animal experiments. They have been co-authored by Norwegian and British experts with long experience both in animal facility management and in helping scientists to plan experiments.

More information is available at https://norecopa.no/PREPARE.

Adrian Smith
Secretary to Norecopa

SUI: ALTEX Prize goes to Uwe Marx

The 2017 ALTEX Prize for the best paper published in ALTEX in 2016 goes to Uwe Marx. The t[4] workshop report, "Biology-inspired microphysiological system approaches to solve the prediction dilemma of substance testing", a collaborative effort by a total of 36 authors, was published in ALTEX 3/16, doi:10.14573/altex.1603161.

The Prize consists of a CHF 2,000 personal prize sponsored by the Doerenkamp Zbinden Foundation. It was presented to a colleague of Dr Marx at the Tenth World Congress on Alternatives and Animal Use in the Life Sciences in Seattle, WA by Mardas Daneshian, President of ALTEX Edition.

The winner was elected by the Board, Editorial Board and Editorial Office of ALTEX out of all main articles published in 2016; articles including members of the ALTEX Board and Editorial Office as first or last authors were excluded.

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UK: NC3Rs calls for applications for international 3Rs prize

Each year the NC3Rs awards a prize to highlight an outstanding original contribution to scientific and technological advances in the 3Rs, published in the last three years. The award consists of a £28k prize grant and £2k personal award. Highly-commended entries receive a £4k grant and a £1k personal award.

The 3Rs prize is for a piece of primary research published in a peer-reviewed journal in the last three years and is open to any researcher, in academia or industry. The prize is awarded to the principal investigator, research team leader, or other nominated author. The competition is open to international groups. Applications are assessed by a dedicated panel, which also selects the winner. Selection is based on the quality of the published research and its impact on the 3Rs.

Deadline: December 4, 2017, 4pm (GMT).
More information: http://bit.ly/2gRrzuB

USA: NIEHS offers grants for *in vitro* systems modeling animal experiments

The National Institute of Environmental Health Sciences (NIEHS) is providing grants for development of novel *in vitro* systems using cells from experimental animal models typically used for toxicology testing. The intent is that these systems will replicate biological responses within the corresponding animal tissues or organs. When developed and validated, these systems will provide information needed to predict toxicity of chemical and drug candidates, enable comparisons with existing *in vivo* animal toxicity data, serve as newer assays for toxicology testing, and have the potential for reducing the numbers of animals used in toxicology testing.

These grants of up to $150,000 for Phase I awards and up to $1 million for Phase II awards are only available to U.S. small businesses.

Deadline: January 12, 2018
More information: http://bit.ly/2yTIg38