IVD_14 - Extending the Shelf-Life of Protein A Gold conjugate for Rapid Syphilis Testing: A Comprehensive Stability Analysis

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Introduction: Rapid diagnostic tests (RDTs) for syphilis are crucial in global health settings for timely disease management and control. Bio-Manguinhos has developed an RDT for syphilis, which detects IgG antibodies in patient serum using a Protein A Gold conjugate (PtnA-Au conjugate). The established expiration date of the PtnA-Au conjugate solution is a critical factor in the Gold Pad spraying process and, therefore, to the RDTs production process. The aim of this study was to investigate the possibility of extending the expiration date from 6 hours to up to 24 or 48 hours prior to spraying the gold pad.

Objectives: To evaluate the physical-chemical stability by assessing the diagnostic performance of PtnA-Au conjugate at 3 different spraying time (T0, T24, and T48 hours) in order to assess a potential extended expiration date to the PtnA-Au conjugate solution.

Methodology: The PtnA-Au conjugate was prepared according to SEFEN's standard operating procedure. The Conjugate solution was divided into three parts of equal volume for spraying at T0, T24, and T48 hours. The diagnostic efficacy of the conjugate was assessed evaluating the qualitative (P1, P2 or P3) and semiquantitative (numeric value obtained using a rapid test reader) results for 48 characterized serum samples (24 positives and 24 negatives for syphilis) in Bio-Manguinhos Syphilis RDTs. Statistical analyses included normality tests, outlier detection, ANOVA, and Kruskal-Wallis tests to compare readings values across different times.

Results: The test reader results for P2 and P3 groups adhered to the normal distribution without outliers, indicating consistent test performance. The P1 group initially showed deviation from normality with an outlier at T24 and T48, which, upon removal, aligned results with the normal distribution. Statistical analysis revealed no significant differences in variance or mean absorbance values across the spraying times for all groups (P1, P2, P3), with a significance level of 0.05.

Conclusion: Our results indicate that the PtnA-Au conjugate could be sprayed up to 48 hours after the solution was ready. This extended expiration date would improve the gold pad spraying process, reducing the risks inherent to the process. Future research should focus on evaluating the shelf life of the RDT produced with the T24 and T48 PtnA-Au conjugates.

Keywords: Diagnostic Stability; Conjugate Shelf-Life Extension; Rapid Test Optimization