

Session S405

Meeting at the Falls: ICATM Partner Updates

Update on recent activities at JaCVAM

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Center for Biological Safety and Research

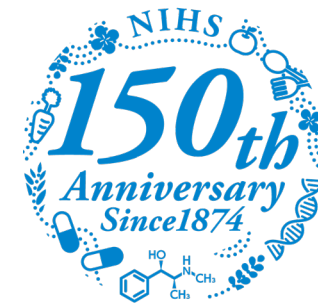
National Institute of Health Sciences

Japan





National Institute of Health Sciences (NIHS)



Enhancing safety and quality of life through scientific research



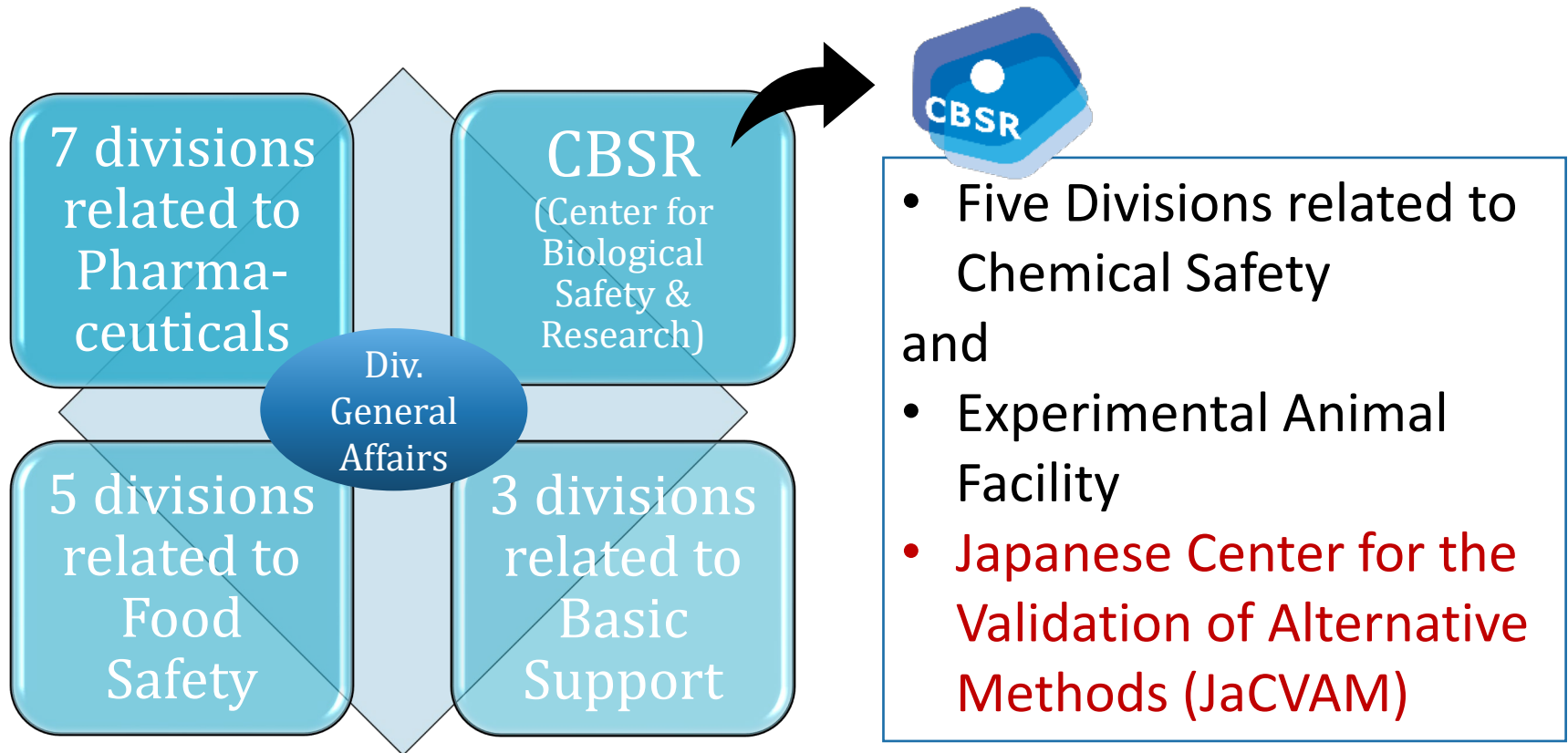
I have no financial relationships to disclose.

This presentation neither intend to represent, nor is restricted by, the policy of MHLW, Japan

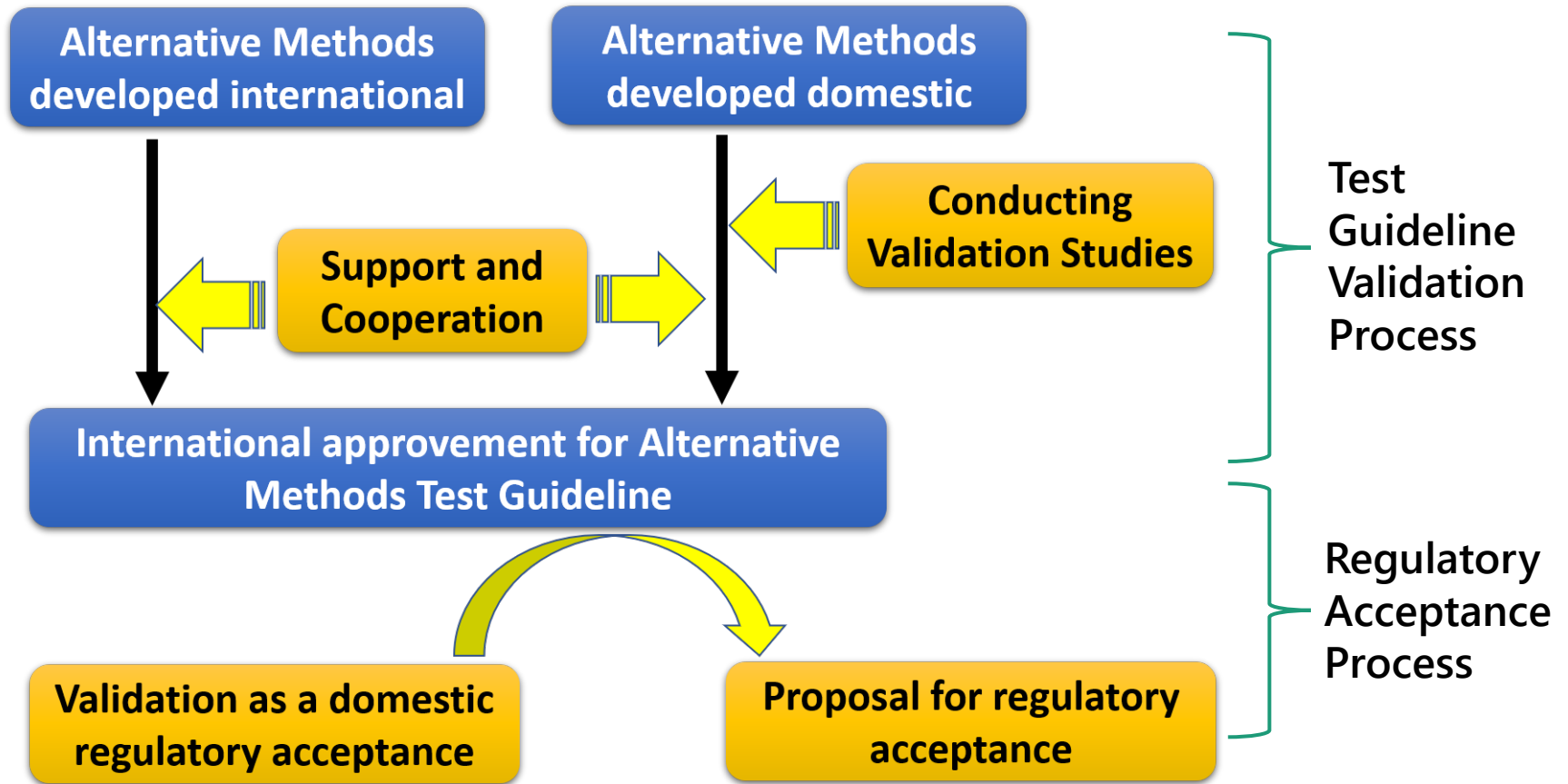




NIHS: Organization



Flowchart of Activities



Number of Test Guidelines on human health that do not use animal testing in OECD (2022)

OECD: <http://www.oecd.org/env/ehs/testing/oecdguidelinesforhetestingofchemicals.htm>

Subject of evaluation	TOTAL	<i>in vitro / in chemico</i>
Skin corrosiveness	3	3
Skin irritation	2	1
Phototoxicity	3	3
Eye Irritation	10	9
Skin sensitization	8	4
Percutaneous absorption	2	1
Genotoxicity	13	5
Endocrine Disruption	6	4
Other	28	0
SUM	75	30

Test Guidelines on human health that do not use animal testing in OECD (2022)

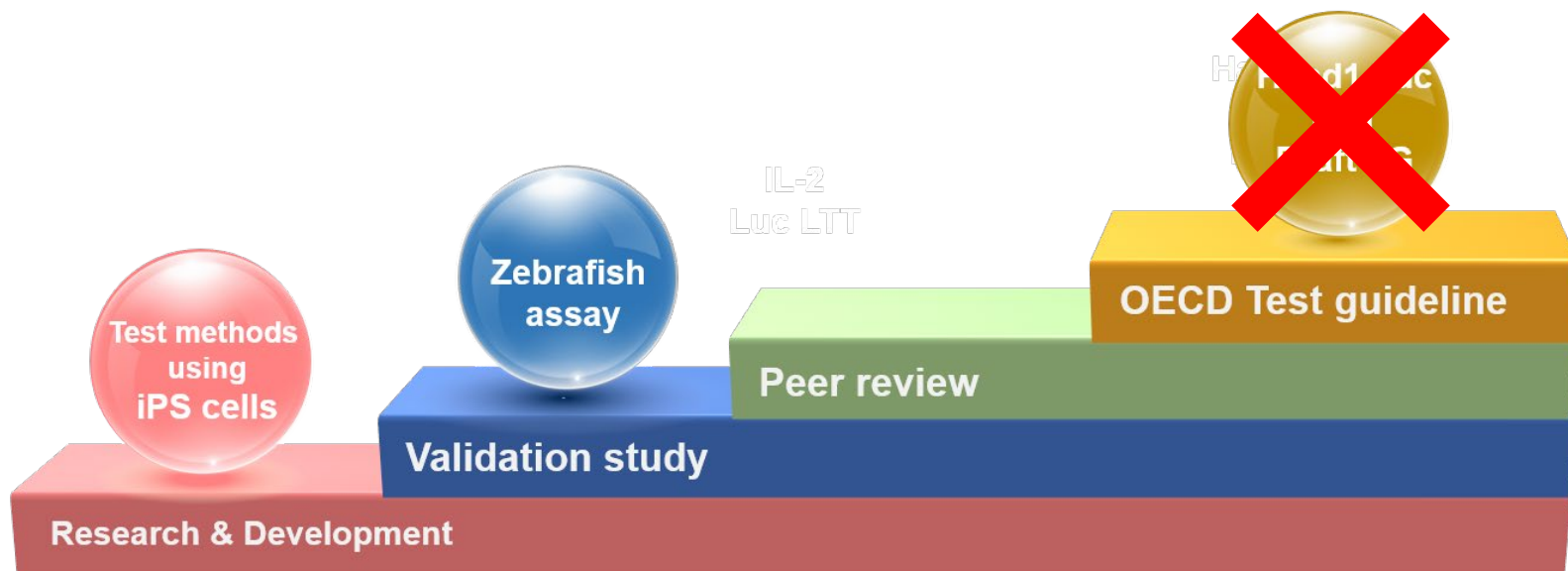
Subject of evaluation	ID No. of Test Guideline
Skin corrosiveness	430, 431 (incl. 5 RhE models), 435
Skin irritation	439 (incl. 6 RhE models)
Phototoxicity	432, 495, 498
Eye Irritation	437, 438, 460, 467, 491, 492 (incl. 4 ChE models), 492b, 494, 496
Skin sensitization	442C (ADRA, DPRA, kDPRA), 442D, 442E (h-CLAT, U-SENS, IL-8 Luc assay, GARD™Skin) , 497
Percutaneous absorption	428
Genotoxicity	471, 473, 476, 487, 490
Endocrine Disruption	455 (incl. 3 test methods), 456, 458, 493 (incl. 2 test methods)

Toward the development of NAM...

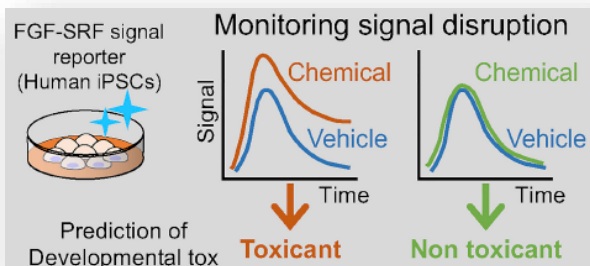
Test methods under development with JaCVAM: a challenge with a particular focus on general toxicity

1. Reproductive and Developmental Toxicity Tests
 - Test Method using human iPS cells
 - Test Method using Zebrafish fertilized eggs
2. Immunotoxicity Test
 - Standardize IL-2 Luc LTT and IL-1 Luc assay, and develop IATA (Integrated Approaches to Testing and Assessment)

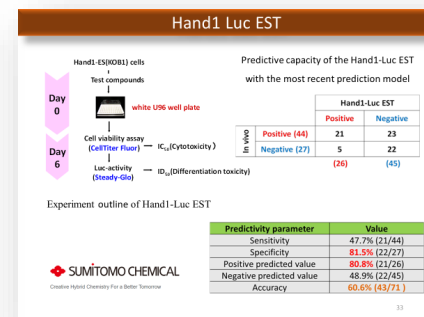
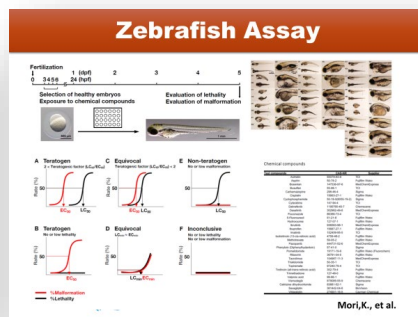
Reproductive Toxicity Tests



iSDT (integrated Signal Disruption Test)



Kanno S, Okubo Y et al: *iScience*, 2022



ICH guidelines specify the use of alternative test methods



INTERNATIONAL COUNCIL FOR HARMONISATION OF TECHNICAL REQUIREMENTS FOR PHARMACEUTICALS FOR HUMAN USE

ICH HARMONISED GUIDELINE

DETECTION OF REPRODUCTIVE AND DEVELOPMENTAL TOXICITY FOR HUMAN PHARMACEUTICALS

S5(R3)

Final version
Adopted on 18 February 2020

ICH S5(R3) Guideline

4.2.2. Alternative Approaches for Addressing EFD Risk

4.2.2.1. Use of Alternative Assays

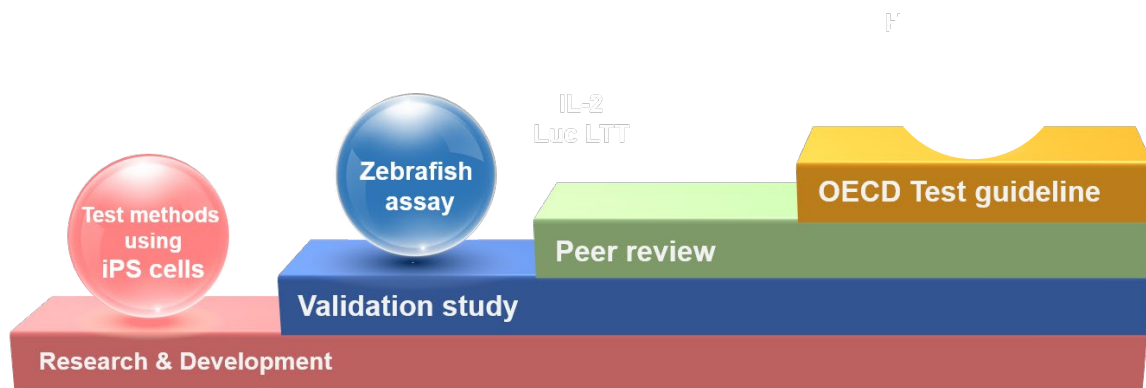
A number of alternative *in vitro*, *ex vivo*, and non-mammalian *in vivo* assays (alternative assays) have been developed to detect potential hazards to embryo-fetal development. They have been used as drug discovery screens for adverse effects on EFD and have assisted in the understanding of the mechanism of toxicity, which can be useful for translating nonclinical data to human risk (especially for human-specific targets). The continued use of alternative assays for these purposes is encouraged.

If properly qualified, alternative assays have the potential to defer or replace (in certain circumstances) conventional *in vivo* studies. This has the added benefit of potentially reducing animal use. Concepts to consider when qualifying these assays, and examples when the use of such assays could be appropriate, appear in Annex 2. Approaches that incorporate alternative assays should provide a level of confidence for human safety assurance at least equivalent to that provided by the current testing paradigms described above. Based on the direction of scientific development as of the writing of this document, it is expected that for regulatory purposes multiple alternative assays will be used within a tiered or battery approach. These testing strategies will be qualified within a certain context of use, which is defined by the chemical applicability domain of the assay, and by characterization of the biological mechanisms covered by the assay.

EFD: embryo fetal development

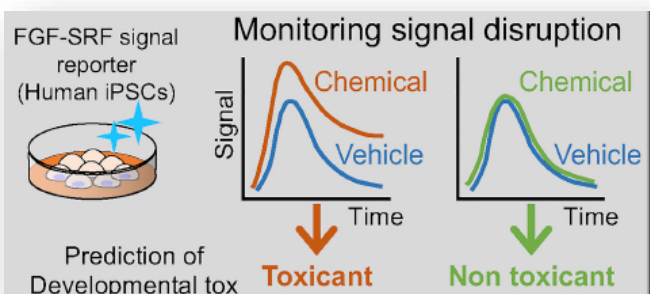


Reproductive Toxicity Tests

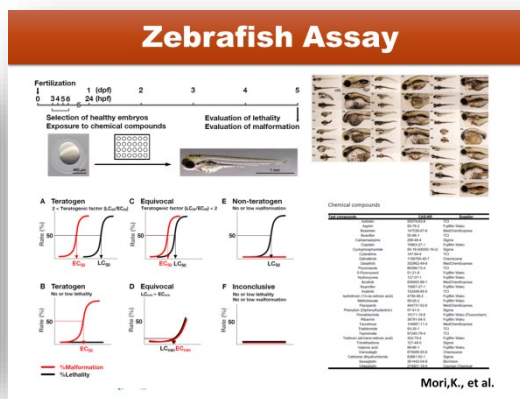


Immediate Goals: inclusion in ICH S5

iSDT (integrated Signal Disruption Test)

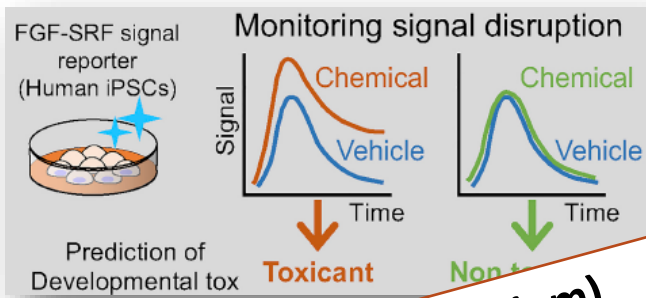


Kanno S, Okubo Y et al: *iScience*, 2022



Attempting to develop a new test method using human iPS cells

iSDT (integrated Signal Disruption Test)



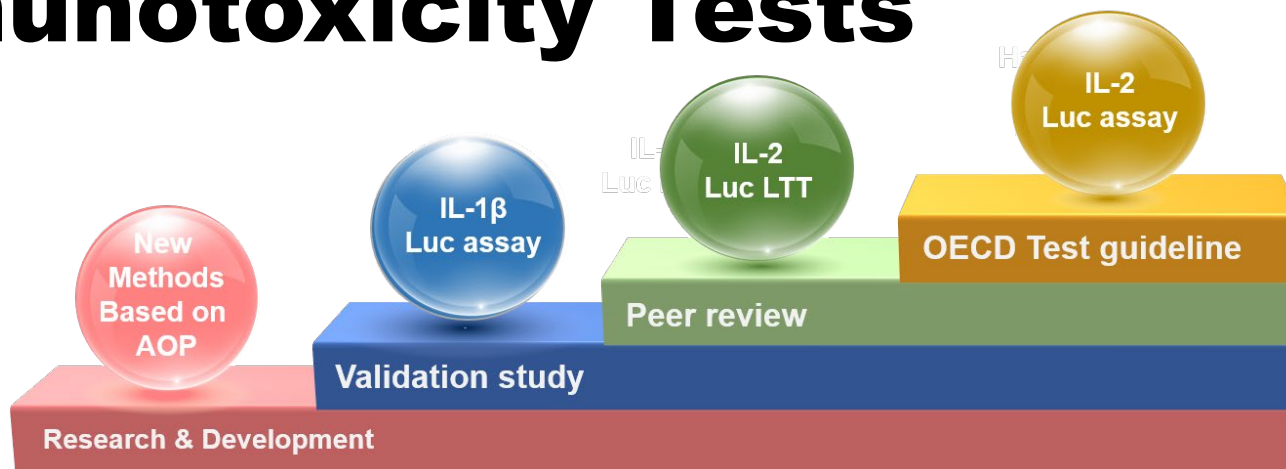
Kanno S, Okubo Y et al

Session S450 (Symposium)

New Approach Methods for Developmental and Reproductive Toxicity Testing

**#137 Developmental toxicity Test using human iPS cells
based on signal disruptions induced by chemical substances
OKUBO, Yusuke (NIHS)**

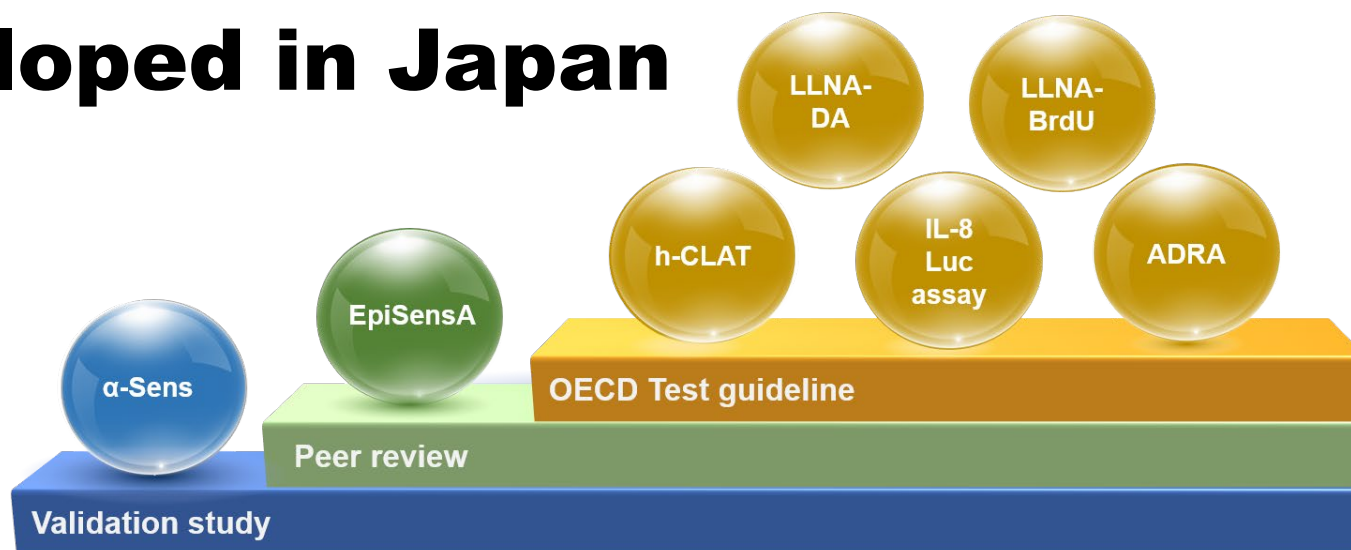
Immunotoxicity Tests



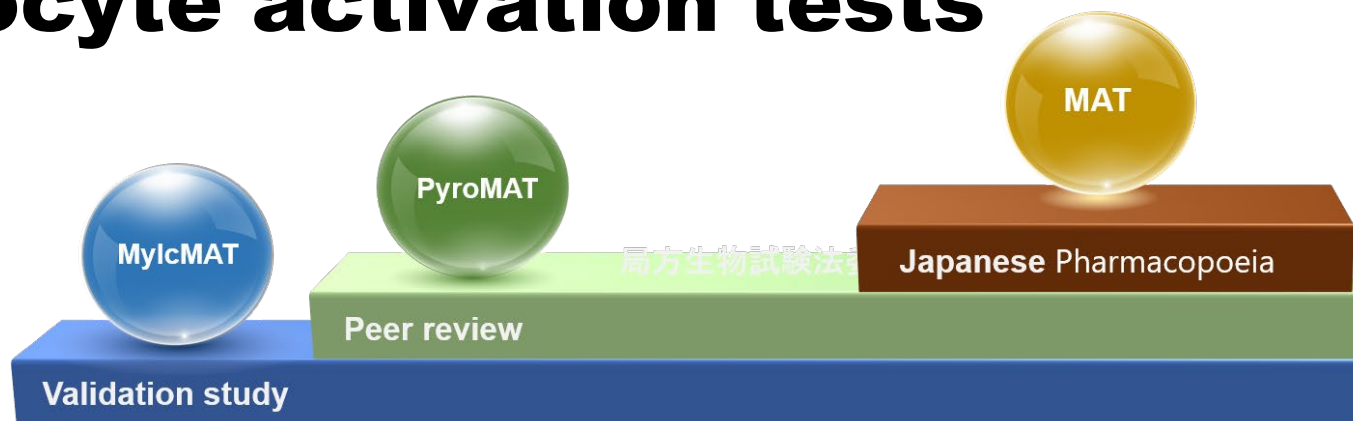
Development phase of alternative immunotoxicity testing methods

1. Developed three AOPs for immunotoxicity
2. Created DRPs for immunotoxicity testing, primarily for test systems to detect immunosuppression
3. Develop test methods based on AOPs
4. Validation studies of the developed assays are conducted and the assays that make up the Multi-ImmunoTox Assay (MITA) are converted to TGs.
5. Finally, an IATA was developed using TG as the core, combined with other information.

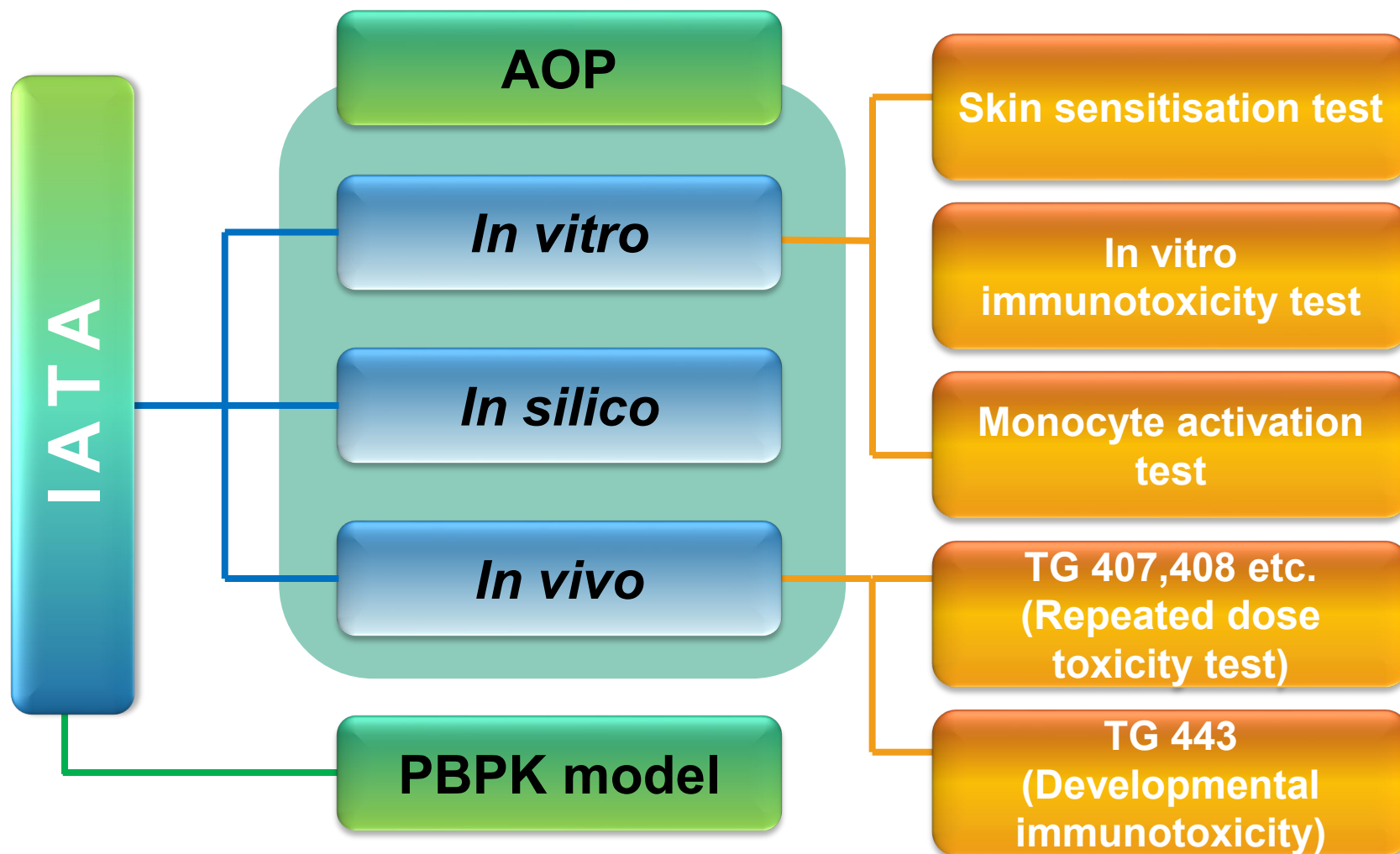
Skin sensitization tests developed in Japan



Monocyte activation tests



Future Plan: Development of IATA for Immunotoxicity Tests



IMPACT

INNOVATIVE EXPLORATORY RESEARCH

RESEARCH INSIGHTS

PROFESSOR TAKESHI YAMADA Fukuoka Prefectural University of Health Sciences	PROFESSOR MASASHIRO GOTO Aizu University	PROFESSOR WOEICHHYN CHU National Tsinghua University	PROFESSOR JAE-GOOK B Seoul National University
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Introductory article of JaCVAM

Impact Objectives

- Contribute to efficient drug development and improved experimental animal welfare
- Explore opportunities where non-animal test methods can quickly evaluate whether new chemical substances are harmful to human health and all life on earth

Innovations to eliminate animal testing

The JaCVAM is leading the way to develop non-animal testing methods in immunotoxicity and reproductive toxicity

Alternatives to animal testing

Director of the Center for Biological Safety and Research (CBSR), National Institute of Health Sciences (NIHS), Dr Yoko Hirabayashi envisions a world in which the use of non-animal methods can replace animal testing. She talks about her interrelated roles and important research

Could you first introduce the important work of the Japanese Center for the Validation of Alternative Methods (JaCVAM) to replace, reduce or refine (the '3Rs') animal testing?

Working hand-in-hand with related international organisations, JaCVAM has contributed to the development of new alternative methods to animal testing, promoted the adoption of these new methods by regulatory agencies and also worked to further the development by Japanese researchers of the novel test methods. It has also contributed to the international standardisation of validation studies and the adoption of more than 15 test methods for the Organisation for Economic Co-operation and Development (OECD) Test Guidelines and International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) guidelines.

How are your roles linked to the 3Rs of animal welfare?

I am the Director of the CBSR, which was established in 1978 as a research facility to investigate the safety of chemical substances and their effects on living organisms, particularly from a neutral standpoint. Its mission is to research and test the safety of substances which are subject to our studies, such as chemical substances, food and medicinal products using biological resources (such as experimental animals and cells) and conduct comprehensive safety

methods based on scientific grounds. The JaCVAM was organised within the CBSR by the National Institute of Health Sciences (NIHS) to promote the use of alternatives to animal testing for regulatory studies. I am the Chair of the JaCVAM Steering Committee. This Committee determines which novel or revised test methods are to be selected and assessed by JaCVAM. It also allocates both the financial and human resources necessary to undertake the scientific validation and assessment of such test methods based on the spirit of the 3Rs of animal welfare, as well as to respond to international trends on how to conduct animal experiments appropriately.

We act as a contact point for international organisations supported by the Ministry of Health, Labor and Welfare (MHLW). We are proud of the Japanese researchers who have done these things and would like to take this opportunity to thank the Japanese developers and collaborators who have contributed.

The ultimate goal of our research is to realise a society in which Artificial Intelligence (AI) can quickly evaluate whether numerous new chemical substances that will be developed in the future may harm people's health and all life on earth.

You are also the representative of the Japanese research group working on ICH-related issues funded by the Japan Agency for Medical Research and Development (AMED). What is the purpose of this work?

The aims are to provide scientific evidence to develop or revise guidelines on safety evaluation and/or quality control of new drug registration in the ICH, and to achieve worldwide harmonisation of processes based on the scientific consensus among academy, regulatory and industry experts. The outcomes in this study are also expected to contribute to prompt and efficient drug development and to experimental animal welfare; the so-called 3Rs. ▶

testing, more costly and complex non-animal methods are required. "Non-animal test methods have been developed with the intention of detecting specific biomarkers based on adverse endpoints in humans. Therefore, to understand toxicity at the individual level, it is currently necessary to combine the results of several non-animal test methods, even if the focus is on biomarkers that can cause serious damage," Hirabayashi clarifies. "The combinations are complicated and diverse, costly and the information is limited. This means there is a need to develop an easy way to obtain a lot of information. Without it, we believe that true non-animal testing will not take root in society," she says.

AI NOT ANIMALS
One element of the research of Hirabayashi and the team involves the development of AI that can be used to evaluate the safety of new compounds, thereby eliminating the need for animal models. Towards realisation of the ultimate goal, the researchers are also developing a number of other non-animal methods. "The methods we are developing include: *in vitro* assays such as ordinary 2-dimensional culture, 3-dimensional culture including organoids or spheroids, reporter gene assay and organ-on-a-chip, and *in silico* assays such as computer toxicology using QSAR and Read Across," Hirabayashi reveals. "The organ-on-a-chip construct that the team is developing is an original microphysiological system (MPS) that will allow the researchers to model human tissues and look at how different compounds affect the tissues," she explains.

Through their study, the researchers have identified the key challenges facing the conversion of animal testing into the use of non-animal methods. One such limitation is the lack of biomarkers to more accurately

innovations emerging and advancing all the time, it makes sense to make use of these in the lab, where possible, to reduce animal testing," observes Hirabayashi. ▶

Project Insights

FUNDING
AMED Grant Number (JP21mk010188, MHLW Grant Number (PMH21K02003) and NIHS Research Grant (2020-2024)

ICATM PARTNER ORGANISATIONS
Interagency Coordinating Committee on the Validation of Alternative Methods, US (ICCVAM), European Union Reference Laboratory for Alternatives to Animal Testing (EURL-ECVAM), Health Canada, Canadian Centre for the Validation of Alternative Methods (CCVAM), Korean Centre for the Validation of Alternative Methods (KCCVAM), Brazilian Centre for the Validation of Alternative Methods (BICVAM)

JACVAM STEERING COMMITTEE
Yoko Hirabayashi Chairperson

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<https://www.ingentaconnect.com/content/sil/impact/2021/00002021/00000008/art00017#>

Thank you very much for your attention



National Institute of Health Sciences,
Center for Biological Safety and Research