

Digital Detection of Group A Streptococcus using Colorex Strep A CHROMagar and WASPLab Chromogenic Detection Module

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BACKGROUND:

- Despite the availability of several diagnostics tools for the diagnosis of Group A *Streptococcus* (GAS) pharyngitis, culture remains one of the primary methods in use today and is still considered the gold standard for the detection of GAS from pharyngeal samples.
- However, in larger volume laboratories, screening for GAS by culture can be cumbersome and streamlined approaches using automated plating instrumentation, smart incubation and image analysis could be helpful.
- This study evaluates the capability of the WASPLab™ Total Laboratory Automation System (TLA) (Figure 1) (Copan Diagnostics, Murrieta, CA) PhenoMATRIX™ Chromogenic Detection Module (CDM) to automatically detect and interpret orange GAS colonies on a novel chromogenic agar called Colorex Strep A Agar (CHROMagar, Paris, France) To date, the Colorex Strep A Agar is not Food and Drug Administration (FDA) cleared.
- There have been 2 excellent published studies demonstrating the ability of the WASPLab™ CDM software in detecting and sorting positive and negative cultures based on pigmentation production on chromogenic media^{1,2}.

MATERIALS/METHODS:

- 250 remnant pharyngeal samples collected from pediatric patients during the period of September 2017 through January 2018 were enrolled in the study.
- Samples were initially tested for the presence of GAS by PCR (Lyra Direct Strep Assay) at Children's Hospital Los Angeles
- Remnant samples, either positive or negative for GAS, were inoculated onto blood agar plate (BAP) and a Colorex Strep A agar and incubated in the WASPLab™ for 24 hours.
- After 24 hours, plates were examined for presence of orange colonies on the Colorex Strep A agar (Figures 2 & 3).
- Results were compared to the results from the PhenoMATRIX Chromogenic Detection Module (CDM).
- Orange colonies observed on Colorex Strep A agar were confirmed via Matrix Assisted Laser Desorption/Ionization – Time of Flight Mass Spectrometry (MALDI-TOF MS).
- Performance of the Colorex Strep A Agar was also compared to growth on the BAP.

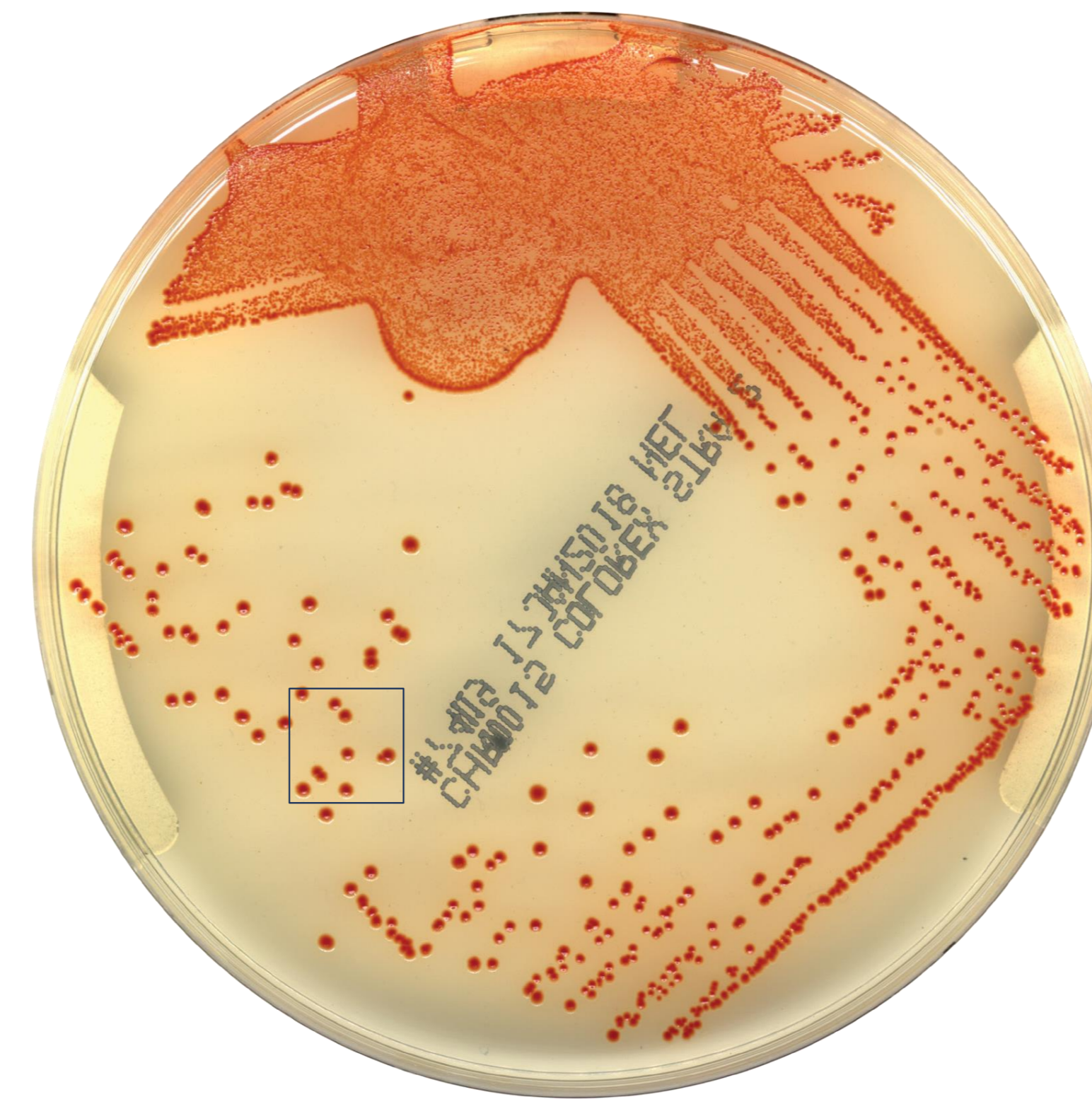


Figure 2. Colorex Strep A Agar, CHROMagar. Orange colonies represent Group A Streptococcus.

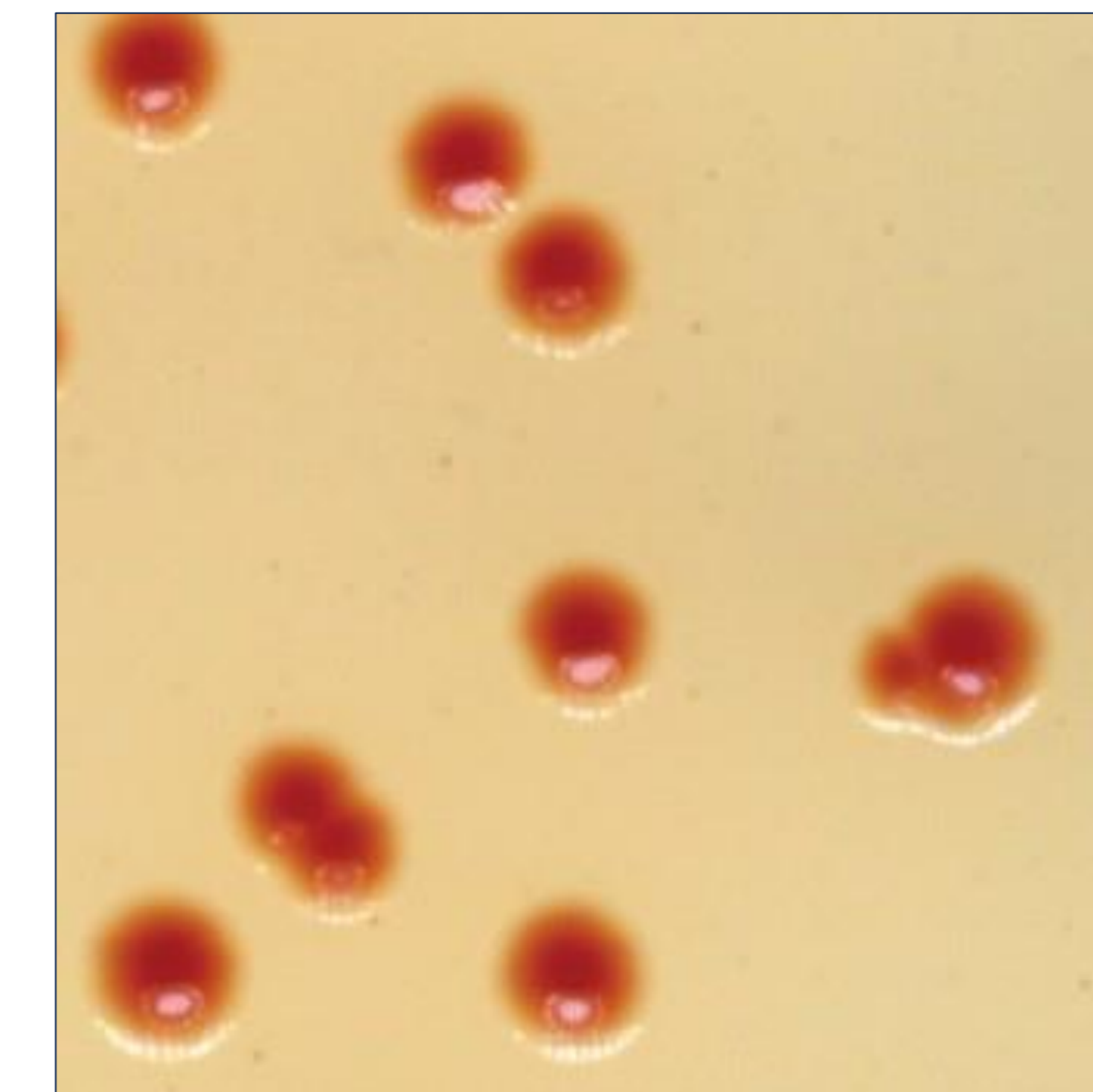


Figure 3. Colorex Strep A Agar, CHROMagar. Orange colonies represent Group A Streptococcus (enlarged view of orange colony)

RESULTS:

- 250 cultures were manually examined by medical technology staff after 24 hours of incubation.
- Plates examined by the CDM software after secondary manual review had a sensitivity of 100% and a specificity of 96.4% (Table 1).
- Plates examined manually by technologist (not assisted by CDM software) after secondary manual review had a sensitivity of 96.5% with a specificity of 100% (Table 2).
- Of the 57 cultures that grew orange colonies on the Colorex Strep A Agar, 51 were confirmed as GAS by MALDI-TOF MS.
- 6 cultures grew orange colonies; but 4 cultures were not identified as GAS and unfortunately, 2 cultures were not tested by MALDI-TOF MS.
- 5 specimens that were initially PCR positive did not grow in culture on the Colorex A Strep Agar.
- Isolation of GAS in blood agar plates had a sensitivity of 78.9% and specificity of 73.6% (Table 3).

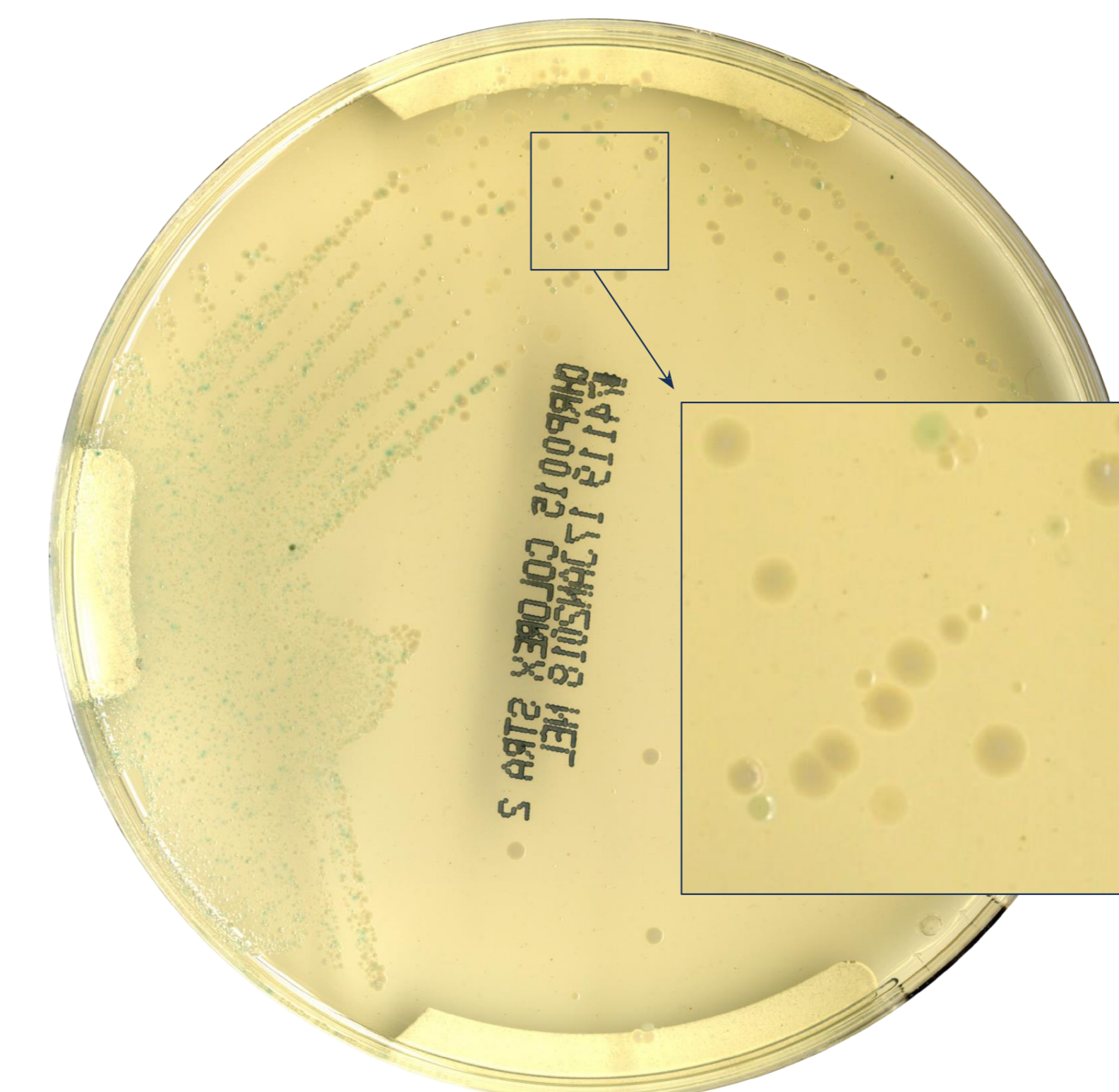


Figure 4. Colorex Strep A Agar, CHROMagar. Colorless colonies are non-GAS.

Table 1. WASPLab examination of Colorex Strep A agar after 24 hours incubation using CDM software with secondary manual review

		Orange Colony	
		Pos	Neg
CHROMagar at 24 h (CDM algorithm)	Pos	57	7
	Neg	0	186
		Sensitivity: 57/57 + 0 = 100%	
		Specificity: 186/186 + 7 = 96.4%	

PPV = 57/57 + 7 = 89.1%; NPV = 186/186 + 0 = 100%

Table 2. Manual examination of Colorex Strep A Agar after 24 hours incubation with secondary manual review

		Orange Colony	
		Pos	Neg
CHROMagar at 24 h (visual)	Pos	55	0
	Neg	2	193
		Sensitivity: 55/55+2 = 96.5%	
		Specificity: 193/193+0 = 100.0%	

PPV = 55/55 + 0 = 100%; NPV = 193/193+2 = 98.9%

Table 3. Comparison of manual examination of BAP versus Colorex Strep A Agar (with secondary manual review)

		Orange Colony	
		Pos	Neg
Beta Hemolysis Present on BAP	Pos	45	51
	Neg	12	142
		Sensitivity: 45/45 + 12 = 78.9%	
		Specificity: 142/142 + 51 = 73.6%	

CONCLUSION:

- This proof of concept study validates the ability of the CDM software in the detection of GAS using a novel CHROMagar, Colorex Strep A Agar.
- This study also demonstrated that the Colorex Strep A Agar has good sensitivity and specificity compared to an FDA cleared PCR assay for GAS and better overall performance compared to culture on a BAP
- The WASPLab™ CDM Software in combination with the Colorex Strep A Agar has the ability to dramatically improve workflow by reducing turn-around-time and redirecting laboratory personnel to other more complex tasks.

REFERENCES:

- Faron et al., 2016. Automatic Digital Analysis of Chromogenic Media for Vancomycin Resistant Enterococcus Screens Using Copan WASPLab. J Clin Microbiol. 54:2464 – 2469
- Faron et al., 2016. Automated Scoring of Chromogenic Media for Detection of Methicillin-Resistant Staphylococcus aureus by use of WASPLab Image Analysis Software.



Figure 1. The WASPLab - Total Laboratory Automation system consisting of the WASP (Walk Away Specimen Processor) plating instrument, track for moving petri dishes, and a smart incubator with a 27-megapixel camera for imaging analysis.

