3035 Evaluation of Two Types of Chromogenic Media for the Detection of MRSA in Wound Swabs

W. Hadley¹, R. Clasen¹, J. Knox¹, J. Eddins², D. Halstead³, M. Sarina⁴, J. Hardy¹; ¹Hardy Diagnostics, Santa Maria, CA, ²Community Hospital, Grand Junction, CO, ³Baptist Health, Jacksonville, FL, ⁴Central Coast Pathology Consultants, San Luis Obispo, CA.

Revised Abstract

In this study HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA media were compared for sensitivity, specificity, color intensity, and time of growth in the identification and detection of clinical MRSA strains. For the purpose of this study 328 clinical wound swabs were collected in duplicate from patients suspected to have MRSA. In order to perform comparison testing, a HardyCHROM[™] MRSA and a BBL[™] CHROMagar[™] MRSA plate were inoculated with each of the clinical swabs. Both manufacturer's plates were allowed to incubate for 24 hours and were then evaluated for positive colonies typical of MRSA growth. Any plates that were negative for MRSA were re-incubated for a total incubation time of 48 hours. Confirmatory testing using a PBP2' latex kit as well as the cefoxitin disk method recommended by the Clinical Laboratory Standards Institute (CLSI) was performed on all MRSA positive samples to verify results.

The percentage of positive cultures for HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA at 24 hours was 28.4% and 28.0% respectively. After 24 hours of incubation the sensitivity rate for HardyCHROM[™] MRSA was 97.9% with a specificity rate of 100%, and the sensitivity rate for BBL[™] CHROMagar[™] MRSA was 96.8% with a specificity rate of 100%. The percentage of positive cultures for HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA at 48 hours was 31.4% and 30.8% respectively. After 48 hours of incubation the sensitivity rate for HardyCHROM[™] MRSA was 97.2% with a specificity rate of 96.6%, and the sensitivity rate for BBL[™] CHROMagar[™] MRSA was 95.3% with a specificity rate of 97.4%.

BBL[™] CHROMagar[™] MRSA media failed to detect three positive MRSA strains that were correctly identified on the HardyCHROM[™] MRSA plates within 24 hours. In comparison HardyCHROM[™] MRSA failed to detect two MRSA strains that were detected on BBL[™] CHROMagar[™] within 24 hours. On both manufacturers' media, eight plates were found to have false positives after 48 hours, which suggests that confirmatory testing should be done on colonies suspected to be MRSA after 48 hours of incubation. While detection rates were similar between both types of media several laboratory technicians expressed a preference for the HardyCHROM[™] MRSA plates based on the cultures producing brighter and more distinguishable colonies within 24 hours. In comparison, the BBL[™] CHROMagar[™] MRSA plates took longer for color development and tended to have less brilliant coloration even after a full 48 hour incubation. Based on these results the HardyCHROM[™] MRSA medium was found to be a reliable growth media for the detection of clinical MRSA strains from wound cultures within 24 hours.

Introduction

In the United States there continues to be an increasing incidence of nosocomial and community acquired infection caused by methicillin-resistant Staphylococcus aureus (MRSA). MRSA strains are often associated with skin infections occurring after accidental injury or surgery, which often results in high morbidity and mortality. For this reason, it is essential to have an effective and reliable test for methicillin-resistant strains to ensure proper antibiotic therapy and infection control.

In this study, HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA were compared for sensitivity, specificity, color intensity, and time of growth for clinical MRSA strains. Both of these products are selective and differential chromogenic media. The chromogens in these media formulations release chromophores when cleaved by enzymes that are produced by MRSA strains. Based on colony color, HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA allow for the reliable detection of methicillin-resistant *S. aureus* from clinical specimens within 24 to 48 hours. Non-MRSA strains are either inhibited by the addition of selective agents or utilized different chromogenic substrates in the media to produce different colored colonies. If none of the substrates are utilized, natural or white colored colonies will be present.

Material and Methods

In this study, 328 clinical wound swabs were collected and cultured by lab personnel affiliated with the Central Coast Pathology Consultants in San Luis Obispo, California, the Community Hospital in Grand Junction, Colorado, and the Baptist Health facility in Jacksonville, Florida. Whenever possible, two swabs were collected from each participating patient. However in situations where this was not possible a single swab was collected and emulsified in 0.5mls of 0.85% saline. The saline suspension was used to inoculate HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA plates to ensure similar inoculum levels. The chromogenic plates were incubated for 24 hours at 35°C and evaluated for the presence of MRSA colonies. Any negative chromogenic plates were held for an additional 24 hours for a total incubation time of 48 hours. Confirmatory testing using a PBP2' latex kit as well as the cefoxitin disk method recommended by the Clinical Laboratory Standards Institute (CLSI) was performed on all MRSA positive samples to verify results.

Results and Data Analysis

A total of 328 clinical wound swabs were cultured on both HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA. 93 cultures were positive on HardyCHROM[™] MRSA within 24 hours and 92 cultures were positive on BBL[™] CHROMagar[™] MRSA plates; three positive cultures were detected only on HardyCHROM[™] MRSA, and two positive cultures were detected only on BBL[™] CHROMagar[™] MRSA after 24 hours of incubation at 35°C (Table 1). This data corresponds into a 28.4% positivity rate for HardyCHROM[™] MRSA and a 28.0% for BBL [™] CHROMagar[™] MRSA for the samples tested.

The sensitivity of HardyCHROM[™] MRSA within 24 hours was 97.9% with a specificity rate of 100%, and the sensitivity for BBL[™] CHROMagar[™] MRSA was 96.8% with a specificity rate of 100%. The percentage of positive cultures for HardyCHROM[™] MRSA and BBL[™] CHROMagar[™] MRSA at 48 hours was 31.4% and 30.8% respectively. After 48 hours of incubation the sensitivity rate for HardyCHROM[™] MRSA was 97.2% with a specificity rate of 96.6%, and the sensitivity rate for BBL[™] CHROMagar[™] MRSA was 95.3% with a specificity rate of 97.4%

On both manufacturers' media eight plates were found to have false positives after 48 hours, which suggests that confirmatory testing should be done on colonies suspected to be MRSA after 48 hours of incubation (Table 2).

Table 1: Summary of Comparison Testing Between HardyCHROM[™] MRSA with BBL[™] CHROMagar[™] MRSA within 24 hours

		BBL [™] CHROMagar [™] MRSA Results (24 hours)		Total
		positive	negative	
HardyCHROM [™] MRSA (24 hours)	positive	90	3*	93
	negative	2*	233	235
TOTAL		92	236	328

* Two true positives detected on BBL™ ĆHROMagar™ MRSA were not detected on HardyCHROM™ MRSA

HardyCHROM[™] MRSA Results at 24 hours

Sensitivity within 24 hours 93/95 = 97.9%

Specificity within 24 hours 235/235 = 100%

Table 2: Summary of Comparison Testing Between HardyCHROM™ MRSA with BBL™ CHROMagar™ MRSA within 48 hours

		BBL [™] CHROMagar [™] MRSA Results (48 hours)		Total
		positive	negative	
HardyCHROM [™] MRSA (48 hours)	positive	98*	5**	103
	negative	3**	222	225
TOTAL		101	227	328

HardyCHROM[™] MRSA Results at 48 hours

Sensitivity within 48 hours 103/106 = 97.2%Specificity within 48 hours 227/235 = 96.6% BBL[™] CHROMagar[™] MRSA Results at 24 hours Sensitivity within 24 hours 92/95 = 96.8% Specificity within 24 hours 236/236 = 100%

USITIVES WERE DETECTED OF DDE CHINOMUQUE MINSA WERE NOT DETECTED OF TUTUYCHNOM MINSA

BBL[™] CHROMagar[™] MRSA Results at 48 hours Sensitivity within 48 hours 101/106 = 95.3%

Specificity within 48 hours 229/235 = 97.4%

Discussion

Based on these results, HardyCHROM[™] MRSA demonstrated a slightly higher positivity rate for detecting clinical MRSA strains in comparison with BBL[™] CHROMagar[™] MRSA within 24 hours. After 48 hours of incubation there was a 98% agreement (n = 101/103) between the two types of chromogenic media used in this study. Five MRSA strains positively identified on HardyCHROM[™] MRSA were not recovered on BBL[™] CHROMagar[™] MRSA, and three MRSA strains positively identified on BBL[™] CHROMagar[™] MRSA were not detected on HardyCHROM[™] MRSA after 48 hours of incubation.

On both manufacturers' media, eight plates were found to have false positives after 48 hours, which suggests that confirmatory testing should be done on colonies suspected to be MRSA after 48 hours of incubation. Confirmatory testing with a PBP2' latex kit as well as the cefoxitin disk method confirmed that all other strains identified as MRSA on both chromogenic media were true positives. The majority of technicians working with the two different brands of media reported that there was brighter colony coloration and faster growth rates on HardyCHROM[™] MRSA in comparison with the BBLTM CHROMagarTM MRSA.

Conclusion

The accurate identification of MRSA positive wound swabs was found to be more reliable on the HardyCHROM[™] MRSA media within 24 hours in comparison with BBL™ CHROMagar™ MRSA media. The faster detection time for the HardyCHROM[™] MRSA media demonstrates that the majority of clinical MRSA strains can be quickly and reliably detected within 24 hours. This advantage helps to streamline the identification process and allows for faster and more appropriate drug therapy to be implemented for patients afflicted with this pathogen.

References

- Weissfeld, American Society for Microbiology, Washington, D.C.



. August, M.J., et al, 1990. Cumitech 3A; Quality Control and Quality Assurance practices in Clinical Microbiology, Coordinating ed, A.S.

HARDY DIAGNOSTICS

A Culture of Service™

2. Murray, P.R. et al. 2003. Manual of Clinical Microbiology, 8th ed. American Society for Microbiology, Washington, D.C.

3. Forbes, B.A., et al. 2002. Bailey and Scott's Diagnostics Microbiology, 11th ed. C.V. Mosby Company, St. Louis, MO.

4. Isenberg, H.D. Clinical Microbiology Procedures Handbook, Vol I & II. American Society for Microbiology, Washington, D.C.

5. Koneman, E.W. et al. 1997. Color Atlas and Textbook of Diagnostics Microbiology, 5th ed. J.B. Lippincott Company, Philadelphia, PA.