

Performance of the OptiSafe Ocular Irritation Assay in a Three-Laboratory Validation Study

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OptiSafe is an *in vitro* test method that assesses a test substance's potential to cause eye irritation by measuring damage caused when the substance is applied to a semi-permeable membrane. The membrane system allows the detection of substances with different mechanisms of ocular injury. NICEATM reviewed a study conducted by Lebrun Labs, which developed OptiSafe, and concluded that the study data compared favorably to other *in vitro* ocular toxicity testing methods. To further assess the transferability of the method to naïve laboratories and the overall performance and applicability domain of the method, NICEATM coordinated a multi-laboratory validation study to evaluate hazard identification of non-surfactants. Phase 1 testing of five chemicals in each laboratory showed that the method could be transferred to naïve laboratories. Thirty coded chemicals selected by a validation management team were then tested by all three laboratories in Phase 2. Test method performance was assessed using both the EPA and GHS eye irritation hazard classification systems. Intralaboratory reproducibility for both classification systems ranged from 93% to 99%. Intralaboratory accuracy using the EPA classification system ranged from 82% to 88%. False negative and false positive rates ranged from 0% to 7% and 23% to 39%, respectively. Intralaboratory accuracy, false negative, and false positive rates using the GHS classification system ranged from 78% to 88%, 0% to 15%, and 23% to 36%, respectively. Interlaboratory reproducibility was 91% for both classification systems. Interlaboratory accuracy and false negative rates were 89% and 0%, respectively, for both classification systems. The false positive rates were 23% for the GHS classification system and 25% for the EPA classification system. Phase 3 testing of an additional 60 substances provided a comprehensive assessment of test method accuracy and defined the applicability domain of the method. These results suggest that the OptiSafe ocular irritation assay may represent a new tool for *in vitro* assessment of the ocular toxicity potential of chemicals in a tiered-testing system. *This project was funded in whole or in part with federal funds from the NIEHS, NIH under Contract No. HHSN273201500010C.*

Category: Alternatives to Mammalian Models III: Liver, Ocular, and Skin Alternatives

Keywords: in vitro and alternatives; alternatives to animal testing; ocular toxicity