Environmental Defense Fund

Comments on

Validation, Qualification, and Regulatory Acceptance of New Approach Methodologies

A Report of the Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM) Validation Workgroup

Submitted: September 05, 2023

Introduction

Environmental Defense Fund (EDF) appreciates the opportunity to provide comments to the Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM) on the Validation, Quantification, and Regulatory Acceptance of New Approach Methodologies.[1] The report, compiled based on the expertise of 17 U.S. federal regulatory and research agencies that use and generate toxicological and safety data, provides recommendations and support of global plans to move towards the use of NAMs as a primary means of testing chemicals for approved use in the environment and in consumer products. The ICCVAM report is positive in that experts carefully review the validation, qualification, and regulatory confidence in NAMs, providing recommendations for their application and implementation going forward. Ultimately this work will support the development and use of NAMs to inform chemical safety in a way that addresses the risks we all face, particularly those faced by the most vulnerable communities and individuals.

However, we do have several concerns, mostly surrounding the validation of NAMs in a uniform manner, as well as the generation of consistent and comparable results across the different entities tasked with switching to NAMs for chemical testing. EDF wants to ensure that chemical assessments completed using NAMs do not dismiss toxicity or potential risk prematurely, and that the data generated via new assays is an advancement compared to the traditional animal testing. Below are our specific comments on the draft ICCVAM report.

[1] ICCVAM, "Validation, Qualification, and Regulatory Acceptance of New Approach Methodologies," 88 FR 54342, August 10, 2023, https://ntp.niehs.nih.gov/go/ICCVAM-submit

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1. ICCVAM should provide clearer guidance on the need for validation of NAMs

We applaud ICCVAM's inclusion of a discussion on validation in its report. EDF understands that the process for interlaboratory ring trials is lengthy, but that its purpose is to demonstrate reproducibility and repeatability of assays between laboratories.[2] Given the nature of chemical testing and that NAMs will be used globally to evaluate chemical safety, EDF feels that it is imperative to have a clear set of steps that will be followed to determine that NAMs are not only fit for purpose, but that findings between different laboratories are in agreement with one another.

A. Validation via interlaboratory comparisons to determine reproducibility should be strongly considered

In its report, ICCVAM should provide clearer guidelines on how NAMs will be validated. The introduction of new assays for first time use by many laboratories producing data for use in regulatory decisions is rife with the potential for the data produced to be highly variable and inconsistent. While we appreciate that ICCVAM raised the importance of interlaboratory testing and validation in several portions of the report, in some cases the recommendations are a bit vague and difficult to follow. For example, on page 6 of the report ICCVAM states "there should be evidence to support that the use of an alternative method will lead a regulatory review to the same or more protective decision as the reviewer would make based on existing methods," but there is not a specific recommendation as to how this evidence would be gathered. In the flow chart on page 20, after "Within laboratory evaluation of assay performance," the next box states "Determination of method transferability (if needed)." If an assay is designed to be used by multiple laboratories for the sake of consistency, it seems that it would always be necessary to determine method transferability and repeatability. For NAMs that will be performed in more than one location, which presumably would be many of them, training and transfer studies to ensure reproducibility should be conducted to demonstrate that a naïve laboratory can produce reliable data from the NAM[2,3] (Hartung et al. 2004, van der Zalm et al. 2022). This may be the intent of the "(if needed)" noted on page 20, but "if needed" leaves the door open to interpretation. This issue is not thoroughly addressed and should be.

[2] Hartung T, Bremer S, Casati S et al (2004) A modular approach to the ECVAM principles on test validity. Altern Lab Anim 32:467–472. https://doi.org/10.1177/026119290403200503

[3] van der Zalm, Anna J. van der, João Barroso, Patience Browne, Warren Casey, John Gordon, Tala R. Henry, Nicole C. Kleinstreuer, Anna B. Lowit, Monique Perron, and Amy J. Clippinger. "A Framework for Establishing Scientific Confidence in New Approach Methodologies." Archives of Toxicology 96, no. 11 (November 1, 2022): 2865–79. https://doi.org/10.1007/s00204-022-03365-4

B. Clarification on how independent review would proceed is needed

Continuing on the points made above, if inter-laboratory testing for determination of method transferability is necessary, a clear plan on how this would proceed is needed. If it is true that under many statutory and regulatory requirements that information from NAMs must not be less protective than existing methods[1], how would a laboratory currently conducting animal testing demonstrate to an independent group or board of reviewers that they had successfully implemented NAMs which satisfactorily replace older assays? This is not explicitly described in the report. While it is stated on page 37 that independent peer review of NAMS may be organized by validation bodies such as NICEATM, EURL ECVAM, ESAC, JaCVAM, etc., the section ends by stating that the extent of independent review will vary depending on the context of use, regulatory framework, and specific method being evaluated. EDF is concerned that a lack of definition and requirements surrounding independent review could lead to industry finding loopholes in the process that allow for assays which are less likely to produce a sufficiently protective regulatory outcome. The report should clearly explain the criteria for demonstrating that NAMs satisfactorily improve upon older assays.

2. , Biological relevance of NAMs is clearly demonstrated, but ICCVAM could provide a better roadmap for how to move forward

EDF is pleased to see that that NAMs are in many cases more representative of human biology than assays with rodents or other model vertebrates, and that their biological relevance, defined as a measure of appropriateness for assessing effects of a chemical within the taxa of interest, is well supported by testing to date.[2] However, we have some concerns about the specificity of the roadmap being provided to laboratories that would be implementing new testing protocols.

A. Clarity is needed to determine how testing should proceed if an AOP to anchor the NAM to is lacking

Adverse outcome pathways, which are defined in the report as a structured representation of sequential events that occur at different levels of organization resulting in an adverse effect when an organism is exposed to a substance, are intended to be anchored to a NAM to demonstrate its biological relevance. However, many new chemicals, as well as chemicals that have been used and studied for many years (e.g. PFAS)[4], lack clear molecular initiating events, or not enough is known about them to match an AOP for an existing chemical to the new chemical (e.g. developmental neurotoxicity)[1].

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[4] Goodrum, Philip E, Janet K Anderson, Anthony L Luz, and Graham K Ansell. "Application of a Framework for Grouping and Mixtures Toxicity Assessment of PFAS: A Closer Examination of Dose-Additivity Approaches." *Toxicological Sciences* 179, no. 2 (February 1, 2021): 262–78. https://doi.org/10.1093/toxsci/kfaa123.

While the report states that ICCVAM recommends comprehensively characterizing and clearly describing what biological event is being measured and how it relates to the adverse outcome or hazard of concern, a flow chart or step-by-step process by which this characterization would take place is needed. The report should explain whether the lack of a detailed AOP that a NAM could be anchored to would trigger additional interlaboratory testing or involve additional independent review. Clear control mechanisms should be in place to ensure that all new assays implemented are biologically relevant for the endpoint being tested.

B. Additional guidance is required to determine which scenarios would lead to traditional animal test methods being used ,

On page 17 of the ICCVAM report, it is mentioned that there may be circumstances under which traditional animal test methods may still need to be used for comparison. Further, later in the document it is mentioned that the use of a whole animal test is up to the sponsor.[1] EDF is concerned that following this recommendation could lead to conflicts of interest, particularly if it is in the interest of the sponsor to have a chemical approved for use quickly. We recommend that decisions such as whether and when to use traditional animal test methods should also be subject to an independent review process.

References

[1] ICCVAM, "Validation, Qualification, and Regulatory Acceptance of New Approach Methodologies," 88 FR 54342, August 10, 2023, http://ntp.niehs.nih.gov/go/ICCVAM-submit

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