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Rapid and efficient approach to urine culture screening using CHROMagar Orientation/ESBL medium



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Fig. 4. Laboratory workflow

Introduction:

CHROMagar Orientation (CHO)/ESBL (Kanto Chemical, Japan) is a bi-plate consisting of CHO medium and CHROMagar ESBL (Fig. 1) for the differentiation and presumptive identification of clinically important gramnegative bacteria (Fig. 2) and detection of extendedspectrum β-lactamase (ESBL) producers (Fig. 3). We report a rapid and efficient approach to urine cultures using CHO/ESBL medium.

Fig. 1. CHROMagar orientation/ESBL medium



Fig. 2. Colonies of selected organisms plated on **CHROMagar Orientation medium**

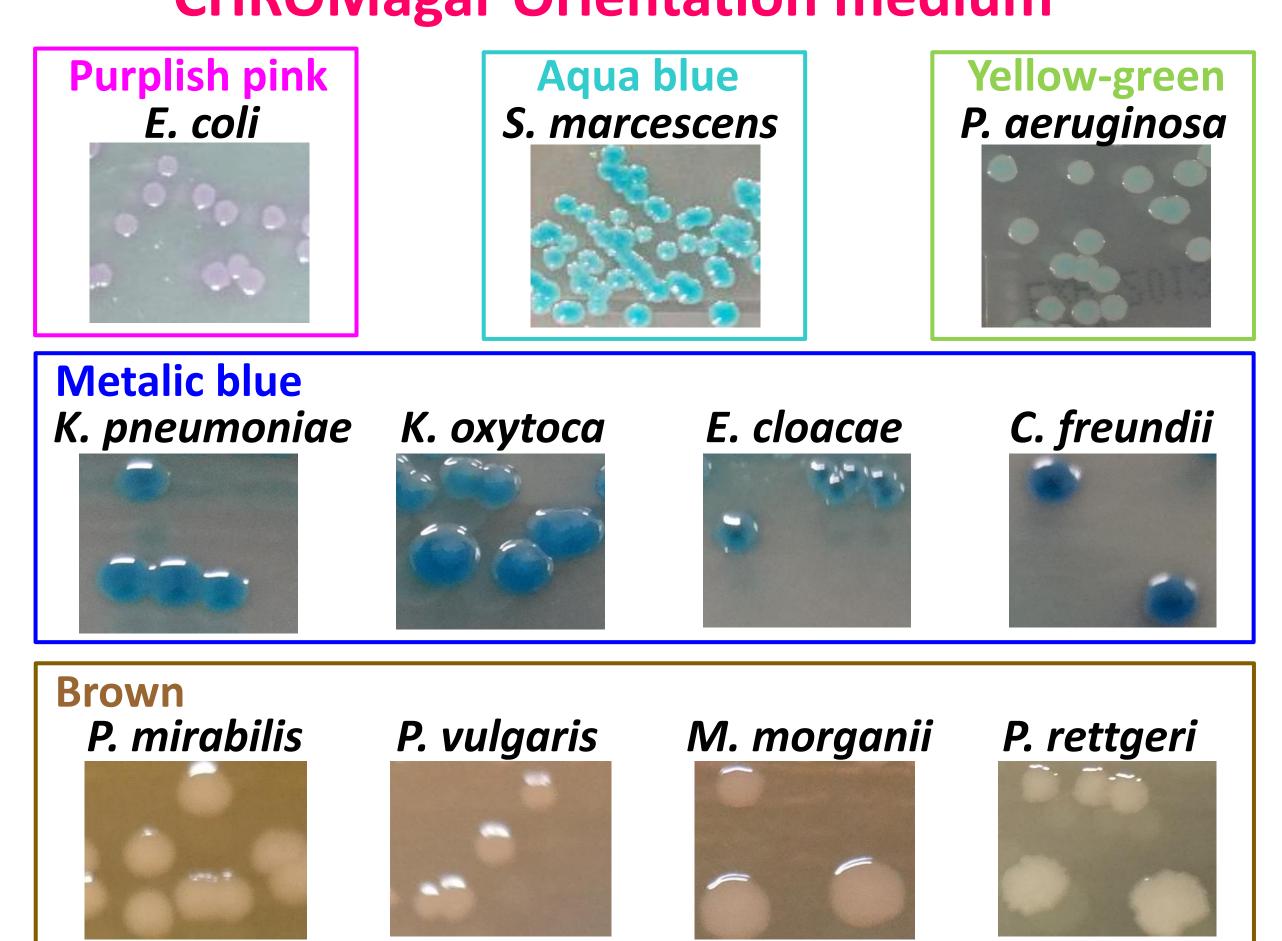
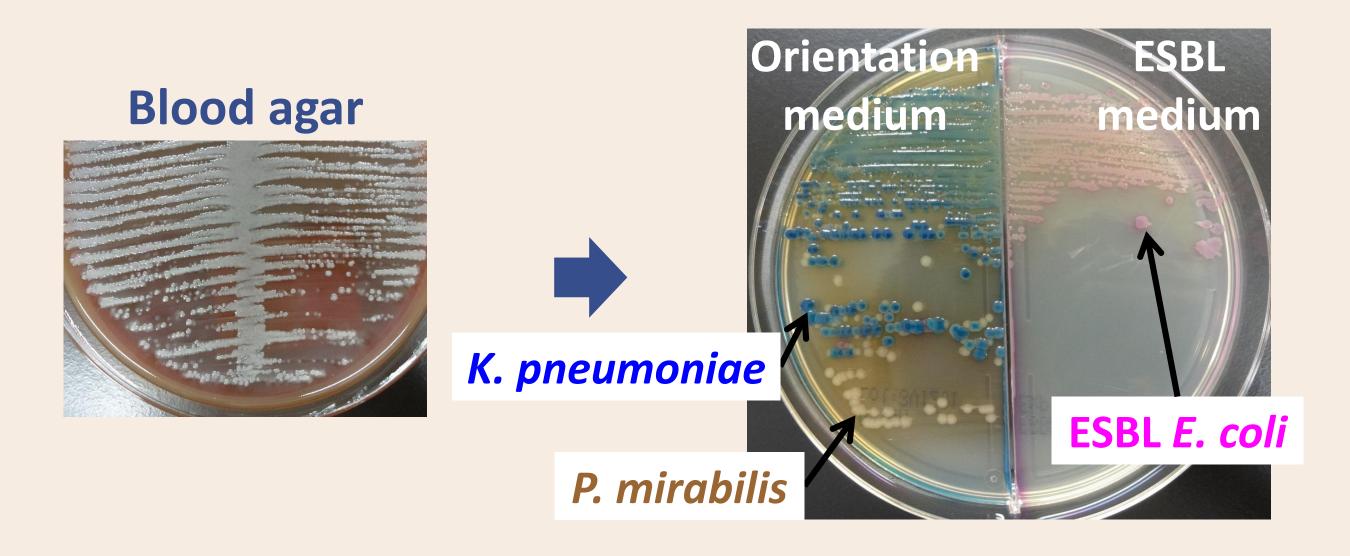


Fig. 3. GNB in mixed cultures of clinical urine sample



Methods:

A total of 1995 urine culture results generated during the months of February, April, July and October of 2012 and 2015, were selected at random for comparison of the turnaround time. In comparing time to result between conventional urine culture and improved urine culture method, 1073 and 922 reports from 2012 and 2015, respectively, were examined. In the conventional method, 10 μ l of urine was streaked on a sheep blood agar plate. A sample of the urine was also prepared for gram-staining. In the improved method, in addition to plating of 10 μ l of urine on sheep blood agar, the urine sample was centrifuged at 1,500 g for 10 min. The supernatant was discarded and the pellet was smeared onto a glass slide for gram staining. If gram-negative organisms were observed, a CHO/ESBL medium was added (Fig. 4).

Conventional method Improved method Streaked 10 μl of urine Streaked plated onto on a plate on a plate a sheep blood agar Centrifuged at 1,500 g for Day 1 10 minutes 10 μl of urine was The pellet was Gram Gram prepared onto smeared onto staining a glass slide a glass slide Streaked CHO/ESBL on a plate medium AST * Day 2 AST ' Confirmatory Test for ESBLs Confirmatory Day 3 Final report Final report

Test for ESBLs

Final report

ID*: identification, AST **: antimicrobial susceptibility testing

Results:

The mean turnaround time was 4.1 days for the conventional method compared to 3.9 days with the improved method. When ESBL-producing organisms were present in the urine specimen, the mean turnaround time was 4.6 days compared to 3.5 days with the improved method. With respect to negative urine cultures, the turnaround time was 3.2 days compared to 2.4 days with the improved method (Table 1).

Table 1. The mean time to reporting (2012 vs 2015)

2012 (Conventional method)									
No. of Urine sample		The mean time to reporting (day)							
		Total	positive	negative	ESBL-producing				
			cultures	cultures	organisms				
February	263	3.5	4.1	3.2	3.8				
April	247	3.7	4.2	3.4	6.0				
July	296	3.5	4.0	3.2	4.3				
October	267	3.5	4.3	3.1	4.4				
Total	1073	3.6	4.1	3.2	4.6				

2015 (Improved method)								
No. of Urine sample		The mean time to reporting (day)						
		Total	positive	negative	ESBL-producing			
			cultures	cultures	organisms			
February	221	3.2	4.2	2.7	3.3			
April	234	3.0	3.7	2.4	3.0			
July	242	3.1	4.0	2.4	4.0			
October	225	2.8	3.7	2.2	3.6			
Total	922	3.0	3.9	2.4	3.5			

Conclusions:

Day 4

The use of CHO/ESBL bi-plates allows differentiation of bacteria the day after plating and providing information on the presence or absence of ESBL-producing organisms. As this medium allows confirmatory testing of resistant organisms to be setup at the same time as antimicrobial susceptibility testing, CHO/ESBL allows reporting of resistant organisms 1 day earlier compared to conventional methodology. Furthermore, the staining of centrifuged urine sediments has reduced false negative microscopies, allowing release of final negative culture reports on the next day as well as workup of cultures with a cutoff of 10³cfu/ml for specimens showing >5 WBC/hpf. The improved efficiency contributes to a rapid report with clinically useful information.