Comparison of BBL™ CHROMagar™ Staph aureus to other Commonly Used Media for the Presumptive Identification of *Staphylococcus aureus*

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**ABSTRACT (revised)**

BBL CHROMagar Staph aureus is a modification to the original CHROMagar Staph aureus (CHROMagar Company, Paris). This chromogenic medium is intended for the isolation and differentiation of *S. aureus* based on the formation of an insoluble pigment. *S. aureus* will produce mauve colored colonies while other organisms are inhibited or produce a distinctly different colony color. The following media were compared for recovery and presumptive identification: BBL CHROMagar Staph aureus (CSA), Mannitol Salt Agar (MSA), and Baird Parker Agar (BPA). Seventy nine strains of *S. aureus*; 40 coagulase-negative-staphylococci; and 11 other organisms, including streptococci, *Enterobacteriaceae*, and *Candida* were evaluated at 18, 24 and 48 hours. *S. aureus* strains were inoculated at a concentration of $10^3$ CFU and all other organisms at a concentration of $10^5$ CFU. At 18 hours, CSA presumptively identified 59 of 79 *S. aureus* (sensitivity, 74.7%), MSA identified 58 of 79 (sensitivity, 73.4%), BPA presumptively identified 15 of 79 (sensitivity, 19.0%). Following 48 hours incubation all sensitivities increased: CSA, 98.7%, MSA, 93.7%, and BPA, 84.8%.

False positives varied by media type. At 18 hours, CSA and BPA demonstrated a specificity of 98.0% and 100%, respectively. MSA had a specificity of 56.9%. At 48 hours, *S. cohnii* produced a false positive result on all 3 media; however, on repeat testing only MSA produced a false positive. BBL CHROMagar Staph aureus provides greater sensitivity and comparable or greater specificity, as compared to MSA and BPA. CSA requires little subjective interpretation. MSA and BPA produce a greater frequency of false positive and questionable results thus increasing the need for supplemental testing or prolonged incubation. BBL CHROMagar Staph aureus also provides more reliable results at 18 and 24 hours.

**INTRODUCTION**

*Staphylococcus aureus* is a well documented pathogen. It is responsible for infections of three general types: superficial, life threatening systemic, and toxinoxis, including food poisoning and toxic shock. Due to the clinical implications and severity of infection associated with *S. aureus,* as well as the potential economic impact on contaminated foods, the isolation and identification of this organism is of utmost importance.

BBL CHROMagar Staph aureus (CSA) is a selective and differential media that has applications in clinical and food microbiology. CSA is composed of a nutritive base that is made selective by the addition of colistin, nalidixic acid and an anti-fungal agent. A proprietary blend of chromogens offers differentiation of *S. aureus* from other organisms. CSA was evaluated against Mannitol Salt Agar (MSA) and Baird Parker Agar (BPA). MSA and BPA are popular selective and differential media; MSA is primarily used in clinical and BPA in food and environmental microbiology.

This study compared recovery and ability to presumptively identify *S. aureus* following 18h, 24h and 48h incubation.
Recovery

Overall recovery of *S. aureus*, independent of color reaction, at 18h was comparable or superior on CSA compared to the other media, isolating 78 of 79 strains (98.7%). MSA recovered 77 of 79 (97.5%), and BPA recovered 75 of 79 (94.9%) at 18h. Following 24h incubation, CSA recovered all 79 strains (100%), MSA recovered 78 of 79 (98.7%), and BPA recovered 75 of 79 (94.9%). MSA and BPA recovered 100% of the strains following 48h incubation.

CSA also provides greater recovery with regards to quadrant of growth at each incubation interval. The quadrant of growth on the CSA media compares closely with that of the nonselective control, TSAII.

Presumptive Identification

Organisms were identified based on production of a mauve colony coloration on the CSA media, yellow zones on MSA and black with clear zones on BPA. Only distinct color reactions for each media type were recorded as positive for identification. Colonies producing questionable coloration, e.g. pale mauve (CSA), red/yellow (MSA) and black w/no clear zones (BPA) were scored as negative reactions.
In general, sensitivities increased with increased incubation time. Following 18h, CSA had a sensitivity of 74.7%, MSA 73.4% and BPA 19.0%. At 24h incubation, the sensitivity for CSA was 89.9%, MSA 87.3% and BPA 48.1%. Percentage of isolates identified again increased following 48h incubation: CSA 98.7% MSA 93.7% and BPA 84.8%.

Specificity decreased slightly over time with all media, except MSA, which demonstrated poor specificity in general. BPA had a specificity of 100% at 18h and 24h; however, many of the organisms scored as negative produced distinct black colonies with no clear zone. These zones were often difficult to detect and subjective. The slight decrease in specificity of the CSA media is attributed to an increase in questionable results. Of the 51 non S. aureus strains tested, CSA produced questionable results with 1 organism at 18h and a total of 4 organisms at 48h. These questionable reactions appear as a very pale mauve coloration in areas of heavy growth; however, isolated colonies remained white or colorless.

Organisms known to produce false positives on CSA include *C. albicans* and *S. cohnii*. Previous testing of *S. cohnii* produced a false positive on all media at 48h. Repeat testing produced questionable results on CSA at 18, 24 and 48 hours and a questionable result on BPA at 48h. MSA was positive at 18h. Additional strains will need to be evaluated to determine if this reaction is organism or strain dependent. It has been demonstrated that some strains of *C. albicans* may produce mauve colored colonies on CSA. The incorporation of an antifungal agent into the BBL formulation has eliminated this potential false positive.

The overall accuracy of CSA is perhaps the most noteworthy. CSA had an accuracy of 83.9%, 91.5% and 96.2% at 18h, 24h and 48h, respectively. MSA had an accuracy of 66.9%, 76.2% and 84.6% and BPA an accuracy of 50.8%, 68.5% and 90.0%.
CONCLUSIONS

- BBL CHROMagar Staph aureus offers a distinct advantage over Mannitol Salt Agar and Baird Parker Agar in overall recovery, sensitivity and accuracy. The specificity of CSA was superior to MSA and comparable to BPA. False positives and questionable results are major short-comings with MSA and BPA, respectively. The color production on CSA is more easily interpreted, identifying more strains of S. aureus than other related media.

- BBL CHROMagar Staph aureus demonstrates superior performance, providing a more rapid, reliable identification at 18h and 24h.