ANALYSIS OF INTERNAL QUALITY CONTROL RESULTS IN ANTI-HLA ANTIBODY DETECTION BY LUMINEX TECHNOLOGY

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Introduction

Anti-HLA antibodies are essential markers in organ transplantation (1). Luminex is a broadly used technology for the detection of anti-HLA antibodies in transplant recipient serum that provides qualitative results (2).
Internal quality control (IQC) plays a key role to ensure the reliability and reproducibility of the test results (3). However, no global consensus exists on the interpretation of anti-HLA antibody detection using Luminex.
Based on our laboratory experience, this study aims to emphasize the importance of IQC in the accurate interpretation of this test.

Materials and Methods

We performed 30 series of anti-HLA antibody detection over three months. We used ONE LAMBDA LABScreen™ mixed classes I and II. Class I anti-HLA antibodies were screened using 12 beads, and Class II anti-HLA antibodies were screened using five beads. Only high control level samples were provided by the supplier. Luminex® platform was used for data acquisition, and HLA fusion™ was used for data analysis. We used Levey-Jennings control charts to analyze IQC results for each bead. The statistical distribution of the IQC values was interpreted according to Westgard rules (4).

WESTGARD RULES

- Rule 1_{2s}: one measurement is between ±2SD and ±3SD.
- Rule 2_{2s}: 2 consecutive values between ±2SD and ±3SD on the same side of the mean.
- Rule 1_{3s}: 1 measurement exceeds 3SD above or below the mean.
- Rule R_{4s}: a difference of 4 SD between 2 consecutive values.
- Rule 4_{1s}: 4 consecutive values exceed 1SD above or below the mean.
- Rule 10_x: 10 consecutive values on the same side of the mean.

Results

In the Levey-Jennings control charts, the x-axis represents the number of runs, and the y-axis represents the MFI results on IQC samples (Figure 1).
The interpretation of Levey-Jennings charts for each IQC bead revealed a distribution of the MFI results around the respective means within the limits of ±3 standard deviations (SD).
We used Westgard rules to study the distribution of the MFI results within the control limits. According to our results, Westgard rule 1_{2s} was violated in the positive control bead, 6 class I beads, and 1 class II bead. Rule 1_{3s} was violated in the positive control bead in one run. The rest of the rules were not violated.

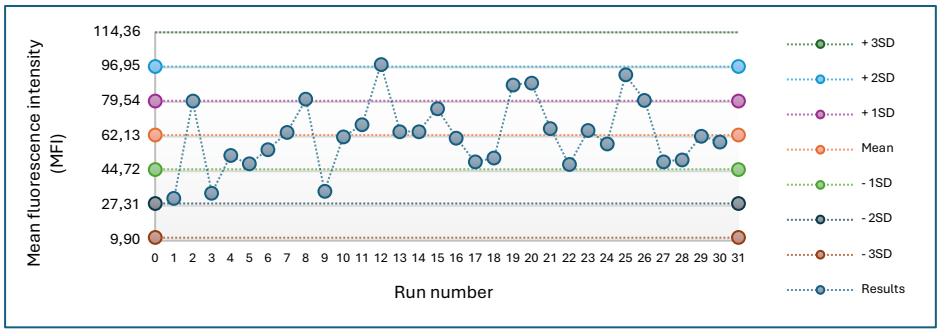

Figure 1: Levey-Jennings control chart for the negative control bead

Discussion

Internal quality control is essential in analyzing the results of anti-HLA antibody detection using Luminex technology. It ensures that each phase, from sample preparation to data collection, is optimally performed, which guarantees accurate and reproducible results (5).

Levey-Jennings control charts, combined with the application of Westgard rules, are used to monitor variations in IQC results and detect anomalies that could compromise data interpretation. The observed deviations may result from factors such as reagents, lot variations, instrumentation fluctuations, or sample-related interferences (3).

The application of Westgard rules in our study revealed the violation of rule 1_{2s} in several beads, which is considered only a warning; and the violation of rule 1_{3s} in one bead during one run.

Although these violations did not affect the validity of our results, this highlights the importance of closely monitoring each bead and raises the question whether an entire run should be rejected due to a violation by a single bead.

Anti-HLA antibodies detection has direct clinical implications in organ transplantation. This emphasizes the pressing need for evident international standards for interpreting IQC results in this context. It also underlines the importance of rigorous scrutiny and continuous adjustment of IQC procedures.

Conclusion

It is necessary to establish standards and guidelines that integrate the features of interpreting IQC results in anti-HLA antibody detection by Luminex technology.

References

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