Introduction

Early identification of methicillin resistant Staphylococcus aureus (MRSA) and Vancomycin resistant enterococci (VRE) in colonized patients from surveillance cultures results in a reduction of colonization, infection and subsequent morbidity and mortality. By reducing the spread of colonization and infection, the overall financial burden on the healthcare system is also lessened by decreasing the length of hospital stay and potential treatments. Since the implementation of the Duo-ESwab™ kit with 2 swabs and WASP™/WASPLab™ automation in our laboratory, we have developed and validated efficacious cost-effective strategies for the prevention of infections caused by MRSA and VRE. We have validated the use of a Colorex™ MRSA/VRE bi-plate (CHROMagar™) using a 1ul dual loop to seed plates on the WASP™ and incubate and analyze plates on the WASLab™. The objective of this study was to validate MRSA – VRE bi-plates for MRSA/VRE screening and the use of the WASP™/WASPLab™ to process and perform imaging analysis.

Materials and Methods

In this study 359 clinical MRSA/VRE specimens were collected with ESwab™ duo swabs kits and processed with our current method using MRSA Select II and VRE CHROMagar with a 30ul inoculum on the WASPLab™. For those same specimens, a new protocol was implemented on the WASP using a 1ul dual loop and a twin loop 2/bi plate streaking pattern for MRSA/VRE CHROMagar bi-plates. An additional 95 specimens were run on the WASP™ using a 10ul loop with a Vertical biplate 2 streaking pattern. After processing, the biplates were incubated in the WASLab™ for 20 hours at which point digital imaging analysis on the WASP Lab was performed. MRSA and VRE target colour colonies were confirmed with LAMP and dd1 identification genes. Vitek MS was performed on non-target colour colonies. Results were compared to routine testing which uses a 30 ul loop on the WASP™.

Results

Of the 359 duo swabs tested, 112 were positive for MRSA and 32 for VRE by our current 2 plate method. With our study protocol using a 1 ul dual loop on the WASP™/WASPLab™ and CHROMagar MRSA-VRE biplates, 110 samples were positive for MRSA on the bi-plates and 32 positives for the VRE side of the bi-plate. 24 samples isolated non-target colour colonies which identified as S. haemolyticus. The two false negatives had only a few colonies on the WASLab™ images with the current method, which confirmed positive when repeated using the 10ul loop on a bi-plate. All results were confirmed with LAMP assays. In the 95 specimens tested using the 10ul loop on a bi-plate, 76 were positive for MRSA and 26 were positive for VRE showing 100% specificity and sensitivity. Colonies growing <10 on the VRE side showed 100% correlation with current imaging analysis on WASPLab™.

Conclusion

Results showed excellent correlation between the current testing method using the 30ul loop on a full CHROMagar plates versus a 10 ul loop on a bi-plate. A timing study was performed with the different protocols on the WASP™. Implementing a protocol with a 10ul loop on a bi-plate results in a 60% reduction of current streaking time on the WASP™. The Hamilton microbiology laboratory currently processes more than 100,000 MRSA/VRE samples annually. Using the duo swabs with the bi-plates reduces the number of specimens and plates by 50% thus doubling the physical space capacity. The reduction in streak time and image analysis will increase the functional capacity of the whole WASP™/WASPLab™ system leading to higher throughput.