



International Workshop on Alternatives to the Murine Histamine Sensitization Test (HIST): State of the Science and the Path Forward

Workshop Introduction and Objectives

Juan L Arciniega, DSc, FDA
ICCVAM Biologics Working Group

November 28, 2012

William H. Natcher Conference Center
Bethesda, MD



Workshop on Alternatives to the Murine Histamine Sensitization Test (HIST) for Acellular Pertussis Vaccines



U.S. Reported Pertussis Incidence 1990-2010

Year	Incidence (per 100,000)
1990	0.5
1995	1.0
2000	1.5
2005	8.0
2010	4.0

International Workshop on Alternatives to the Murine Histamine Sensitization Test (HIST) for Acellular Pertussis Vaccines: State of the Science and the Path Forward

November 28–29, 2012
William H. Natcher Conference Center
National Institutes of Health
Bethesda, Maryland, USA

Organized by members of the International Cooperation on Alternative Test Methods:
NICEATM - National Toxicology Program Interagency Center for the Evaluation of Alternative Toxicological Methods
ICCVAM - Interagency Coordinating Committee on the Validation of Alternative Methods
EURL ECVAM - European Union Reference Laboratory for Alternatives to Animal Testing
JasCVAM - Japanese Center for the Validation of Alternative Methods
KorCVAM - Korean Center for the Validation of Alternative Methods
Health Canada

For more information and to register, please contact NICEATM:
<http://iccvam.niehs.nih.gov/> — (919) 541-2384 — niceatm@niehs.nih.gov

Individuals with disabilities who need accommodation to participate in this event should contact Debbie McCarley at 919-541-2384 or mccarley@niehs.nih.gov. TTY users should contact the Federal TTY Relay Service at 800-877-8339. Requests should be made at least 5 days in advance of the event.

ICCVAM Agencies:

• Agency for Toxic Substances and Disease Registry	• Department of Energy	• National Institute of Environmental Health Sciences
• Consumer Product Safety Commission	• Food and Drug Administration	• National Institutes of Health
• Department of Agriculture	• National Cancer Institute	• National Library of Medicine
• Department of Defense	• Department of Transportation	• Department of the Interior
	• National Institute for Occupational Safety and Health	• Occupational Safety and Health Administration
		• Environmental Protection Agency

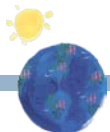
Photomicrograph courtesy of Jeremy Casan, Ph.D., Department of Pathology, University of North Carolina School of Medicine.
Line graph courtesy of U.S. Centers for Disease Control and Prevention.
Vaccination photograph courtesy of James Salzman, U.S. Centers for Disease Control and Prevention.



- November 28-29, 2012
- William H. Natcher Conference Center
National Institutes of Health,
Bethesda, MD
- Experts from government, academia
and industry expected to attend
- Plenary and Breakout Group Sessions
- NICEATM coordinating with the
International working group on
alternatives to HIST
- Review of *in vitro* safety data on
spiked Pertussis toxin vaccine
preparations
- Workshop report to be published
- 47 registrants to date from 11
countries

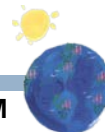
Further information, including registration, is available at:

<http://iccvam.niehs.nih.gov/meetings/HISTWksp-2012/HISTWksp.htm>



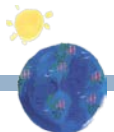
Alternatives to HIST Workshop Objectives (1)

- Review the usefulness and limitations for alternative *in vitro* test methods proposed to replace the current *in vivo* HIST
- Review *in vitro* protocols and data generated by participants of the International Working Group on Alternatives to HIST
 - Use of common set of vaccines, pertussis toxin (reference standard), and protocol for spiking
 - *In vitro* assays tested
 - Biochemical assays
 - Binding assay: used to assess the amount of pertussis toxin/toxoid binding activity to the glycoprotein fetuin.
 - Enzymatic assay: monitors the residual ADP-ribosylation of the pertussis toxin
 - Cell-based assays
 - Human cells measuring ATP reduction
 - Rat cells measuring cAMP increase
 - CHO cell (morphological, cytopathic)



Alternatives to HIST Workshop Objectives (2)

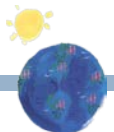
- Discuss application of *in vitro* assays for monitoring consistency of vaccine manufacture as alternatives to the HIST
- Establish framework for international collaboration to validate *in vitro* assay(s) for acellular pertussis vaccine testing
- Identify regulatory acceptance requirements for *in vitro* assays as alternatives to the HIST



Workshop Program – Day 1 (1)

Opening Session

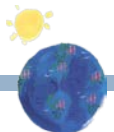
- Plenary Presentation: The Many Faces of Pertussis Toxin
- Current Regulatory Requirements for Residual Pertussis Toxin Testing of Acellular Vaccines
- 2010 International Collaborative Study on Validation of an *In Vitro* Assay System as an Alternative to the Current Histamine Sensitization Test for Acellular Pertussis Vaccines
- Overview of the International Working Group for Alternatives to HIST (Phase 1)



Workshop Program – Day 1 (2)

Session 1: Alternatives to HIST: Methods and Evaluations

- Analyses of Pertussis Toxin ADP-ribosyltransferase and Carbohydrate-Binding Activities as an *In Vitro* Alternative to the *In Vivo* HIST
- *In Vitro* Assays for the Detection of Pertussis Toxin: BSP114 Collaborative Study Results
- Alternative *In Vitro* Methods for Detection of Pertussis Toxin in Component Pertussis Vaccines: Results of a BSP114 Phase 1 International Collaborative Study
- Characteristics of Enzymatic and Binding Activities of Pertussis Toxin According to Chemical Detoxifying Agents
- Detection of PTx in Acellular Pertussis Vaccines Using a CHO Cell Clustering Assay, a Carbohydrate-Binding Assay, and an Enzymatic/HPLC Assay

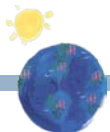


Workshop Program – Day 1 (3)

Session 1: Alternatives to HIST: Methods and Evaluations cont.

- Cell-Based Assays for Detection of Pertussis Toxin in Acellular Vaccines: The Pertussis ATP Test (PAT) and the cAMP-PTx Assay
- Using Pertussis Toxin-Sensitive Genes in Dendritic Cells to Evaluate the Safety of Acellular Pertussis Vaccines
- Summary Analysis of Reported Data Sets

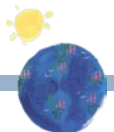
- Session 1B: Alternative *In Vitro* Methods to the Murine Histamine Sensitization Test
- Roundtable Discussion



Workshop Program – Day 2 (1)

Session 2: The Path Forward: Gaps to Cross and Bridges to Build in the Road Toward the Adoption of Alternatives to HIST

- Plenary Presentation: Pertussis Toxin and the CHO Cell Response
- Session 2A: CHO Cell Assay: Potential Use for Standardization and as an Alternative to HIST
- Roundtable Discussion
- Session 2B: Issues with Pertussis Toxin Adsorption/Desorption
- Roundtable Discussion



Workshop Program – Day 2 (2)

Session 2C: The Path Forward: Harmonizing the Adoption of Alternative Assays

- Roundtable Discussion
- Session 3: Next International Collaborative Validation Study on Alternative Assay(s) with Spiked Vaccines
- Roundtable Discussion
- Closing Remarks and Adjournment



Acknowledgements: NICEATM and Support Staff

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Director

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Deputy Director

Debbie McCarley

Special Assistant

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Lori Rinckel, PhD, Principal Investigator

Steven Morefield, MD, Project Manager

Vivian Doelling, PhD

Brett Jones, PhD

Nelson Johnson, BS

Linda Litchfield

Anna Lee Mosley

Catherine Sprankle

Linda Wilson

