Preface

Allergic contact dermatitis (ACD) is an adverse health effect that frequently develops in workers and consumers exposed to skin-sensitizing chemicals and products. ACD results in lost workdays[1] and can significantly diminish quality of life (Hutchings et al. 2001; Skoet et al. 2003). To minimize the occurrence of ACD, regulatory authorities require testing to identify substances that may cause it. Sensitizing substances must be labeled with a description of the potential hazard and the precautions necessary to avoid development of ACD.

Skin sensitization testing has typically required the use of guinea pigs (Buehler 1965; Magnusson and Kligman 1970). However, in 1998, the Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM) evaluated an alternative known as the murine (mouse) local lymph node assay (traditional LLNA[2]). ICCVAM concluded that the traditional LLNA provided several advantages over the commonly accepted guinea pig test methods, including elimination of potential pain and distress, use of fewer animals, less time to perform, and availability of dose-response information. ICCVAM recommended the LLNA as an alternative test method for assessing the skin sensitization potential of most types of substances. United States and international regulatory agencies subsequently accepted the traditional LLNA as a valid alternative test method for ACD testing.

In 2007, the U.S. Consumer Product Safety Commission requested that ICCVAM evaluate several modifications of the traditional LLNA,[3] including the "reduced LLNA" (rLLNA), also referred to as the "cut-down" or "limit dose" LLNA. ICCVAM assigned this activity a high priority after considering comments from the public and ICCVAM's Scientific Advisory Committee on Alternative Toxicological Methods (SACATM). As part of their ongoing collaboration with ICCVAM, scientists from the European Centre for Validation of Alternative Methods and the Japanese Center for Validation of Alternative Methods served as liaisons to the ICCVAM Immunotoxicity Working Group (IWG). A detailed timeline of the rLLNA test method evaluation is included with this report.

This Test Method Evaluation Report provides ICCVAM's recommendations regarding the usefulness and limitations of the rLLNA for assessing the ACD potential of substances. When deemed appropriate for use, the rLLNA can reduce by 40% the number of animals used for each test compared to the traditional LLNA. The report also provides the updated ICCVAM-recommended LLNA test method protocol, which addresses the rLLNA procedure. The database of substances used to validate the rLLNA is discussed and summarized.

ICCVAM carefully compiled and assessed all available data and arranged an independent scientific peer review. ICCVAM and the IWG solicited and considered public comments and stakeholder involvement throughout the rLLNA evaluation process. The National Toxicology Program Interagency Center for the Evaluation of Alternative Methods (NICEATM), ICCVAM, and the IWG began the process by preparing a draft background review document (BRD) describing the validation status of the rLLNA test method, including its reliability and

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[1] http://www.bls.gov/IIF.

[2] The "traditional LLNA" refers to the validated ICCVAM-recommended LLNA test method protocol, which measures lymphocyte proliferation based on incorporation of tritiated thymidine into the cells of the draining auricular lymph nodes.

[3] Available at http://iccvam.niehs.nih.gov/methods/immunotox/llnadocs/CPSC_LLNA_nom.pdf.

accuracy for the substances evaluated, and draft test method recommendations for usefulness and limitations. ICCVAM released these documents to the public for comment on January 8, 2008, at which time ICCVAM also announced a meeting of the international independent scientific peer review panel (Panel) (*Federal Register* 73 FR 1360[4]).

The Panel met in public session on March 4–6, 2008, to review the ICCVAM draft BRD for completeness and accuracy. The Panel then evaluated (1) the extent to which the draft BRD addressed established validation and acceptance criteria and (2) the extent to which the BRD supported ICCVAM's draft test method recommendations. Before concluding their deliberations, the Panel considered written comments and comments made at the meeting by public stakeholders. The final Panel report was made available to the public for comment on May 20, 2008.[5]

ICCVAM provided SACATM with the draft BRD and draft Test Method Evaluation Report, the Panel report, and all public comments for discussion at their meeting on June 18-19, 2008, where public stakeholders were given another opportunity to comment.

After SACATM's meeting, ICCVAM considered the SACATM comments, the Panel report, and all public comments before finalizing the ICCVAM Test Method Evaluation Report and Background Review Document, which is provided as an appendix to this report. The consolidated document will be provided to U.S. Federal regulatory agencies for consideration and be made available to the public. The ICCVAM Authorization Act requires that Federal agencies respond to ICCVAM within 180 days after receiving the ICCVAM test method recommendations. Agency responses will be posted on the NICEATM–ICCVAM website[6] as they become available.

We gratefully acknowledge the many individuals who contributed to the preparation, review, and revision of this report. We especially recognize the Panel members for their thoughtful evaluations and generous contributions of time and effort. Special thanks are extended to Dr. Michael Luster for serving as the Panel Chair and to Dr. Michael Woolhiser, Dr. Michael Olson, and Ms. Kim Headrick for their service as Evaluation Group Chairs. We thank the IWG for assuring a meaningful and comprehensive review. We especially thank Dr. Joanna Matheson (Consumer Product Safety Commission) and Dr. Abigail Jacobs (U.S. Food and Drug Administration Center for Drug Evaluation and Research) for serving as Co-Chairs of the IWG. Integrated Laboratory Systems, Inc., the NICEATM support contractor, provided excellent scientific and operational support, for which we thank Dr. David Allen, Mr. Thomas Burns, Ms. Linda Litchfield, Mr. Michael Paris, Dr. Eleni Salicru, Ms. Catherine Sprankle, Dr. Judy Strickland, and Ms. Linda Wilson; and Dr. Joseph Haseman, ILS consulting statistician, for statistical support. We also acknowledge Dr. Raymond Tice, Deputy Director of NICEATM, for his efforts. Finally, we thank Dr. Silvia Casati and Dr. Hajime Kojima, the IWG liaisons from the European Centre for the Validation of Alternative Methods and the Japanese Center for the Validation of Alternative Methods, respectively, for their participation and contributions.

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[4] Available at http://iccvam.niehs.nih.gov/SuppDocs/FedDocs/FR/FR_E7_25553.pdf.

[5] Announced in 73 FR 29136 (http://iccvam.niehs.nih.gov/SuppDocs/FedDocs/FR/FR-E8-11195.pdf); available at http://iccvam.niehs.nih.gov/docs/immunotox_docs/LLNAPRPRept2008.pdf.

[6] http://iccvam.niehs.nih.gov/methods/immunotox/rLLNA.htm

This comprehensive ICCVAM evaluation of the rLLNA should facilitate regulatory agency decisions on the acceptability of the method. Following regulatory acceptance, use of the method by industry can be expected to significantly reduce the number of animals required for ACD testing while continuing to support the protection of human health.

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