

1 **ICCVAM NOMINATION FOR FUTURE STUDY: EVALUATION OF THE**  
2 **OPTIMAL CORNEAL HOLDER AND VEHICLE FOR THE BOVINE CORNEAL**  
3 **OPACITY AND PERMEABILITY TEST METHOD**

4 **Description of Project**

5 The nominated activity is the evaluation of alternative corneal holders and vehicles for the  
6 Bovine Corneal Opacity and Permeability (BCOP) test method.

7 **Background/Introduction**

8 In October 2003, the U.S. Environmental Protection Agency (EPA) nominated four *in vitro*  
9 test methods proposed for identifying potential ocular corrosives and severe irritants in a  
10 tiered-testing strategy for review of their current validation status. The test methods were the  
11 Bovine Corneal Opacity and Permeability (BCOP) assay, the Hen’s Egg Test -  
12 Chorioallantoic Membrane (HET-CAM) assay, the Isolated Chicken Eye (ICE) assay, and  
13 the Isolated Rabbit Eye (IRE) assay. The National Toxicology Program (NTP) Interagency  
14 Center for the Evaluation of Alternative Toxicological Methods (NICEATM), in conjunction  
15 with the Ocular Toxicity Working Group (OTWG), prepared a background review document  
16 (BRD) for each test method to describe its current validation status. The Interagency  
17 Coordinating Committee on the Validation of Alternative Methods (ICCVAM) convened an  
18 Expert Panel to assess the validation status of the methods. The Expert Panel Report  
19 (ICCVAM 2005) concluded the following:

20 “For the purposes of detecting severe eye irritants in the tiered testing scheme outlined in the  
21 BRD, the proposed BCOP test method protocol is useful for identification of severe corrosive  
22 ocular irritants with the following caveats: ...

- 23           • 0.9% NaCl should be used instead of distilled water as the test substance  
24           diluent.”

25 "The following are recommended as modifications that might improve the accuracy and  
26 reliability (repeatability/reproducibility) of the BCOP test method:

- 27           • Use of the larger holder as suggested by Ubels et al. (2002, 2004)"

28 The draft BRDs and the Expert Panel report were made available to the Scientific Advisory  
29 Committee on Alternative Toxicological Methods (SACATM) for their consideration at their  
30 meeting on December 12, 2005. SACATM agreed with the conclusions of the Expert Panel.  
31 ICCVAM subsequently prepared final test method recommendations based on the Expert  
32 Panel report and SACATM comments, which will be made publicly available and provided  
33 to U.S. Federal agencies (Available: <http://iccvam.niehs.nih.gov/>).

34 ICCVAM also convened a symposium, *Mechanisms of Chemically-Induced Ocular Injury*  
35 *and Recovery*, to review the state-of-the-science and understanding of the pathophysiology  
36 and mechanisms of chemically-induced ocular injury and recovery (reversibility vs.  
37 irreversibility) (Available:  
38 <http://iccvam.niehs.nih.gov/methods/ocudocs/ocumeet/sympinfo.htm>). At that symposium,  
39 Dr. John Ubels gave a presentation entitled, *In Vitro Models of Ocular Injury: Bovine Cornea*  
40 *Opacity and Permeability Assay*, during which he discussed the rationale and advantages of  
41 using the new corneal holder in the BCOP assay.

42 Based on the conclusions in the Expert Panel Report and the presentation at the symposium,  
43 ICCVAM concluded that studies should be conducted to evaluate the impact of using a  
44 corneal holder that maintains normal corneal curvature (e.g., the corneal mounting system  
45 designed by Ubels et al. 2002) on accuracy and/or reliability of the BCOP test method.  
46 ICCVAM also concluded that the effect of using 0.9% sodium chloride (NaCl) instead of  
47 distilled water as the diluent should be evaluated.

#### 48 **Objective**

49 To define the optimum corneal holder and vehicle for the BCOP test method.

#### 50 **Method/Proposed Activity**

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- 52 • NICEATM, working in partnership with the OTWG and with interested  
53 stakeholders, will manage a study, using ICCVAM recommended reference  
54 substances, to evaluate the performance (accuracy and reliability) of the  
55 BCOP using alternative corneal holders (e.g., holder developed by Ubels et al.  
56 2002) and test substance vehicles (e.g., 0.9% NaCl).

56 **DRAFT ICCVAM Recommended Priority: High**

57

58 **References**

59 ICCVAM Test Method Evaluation Report: In Vitro Ocular Toxicity Test Methods for  
60 Identifying Severe Irritants and Corrosives (Available: <http://iccvam.niehs.nih.gov/>).

61 ICCVAM. 2005. EXPERT PANEL FINAL REPORT: Evaluation of the Current Validation  
62 Status of In Vitro Test Methods for Identifying Ocular Corrosives and Severe Irritants  
63 (Available: <http://iccvam.niehs.nih.gov/methods/ocudocs/EPreport/ocuEPrpt.pdf>).

64 Stokes WS, Tice RR, Allen DG, Choksi NY, Truax JF. The ICCVAM/NICEATM/ECVAM  
65 symposia on mechanisms of chemically induced ocular injury and recovery  
66 (<http://iccvam.niehs.nih.gov/methods/ocudocs/ocumeet/sympinfo.htm>).

67 Ubels JL, Ditlev JA, Clousing DP, Casterton PL. 2004. Corneal permeability in a redesigned  
68 corneal holder for the bovine cornea opacity and permeability assay. *Toxicol In Vitro*  
69 18:853-857.

70 Ubels JL, Paauw JD, Casterton PL, Kool DJ. 2002. A redesigned corneal holder for the  
71 bovine cornea opacity and permeability assay that maintains normal corneal morphology.  
72 *Toxicol In Vitro* 16:621-628.