Retrovirology

Poster presentation

P15-04. Development and implementation of an international proficiency testing program for a neutralizing antibody assay for HIV-1 in TZM-bl cells

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Background

Recent advances in assay technology led to major improvements in how HIV-1 neutralizing antibodies are measured. A luciferase reporter gene assay performed in TZM-bl (JC53bl-13) cells has been optimized, and many performance parameters of this assay have been validated. Because this assay has been adopted by multiple laboratories world-wide, an external proficiency testing program was developed to qualify laboratories to perform a Good Clinical Laboratory Practice (GCLP) compliant neutralizing antibody assay for HIV/AIDS vaccine clinical trials.

Methods

The program was optimized by conducting three independent rounds of testing, with an increased level of stringency from the first to third round. Results from the participating domestic and international laboratories improved at each round as factors that contributed to inter-assay variability were identified and minimized. Key contributors to increased agreement were experience among laboratories, standardization of reagents and adherence to GCLP.

Results

Based on results from the three rounds of testing, standardized proficiency test kits were assembled at QBI, Inc. Each kit consists of identical stocks of Env-pseudovirus and five blinded serologic reagents. At the beginning of the external proficiency program, five experienced reference laboratories will utilize the kits, in three repeats, to derive gold standard reference values and acceptance intervals for each antibody-isolate combination. The acceptance interval is defined by a 90% prediction interval with mean and variance estimated using logarithm transformed ID50 values measured in the five reference laboratories.

Conclusion

A 2-fold systematic difference in ID50 values was set as the acceptance criteria to qualify a lab such that results of two vaccine trials, each with 30–50 volunteers, could be compared, by qualified laboratories, with adequate statistical power to detect moderate differences in the magnitude and breadth of the neutralizing antibody response. This program is jointly sponsored by the NIH and the Collaboration for AIDS Vaccine Discovery.