PREFACE

During the past 60 years, government regulatory agencies have implemented safety testing requirements to identify potential hazards of various chemicals and products in order to protect human health and the environment. Testing results are used for hazard classification and labeling and to identify appropriate risk management practices necessary to reduce or avoid human injury, disease, disability, and/or death. The first standardized toxicity test method developed for assessing the safety of a chemical ingredient or new product was for chemically-induced eye injuries (Draize et al. 1944). The U.S. Food and Drug Administration (FDA) developed this test in response to new laws implemented as a result of permanent eye injuries from various cosmetic products in the 1930s (Calabrese 1983). Various national and international regulatory authorities now require updated versions of this test method to assess whether substances can potentially cause eye irritation or corrosion. The U.S. Consumer Product Safety Commission (CPSC), the U.S. Environmental Protection Agency (EPA), FDA, and the U.S. Occupational Health and Safety Administration (OSHA) have testing requirements and guidelines in place for assessing the ocular irritation of various substances such as pesticides, hazardous household products, pharmaceuticals, cosmetics, and agricultural and industrial chemicals.

While ocular safety assessments have clearly supported appropriate protection of consumers and workers, there have been concerns raised about the humane aspects of this test method. Various modifications to the Draize rabbit eye test (Draize et al. 1944) have now been adopted by regulatory authorities that reduce the numbers of animals used and that reduce the potential pain and distress associated with the procedure. Significant progress has been made during the last decade, with only one to three rabbits now required per test compared to six rabbits in the original protocol, and addition of provisions that allow for humane euthanasia of animals with severe lesions or discomfort. In addition, a number of scientists and organizations began to develop nonanimal alternatives in the early 1980s that might be useful in further reducing or replacing the need for animals for the assessment of ocular irritancy and corrosion. Although a great deal of progress has been made, there is currently no accepted nonanimal alternative test method for ocular irritancy in the United States. Cognizant of various *in vitro* methods that had been developed and have undergone some degree of validation, the Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM) Scientific Advisory Committee on Alternative Toxicological Methods (SACATM) recommended in August 2003 that ICCVAM give high priority to reviewing the validation status of *in vitro* test methods proposed for identifying ocular irritants/corrosives. In October 2003, the EPA formally nominated several ocular irritation test methods and related activities for evaluation by ICCVAM. This included review of the validation status of four *in vitro* methods for identifying potential ocular corrosives and severe irritants in a tiered testing strategy. Validation[1] of a test method is a prerequisite for it to be considered for regulatory acceptance (ICCVAM 1997, 2003). The four test methods were the Bovine Corneal Opacity and Permeability (BCOP) assay, the Hen's Egg Test - Chorioallantoic Membrane (HET-CAM) assay, the Isolated Chicken Eye (ICE) assay, and the Isolated Rabbit Eye (IRE) assay.

[1] Validation is the process by which the reliability and relevance of a test method are established for a specific purpose (ICCVAM 1997, 2003).

ICCVAM, which is charged with coordinating the technical evaluations of new, revised, and alternative test methods with regulatory applicability (ICCVAM Authorization Act of 2000, Public Law [P.L.] 106-545), unanimously agreed that the four nominated *in vitro* test methods should have a high priority for evaluation. An ICCVAM Ocular Toxicity Working Group (OTWG) was established to work with the National Toxicology Program (NTP) Interagency Center for the Evaluation of Alternative Toxicological Methods (NICEATM) to carry out these evaluations. ICCVAM and NICEATM also collaborate closely with the European Centre for the Validation of Alternative Methods (ECVAM), a component of the European Commission's Joint Research Centre. Accordingly, an ECVAM liaison was designated for the ICCVAM OTWG to ensure input and contributions during the evaluation and review process.

NICEATM, which administers the ICCVAM and provides scientific support for ICCVAM activities, subsequently prepared four comprehensive background review documents (BRDs) that provided information and data about the current validation status of the four nominated *in vitro* test methods (i.e., BCOP, HET-CAM, ICE, and IRE) for detecting ocular corrosives and severe irritants. These draft BRDs were based on published studies using the identified test methods, and other data and information submitted in response to a 2004 *Federal Register* (*FR*) request (Available: http://iccvam.niehs.nih.gov/methods/eyeirrit.htm), and were made available to the public on November 1, 2004 (Available: http://iccvam.niehs.nih.giv/methods/ocudocs/ocu_brd.htm). Notification for data also was made through the ICCVAM electronic mailing list.

ICCVAM subsequently convened an Expert Panel meeting on January 11-12, 2005, to independently assess the validation status of these four *in vitro* test methods for identifying ocular corrosives or severe irritants. Prior to this meeting, public comments on the BRDs were received from three organizations and provided to the Expert Panel for their consideration. Public comments at the meeting revealed that additional relevant data was available that had not previously been provided in response to earlier requests for data. The Expert Panel recommended that the additional data be requested and that a reanalysis of the accuracy and reliability of each test method be conducted, where appropriate (the Expert Panel report from this meeting is available at http://iccvam.niehs.nih.gov/methods/eyeirrit.htm).

In response to this recommendation, an *FR* notice was published on February 28, 2005 (Available: http://iccvam.niehs.nih.gov/methods/eyeirrit.htm), which requested all available *in vitro* data on these four *in vitro* ocular irritancy test methods and corresponding *in vivo* rabbit eye test method data, as well as any human exposure data (either via ethical human studies or accidental exposure). A request for relevant data was resent directly to the primary developers or users of each test method. In response to these requests, additional *in vitro* test method data and corresponding *in vivo* rabbit eye test results were submitted for the BCOP, HET-CAM, and ICE test methods. These additional data were used to update the performance statistics of the test methods. Several U.S. Federal agencies (OSHA, CPSC, and the National Institute for Occupational Safety and Health [NIOSH]), along with the US Eye Injury Registry (USEIR) were also contacted directly for data resulting from accidental human exposures. However, given the lack of details about the specific nature of the

substances reported and their associated exposure conditions, these types of accidental human exposure injury data were not useful for evaluating the accuracy of the BCOP test method for predicting human ocular hazard.

Further clarification of hazard classification rules for severe irritants also was obtained subsequent to the release of the four draft BRDs. This change resulted in a small number of substances previously classified as nonsevere irritants now being classified as severe irritants (from 10 to 15, depending on the test method and the classification system used). This change necessitated a reanalysis of the accuracy and reliability of all four of the test methods previously evaluated.

The original draft BRDs also provided an evaluation of the accuracy of each test method by chemical class. Subsequent to the release of the draft BRDs, the chemical classes assigned to each test substance were revised based on a chemical classification system consistent with the U.S. National Library of Medicine's Medical Subject Headings (MeSH; Available: http://www.nlm.nih.gov/mesh), an internationally recognized standardized classification scheme. This scheme was used to ensure consistency in classifying substances by chemical class among all the *in vitro* ocular test methods under consideration, and resulted in some chemicals being reclassified into different chemical classes. As a result, the accuracy of each test method by chemical class was reanalyzed.

To incorporate the additional data submitted, the changes in irritancy classification, and the revised chemical classes, a BRD Addendum was developed. The purpose of this document was to highlight changes in the performance statistics due to the above noted updates. The BRD Addendum was released on July 26, 2005, with notification of its release via an *FR* notice and notification through the ICCVAM electronic mailing list (and is available in electronic format on the ICCCVAM/NICEATM website, http://iccvam.niehs.nih.gov/methods/ocudocs/reanalysis.htm). The Expert Panel was subsequently reconvened via public teleconference on September 19, 2005 to discuss the BRD Addendum. Prior to this meeting, public comments on the Addendum were received from three organizations and provided to the Expert Panel for their consideration (no public comments were provided during the public teleconference). The Expert Panel then provided final endorsement regarding the effects, if any, of the information in the BRD Addendum on their original evaluation from the January 11-12, 2005 meeting (the Expert Panel report from this meeting is available at http://iccvam.niehs.nih.gov/methods/ocudocs/EPreport/EPrptAddend.htm).

NICEATM has subsequently prepared revised BRDs to reflect a compilation of the updated information for each test method. Each BRD provides a comprehensive summary of the current validation status of the *in vitro* test method, including what is known about its reliability and accuracy, and the scope of the substances tested. Raw data for these test methods will be maintained for future use. Therefore, the performance statistics of these test methods will be updated as additional information becomes available.

The ICCVAM and its OTWG will consider both Expert Panel reports, the updated performance statistics presented in the BRDs, and any public comments in preparing its final

test method recommendations for these *in vitro* ocular test methods. These recommendations will be made available to the public and provided to the U.S. Federal agencies for consideration, in accordance with the ICCVAM Authorization Act of 2000 (Public Law 106-545) (Available: http://iccvam.niehs.nih.gov/about/PL106545.pdf).

We want to acknowledge the excellent cooperation and contributions from the many organizations and scientists who provided critical data and information necessary for the BRD. The efforts of the many individuals who contributed to the preparation of this document also are gratefully acknowledged. These include David Allen, Ph.D., Bradley Blackard, M.S.P.H., Thomas A. Burns, Jr., M.S., Jeffrey Charles, Ph.D., M.B.A., D.A.B.T., Neepa Choksi, Ph.D., and James Truax, M.A. of Integrated Laboratory Systems (ILS), Inc., the NICEATM Support Contractor, as well as the members of the ICCVAM OTWG and ICCVAM representatives who reviewed various drafts. We also want to thank Raymond Tice, Ph.D., Deputy Director of NICEATM, for his extensive efforts on this project. Finally, we want to recognize the excellent leadership of the OTWG Co-chairs, Dr. Karen Hamernik (U.S. Environmental Protection Agency) and Dr. Jill Merrill (U.S. Food and Drug Administration).

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