Automated 4-Hour Detection of Heteroresistant Vancomycin-Intermediate Staphylococcus aureus (hVISA)

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Background: The hVISA phenotype presents a diagnostic dilemma with infected patients, as standard susceptibility methods frequently underestimate hVISA prevalence. To improve this diagnostic tool, we developed an automated fast screening method. This study presents the evaluation of a multiplexed automated digital microscopy (MADM) system to identify hVISA strains and compare results to the gold standard.

Methods: 30 isolates were characterized by the broth microdilution analysis profile and experiment control software. A PCR cut custom image analysis (CIA) was used to identify organisms using abbreviated population analysis profiles. 2-channel disposable cassettes (Fig. 1) enabled live microbial cell immobilization for microscopy and fluid exchanges for different test media. Automated Staphylococcus aureus (SA) clinical isolates and isolates from a CDC SA collection using 48-hour broth microdilution abbreviated population analysis profiles (BMD-PAP), which served as the control condition.

Results: We screened the reference. The PAP area under the curve (AUC) was calculated and hVISA isolates with 1.0, 2.0, or 4.0 µg/mL dropped onto sectors of VAN agar plates containing from 0 to 6 µg/mL. The MADM-PAP-AUC was censored as a technical error. MADM correctly classified 14/15 hVISA+ isolates characterized by a broth microdilution methods for discrimination between hVISA+ and VSSA strains determined by the plus sign in hVISA+.

Discussion: Growth analysis by MADM revealed an identification criterion for using abbreviated PAPs of individual isolates to screen for VAN susceptibility using an arbitrary units for areas. Normalized data. Horizontal dotted line shows the BACcel criterion AUC level, and vertical dotted line shows BMD-PAP AUC detection criterion.

Conclusion: Automated MADM can identify the hVISA phenotype within 4 hours using a gas exchange film in multiple wells in different VAN concentrations to use for expanded studies. Automated and rapid automation. At its present state, MADM appears applicable for use with clinical isolates to identify hVISA+ within 4 hours. This enables replication with larger screening studies to help estimate phenotype prevalence as well as characterizing statistical performance.

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PAPs for positive strains had down-sloping characteristics of hVISA as did the BMD-PAPs. This study identified a narrow range of VAN concentrations to be used for expanded studies, automated and rapid automation. At its present state, MADM appears applicable for use with clinical isolates to identify hVISA+ within 4 hours. This enables replication with larger screening studies to help estimate phenotype prevalence as well as characterizing statistical performance.

The number of small cells required is also compatible with the number available from lower respiratory tract specimens at the diagnostic threshold. Additional research with polymicrobial samples will determine potential for inclusion in a practical rapid diagnostic system.