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Institute for Health and Consumer Protection (Ispra)
EURL ECVAM

EURL ECVAM Workshop "Inaugural Meeting of EU-NETVAL Members" 26-27 June 2014

All documents and presentations related to this meeting are made available to all EU-NETVAL members through the CIRCA website https://circabc.europa.eu
Go to: interest group/EU-NETVAL/ Library/ Inaugural meeting

Participants: Representatives of EU-NETVAL, EURL ECVAM, DG ENTR, DG ENV, and OECD (see CIRCA website for full participants list).

Sandra Coecke (SCO, chair) and Maurice Whelan (MWH, Unit head of the Systems Toxicology Unit (STU) welcomed the participants.

Maurice Whelan gave an introduction to EURL ECVAM, which is an integral part of the STU that gets its legal mandate from the Directive 2010/63/EU on the protection of animals used for scientific purposes.

EURL ECVAM works together with many different networks such as:

- PARERE (Preliminary Assessment of Regulatory Relevance), the network of regulators to get early advice about the relevance of *in vitro* methods and the impact on the 3Rs principles,
- ESTAF (ECVAM Stakeholder Forum), the platform representing non regulatory stakeholders such as Academia, Industry, etc.
- ESAC (ECVAM Scientific Advisory Committee), giving independent scientific advice about the validation studies.
- ICATM (International Cooperation on Alternative Test Methods), a partnership between validation bodies worldwide.

Important deliverables of EURL ECVAM are the strategy documents, outlining ECVAMs views on specific topics and ECVAM recommendations on specific *in vitro* methods. Currently two such strategy documents exist. One for the 'Replacement of Animal Testing for Skin Sensitisation Hazard Identification and Classification' and a second on 'how to avoid and reduce animal use for assessing chemicals for genotoxicity'.

For successful validation an *in vitro* method must be: well defined in the standard operating procedure, transferable to other laboratories, give reproducible results within and between laboratories and have a good predictive capacity of the hazards exerted by chemicals. On a case by case basis, depending on the information already available, it is decided which information gaps must be filled to complete the validation scheme.

Susanna Louhimies of DG ENV gave an overview of the legislative anchor. The objective of Directive 2010/63/EU is to increase animal welfare and promote the use of alternatives in academia and industry. Article 47 is the corner stone for the 3Rs.

Member States must, according to the Directive, appoint a national contact point and contribute to the development and validation of alternative approaches. This led to the establishment of EU-NETVAL, the network of validation laboratories, with currently 26 test facilities from 11 member states. Its terms of reference were presented in detail by Sandra Coecke and can be found on the following website: http://ihcp.jrc.ec.europa.eu/our_labs/eurl-ecvam/eu-netval/EU-NETVAL-tor-november-2013.pdf. Several EU-NETVAL members had agreements with their government to get financial support for validation studies, others had to prepare project proposals and apply for funding on a case by case basis. The reported initiatives were considered good examples of how member states can contribute to the development and validation of alternative approaches, in line with article 47 of Directive 2010/63/EU.

Valerie Zuang from EURL ECVAM presented the validation workflow in detail and announced that upcoming EURL ECVAM strategy documents will be in the field of aquatic toxicity, acute systemic toxicity, toxicokinetics and developmental neurotoxicity.

The presentation by Natalie Delrue from OECD focussed on in vitro methods and the procedures to develop test guidelines and adverse outcome pathways. The working group of the test guidelines programme (WNT) is responsible for the development and update of new and existing test guidelines and consists of national coordinators of OECD member countries, non-OECD member countries, industry and animal welfare organisations. Proposals for new methods or fields for collaboration must be provided in standard project submission forms (SPSFs). Specific methods are being followed by expert groups. E.g. VMG-Non Animal is dealing with endocrine disruptor methods. After validation by an organisation or validation body such as EURL ECVAM, the expert group drafts the technical guidance (TG). The WNT comments, updates the TG and gives its final approval. Both EURL ECVAM and OECD put a lot of effort into the development of Performance Based Test Guidelines (PBTGs) and Performance Standards (PS). EU-NETVAL members were encouraged to seek contact with the member state national contact points and contact points in the PARERE network (published by DG ENV on their website; http://ec.europa.eu/environment/chemicals/lab_animals/home_en.htm), as there is a need to reinforce expert networks.

EU-NETVAL test facilities were invited to present themselves. There is a wide variety of expertise available in the network, with competence in biomedical devices, vaccines, mixtures, toxicokinetics, analytical methods, omics, metabolomics, nanotoxicology, ecotoxicology etc.

A few examples were given of EURL ECVAMs activities. One of the priorities for EURL ECVAM is the development of alternative methods in the field of toxicokinetics, as it is important for systemic toxicity and is a building block for *in vitro* testing strategies. The Cytochrome P450 induction multi-study validation trial, looking at induction of 3 cytochrome P450 enzymes in two human-derived metabolically competent hepatic *in vitro* test systems, was presented by Sandra Coecke. This project is being followed by the project on human hepatic metabolic clearance/stability (see presentation by Varvara Gouliarmou), with the ultimate aim to develop EURL ECVAM standards. EU-NETVAL input on the standards to be developed for phase II metabolism, was welcomed.

The first multi study validation trial that will be carried out with EU-NETVAL test facilities, concerns an Androgen Receptor Transactivation Assay (ARTA) for the detection of compounds with (anti)androgenic potential using AR-CALUX® cells. Sandra Coecke reported that eleven test facilities responded to the invitation to participate in the validation project and provided the

requested information. Six test facilities met the requirements and were evaluated against the selection criteria. Until the collaboration agreement is signed by the three facilities with the highest ranking, no further information can be provided. Besides the planning of the validation, an SPSF has been sent by EURL ECVAM to OECD to propose a performance based test guideline for androgen receptor transactivation, based on validation of the AR-CALUX assay and the Human Androgen Receptor Mediated Reporter Gene Assay using AR-EcoScreen™ cells (EcoScreen assay). Anne Milcamps, the validation coordinator of the AR-CALUX multi-study validation trial, presented the principle of the AR-CALUX *in vitro* method and the validation procedures. After theoretical and GLP compliant experimental assessment, an optimised SOP will be made available to the selected EU-NETVAL facilities for training and subsequent transfer to their laboratories. Only trained staff may perform the work for the validation studies.
A Validation Management Group will follow the whole process, before, during and after experimental phase.

One of the eligibility criteria of an EU-NETVAL facility is to have a quality system, preferably GLP. Maurits-Jan Prinz from DG Enterprise explained that the OECD council act on the Mutual Acceptance of Data, containing the OECD test guidelines and GLP principles as annexes, is implemented in the EU through Directives 2004/9/EU and 2004/10/EU. GLP ensures quality and validity of the data. For medical devices, requirements differ around the world. FDA asks for GLP, the EU not. It will be controlled by Maurits-Jan Prinz if the updated Regulation, which is in preparation, includes GLP requirements. It became clear during the meeting that not all monitoring authorities are interpreting the GLP guidelines in the exact same way. On the DG ENTR website there is a Q&A document clarifying several questions that frequently lead to debate http://ec.europa.eu/enterprise/sectors/chemicals/specific-chemicals/index_en.htm.

As explained by Sandra Coecke, the development of a technical Guidance on the implementation of in vitro methods within a GLP environment to support regulatory human safety assessment of chemicals (GIVIMP)' is an identified area for collaboration with EU-NETVAL. This guidance should be complementary to the OECD GLP principles and guidance document 34 and the ECVAM guidance for good cell culture practices (GCCP) and provides guidance for handling test items, test system, experimental design, setting acceptance criteria, good reporting standards, equipment requirements etc. The aim is that the relevant information is delivered to the regulatory authorities that must interpret the data. From the EU GLP working group several members have already stated they want to collaborate. Also EU-NETVAL members were encouraged to become involved in the drafting of GIVIMP guidance. All members will be invited to review the guidance once it is developed. During this meeting, the participants were divided into two breakout groups to discuss several topics that could be included in GIVIMP guidance. The presentations from the rapporteurs are placed on the CIRCA website.

Follow up actions:

Action 1: EURL-ECVAM gives the EU-NETVAL participants access to all meeting documentation through the CIRCABC website.
Action 2: EU NETVAL members were asked to express their interest for collaboration and contribution to the metabolic clearance project and GIVIMP guidance.
Action 3: Maurits-Jan Prinz will verify if the draft revision of the Regulation that is covering medical devices is including GLP requirements.

Next meeting is planned for the year 2015, as one EU-NETVAL meeting is foreseen per year.