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Validation of an Assessment of Chromogenic Media Against Conventional Culture Techniques for Isolation, Identification, and Direct Antibiotic Susceptibility Testing of Uropathogens in Resource-Poor Settings

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Abstract

The study was planned to evaluate chromogenic media against conventional techniques in terms of correct identification, ease of reporting, and reduction in cost. This cross-sectional prospective analytical study was carried out from January 1, 2022, to September 30, 2023, at Vedantaa Institute of Medical Sciences, Palghar. Urine samples were inoculated on MacConkey agar, blood agar, CLED agar, and chromogenic media simultaneously and incubated overnight. Imparting a distinct color to the isolated bacterial colony was visualized and identified by Hichrome UTI agar. Direct susceptibility testing by disc diffusion on clinical samples offers a rapid and inexpensive method of obtaining information to guide antimicrobial therapy. A total of 531 samples were culture-positive clinical isolates. Significant growth was obtained in 531 (100%) plates of HiCrome UTI, followed by CLED agar 521 (98.11%), Blood agar 511 (96.23%), and MacConkey agar 497 (93.59%). The sensitivity patterns of S. aureus to the following antibiotics: gentamicin, tetracycline, penicillin, and levofloxacin were 12 (85.71%), 11 (78.57%), 9 (64.28%), and 7 (7.00%), respectively. The sensitivity patterns of Pseudomonas spp. to the following antibiotics: piperacillin-tazobacta, amikacin, ceftazidime-avibactam, and gentamicin were 62 (87.32%), 43 (60.56%), 35 (49.29%), and 21 (29.57%). Enterococcus spp. was sensitive to Penicillin 79 (94.94%), Ampicillin 68 (80.95%), Vancomycin 57 (67.85%), and Tetracycline 46 (54.76%). Enterobacteriaceae showed high sensitivity to Meropenem (84.25%), Ceftazidime (67.68%), Piperacillin-Tazobactam (55.80%), and Ampicillin-Sulbactam (41.44%). Concluded that the Hichrome agar medium can be a desirable, simple primary isolation and identification medium that significantly lessens the daily workload associated with urine culture in microbiology laboratories.

Keywords

Bacterial growth, CLED agar, Conventional culture system, Hi Chrome UTI agar, Urinary tract infection

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INTRODUCTION

Urinary tract infections (UTIs) are among the most common bacterial infections worldwide, affecting all age groups in community and hospital settings (Ali *et al*., 2023; Harris and Fasolino, 2022). They represent 10–20% of all infections treated in primary care and 30–40% of infections treated in hospitals (Nahar *et al*., 2020). It is common in females due to the short urethra, and about 60% of women are reported to have experienced the UTI once in their lifetime. The increasing risk factor in females is due to short urethra, pregnancy, and the absence of prostatic secretions, which easy contamination of the urinary tract. 90% of pregnant women develop urethral dilation and are susceptible to UTIs. In the need to confirm the diagnosis and manage the patient, a microbiological diagnosis is a must (Jain *et al.*, 2023; Bajoria *et al*., 2019).

Urine samples constitute a large share of the daily workload of microbiological laboratories (Stefaniuk, 2018). For many years, blood agar, CLED agar (cystine-lactose-electrolyte-deficient agar), and MacConkey agars have been used for the detection of urinary tract pathogens as well as for differentiation. These current detection methods are time-consuming and require at least 24 hours for a negative culture and up to three days for susceptibility data if positive cultures are discovered (Jadhav *et al*., 2016; Bouslah *et al*., 2020).

The problem of urine culture has been addressed by the introduction of chromogenic agar medium, which is commercially available. CA medium is increasingly being used as a versatile tool in the early differentiation and identification of bacterial isolates from clinical specimens (Sachu and Samuel, 2022). Chromogenic Media: chromogenic substrates are specifically broken down by the enzyme possessed by the particular bacteria, thus imparting a distinct color to the isolated bacterial colony, which can be visualized and identified. Chromogenic media have shown advantages, which include a 20% reduction in time for identification, a reduction in workload, easier recognition of mixed growth, and a reduction in the number of biochemical tests performed for the identification of bacteria, which ultimately results in cost reduction (Khalid *et al*., 2021; Akter *et al*., 2014). The present study was undertaken to evaluate and compare the effectiveness of conventional culture systems and chromogenic agar medium for the isolation, identification, and direct testing of uropathogens' antibiotic susceptibility. Which will help reduce the cost in resource-poor settings.

MATERIALS AND METHODS

The study comprised 1289 clinically suspected acute UTI patients of all ages and gender. This study was undertaken at the Vedantaa Institute of Medical Sciences, Vedantaa Hospital and Research Centre, Dahanu, Palghar, situated in a rural setting. The study was carried out to compare the utility of the conventional method with HiCrome UTI Medium for the detection of uropathogens. It was conducted between 1 January 2022 and 30 September 2023 (Jain *et al*., 2023).

All clinically suspected cases of urinary tract infections admitted to this hospital were included in the study. Either clean mid-stream urine samples or from the sampling port of an indwelling catheter with a sterile syringe and needle were collected. Criteria of inclusion for those indoor patients with UTI were the following: a temperature greater than 38°C, the presence of plenty of pus cells or a high-power field in urine samples, >105 colony-forming units per ml of uncentrifuged urine (Bose, 2015).

The urine samples were inoculated on MacConkey's agar, Blood agar and CLED agar (cystine-lactose-electrolyte-deficient agar). The inoculated plates were incubated aerobically at 37°C for 24 hours. In the conventional method, *Enterobacteriaceae* isolates were identified by the following biochemical tests: motility, indole production, nitrate reduction, glucose fermentation with or without gas production, hydrogen sulfide production, urea hydrolysis, lysine and ornithine decarboxylation, and sodium citrate utilization. Gram-negative microorganisms other than *Enterobacteriaceae* were identified by colony morphology, pigment production, gelatin, and the catalase test. The oxidase test was performed for non-fermenters. Staphylococcus aureus and coagulase-negative Staphylococci were identified by Gram staining, catalase, and coagulase tests, and novobiocin susceptibility (Ali *et al*., 2023). *Streptococci* were identified up to species level by PYR test, the bacitracin sensitivity test, the bile solubility test, and the aesculin hydrolysis test. All the media and biochemicals were procured from Hi Media Laboratories, India.

For the isolation and identification of uropathogens using HiCrome UTI Medium. The urine samples were inoculated on chromogenic UTI agar. The inoculated plates were incubated aerobically at 37°C for 24 hours. The gold standard of this method was the absence of biochemical tests for the identification of isolates.

All urine samples that were received in the lab underwent direct susceptibility testing right away. An unadjusted, well-mixed urine sample was used to test for confluence on a HiChrome agar plate using a sterile cotton swab. The extra fluid was then expelled. Using sterile forceps, commercial antimicrobial discs were firmly placed on the agar surface after being distributed via a multidisc

dispenser. Following incubation at 35°C for 16 to 18 hours, plates were read the following day. According to the Clinical and Laboratory Standards Institute (CLSI), zone diameter was measured and interpreted. In accordance with CLSI standards, the antimicrobial susceptibilities of isolates from pure cultures were evaluated for comparison using a conventional disc diffusion method.

Control strains used for the study were the following: Color of the colony.

Enterococcus faecalis ATCC 29212: blue, small
Escherichia coli ATCC 25922: pink purple
Klebsiella pneumoniae ATCC 13883, blue purple
Pseudomonas aeruginosa ATCC 27853: colorless (greenish pigment may be observed)
Proteus mirabilis ATCC 12453, light brown
Staphylococcus aureus subsp. aureus ATCC 25923: golden yellow

Statistical analysis: The data was anonymized and presented in percentages and graphs.

RESULTS AND DISCUSSION

A total of 531 patients of different age and sex suffering from UTI were included in this study. Among the patients, 215 (40.48%) were male and 316 (59.51%) were female. Their age distribution showed that 335/531 (63.08%) patients were in the age group 01 to 40 years, followed by 196/531 (36.91%) in the age group 41 to above 70 years. Majority of the UTI patients 435/531 (81.90%) were in the 20-50 years age group followed by >50 years 40 (7.53%) and below 20 years 56(10.54%). Regarding rate of bacterial growth, slightly higher number of culture positive cases was noted among female 316/531 (59.51%) than male 215/531 (40.48%) Fig. (1). This was in correlation with the study conducted by Moyo *et al*., (2010), in which the isolation of pathogens from females was at a rate of 54.4% and from males was at a rate of 45.6%. Another study by Nehar *et al*., (2020) also showed that among the patients, 121 (40.33%) were male and 179 (59.67%) were female, with a male-female ratio of 1:1.48. Anatomic factors like the short urethra, hormonal factors, and pregnancy contribute to the female preponderance of the UTI s. Our study demonstrated that the highest incidence of urinary isolates was in the 20–50 (81.90%)-year-old age group. This is coincident with the study by Shafiyabi *et al*., (2014). In this study, a peak incidence of UTI was noted in the age group between 21 and 50 years (55%).

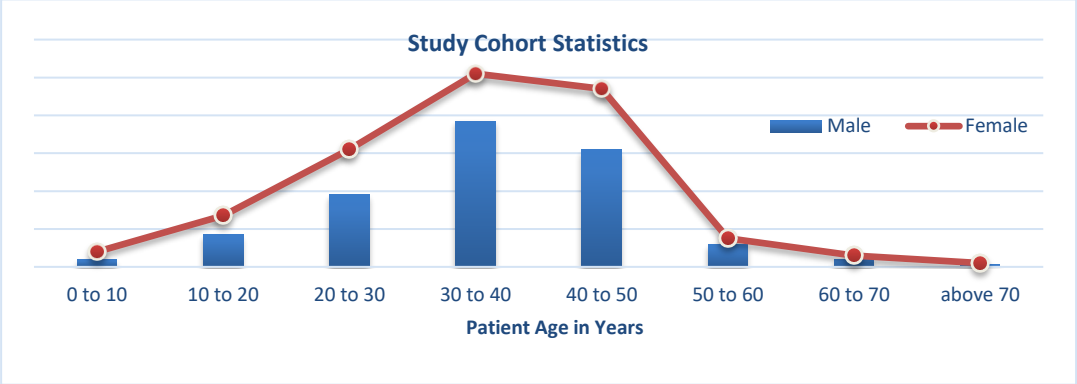

Fig. 1. Culture positive cases detected in chromogenic agar medium in relation to the age and gender of the patients (N=531)

Table 1. Spectrum of organisms isolated from culture positive cases (n=531)

Organism	Total isolates	Percentage (%)
E. coli	258	48.59
Klebsiella spp.	92	17.33
Enterococcus spp.	84	15.82
Pseudomonas spp.	71	13.37
Staphylococcus aureus	14	2.64
Acinetobacter spp.	9	1.69
Proteus spp.	3	0.56
Total	531	100.00

Table 2. Evaluation of four culture media for the rate of isolation of uropathogens (n=531)

Bacteria	Total isolates (n)	HiCrome UTI Agar (%)	CLED agar (%)	Blood Agar (%)	MacConkey agar (%)
E. coli	258	258 (100%)	258 (100%)	258 (100%)	258 (100%)
Klebsiella spp.	92	92 (100%)	92 (100%)	92 (100%)	92 (100%)
Enterococcus spp.	84	84 (100%)	84 (100%)	76 (100%)	76 (90.47%)
Pseudomonas spp.	71	71 (100%)	71 (100%)	71 (100%)	71 (100%)
Staphylococcus aureus	14	14 (100%)	14(100%)	14 (100%)	00 (00%)
Acinetobacter spp.	9	9 (22.22%)	0 (0%)	0 (0%)	0 (0%)
Proteus spp.	3	3 (100%)	2 (66.66%)	0 (0%)	0 (0%)
Total	531	531 (100%)	521 (98.11%)	511 (96.23%)	497 (93.59%)

Table 3. Rate of matching of bacterial isolates on HiCrome UTI agar medium with the standard colony color (N=531)

Bacteria	Total isolates (n)	Matched (%)	Not matched (%)
E. coli	258	255 (98.83%)	3 (1.16%)
Klebsiella spp.	92	92 (100%)	0 (0%)
Enterococcus spp.	84	84 (100%)	0 (0%)
Pseudomonas spp.	71	71 (100%)	0 (0%)
Staphylococcus aureus	14	14 (100%)	0 (0%)
Acinetobacter spp.	9	9 (100%)	0 (0%)
Proteus spp.	3	3 (100%)	0 (0%)
Total	531	528 (99.43%)	3 (0.56%)

Table 4. Antimicrobial susceptibility rate found in Staphylococcus aureus, Pseudomonas spp., Enterococcus spp. and total Enterobacteriaceae (n= 531)

Staphylococcus aureus (n=14)		Pseudomonas spp. (n= 71)		Enterococcus spp. (n = 84)		Total Enterobacteriaceae (n= 362)	
Antibiotics	Sensitivity	Antibiotics	Sensitivity	Antibiotics	Sensitivity	Antibiotics	Sensitivity
Azithromycin	2 (14.28%)	Ceftazidime	19 (26.76%)	Ampicillin	68 (80.95%)	Ampicillin	102 (28.18%)
Clarithromycin	3 (21.42%)	Gentamicin	21 (29.57%)	Penicillin	79 (94.94%)	Cefazolin	78 (21.55%)
Clindamycin	5 (35.71%)	Tobramycin	2 (2.81%)	Linezolid	21 (25%)	Gentamicin	105 (29.01%)
Cefoxitin	1 (7.14%)	Piperacillin-tazobactam	62 (87.32%)	Teicoplanin	31 (36.90%)	Tobramycin	43 (11.88%)
Penicillin	9 (64.28%)	Amikacin	43 (60.56%)	Vancomycin	57 (67.85%)	Amikacin	77 (21.27%)
Trimethoprim-Sulfamethoxazole	4 (28.57%)	Aztreonam	21 (29.57%)	Gentamicin (High-level resistance testing only)	39 (46.42%)	Amoxicillin–clavulanate	23 (6.35%)
Ceftaroline	2 (14.28%)	Cefepime	11 (15.49%)			Ampicillin–sulbactam	150 (41.44%)
Linezolid	2 (14.28%)	Ceftazidime-avibactam	35 (49.29%)	Streptomycin (High-level resistance testing only)	23 (27.38%)	Azithromycin	56 (15.47%)
Tetracycline	11 (78.57%)	Levofloxacin	16 (22.53%)			Cefepime	60 (16.57%)
Levofloxacin	7 (7.00%)	Meropenem	13 (18.30%)	Levofloxacin	12 (14.28%)	Cefotaxime	34 (9.39%)
Gentamicin	12 (85.71%)			Fosfomycin	21 (25%)	Cefotetan	46 (12.71%)
Minocycline	3 (21.42%)			Nitrofurantoin	14 (16.66%)	Ceftazidime–avibactam	24 (6.63%)
Nitrofurantoin	2 (14.28%)			Tetracycline	46 (54.76%)	Ceftriaxone	7 (1.93%)
Sulfisoxazole	1 (7.14%)					Cefuroxime	86 (23.76%)

Trimethoprim	1 (7.14%)	Meropenem	305 (84.25%)
		Piperacillin–Tazobactam	202 (55.80%)
		Aztreonam	23 (6.35%)
		Ceftazidime	245 (67.68%)
		Ceftaroline	89 (24.59%)
		Tetracycline	24 (6.63%)
		Cefazolin	56 (15.47%)
		Fosfomycin	34 (9.41%)
		Nitrofurantoin	32 (8.84%)

Out of 531 isolates, E. coli was the leading bacteria 258 (48.59%) followed by *Klebseilla spp*. 92 (17.33%), *Enterococcus* spp. 84 (15.82%), *Pseudomonas* spp. 71 (13.37%), *Staphylococcus aureus* 14 (02.64%), *Acinetobacter spp*. 09 (01.69%) and *Proteus spp*. 03(0.56%) Table (1). A similar pattern and rate of isolation of uropathogens were also observed by Nehar *et al.,* (2020). Another study by Jadhav *et al*, (2016); Karah *et al*., (2020) showed that among all the bacterial isolates (n = 578), E. coli had the highest number (195), followed by *Klebsiella* spp., which was found in a total of 81 samples, and *Staphylococcus aureus*, which constitutes 12.97% of the total urinary isolates, respectively. This is in accordance with a study carried out by Akter *et al*., (2014), which showed patterns of bacterial isolates from urine culture. *E. coli* was the leading bacteria isolated from 118 (59.30%) samples, followed by *Klebseilla* spp. 38 (19.09%), *Enterococcus spp.* 23 (11.56%), *Pseudomonas spp.* 04 (2.01%), and *Proteus spp*. 03 (1.51%). These urinary isolates were presumptively identified on Hicrome UTI agar, CLED agar, Blood, and MacConkey agar. The evaluation results of four culture media for the rate of isolation of uropathogens are shown in Table Table (2), maximum number of isolates was presumptively identified on Hicrome UTI agar (100%) and the rate of presumptively identification from CLED agar 521 (98.11%), Blood agar 511 (96.23%) and MacConkey agar 497 (93.59%).The studies carried out by Jain *et al*. (2023) showed chromosomal agar (CA) supported the growth of all isolates grown on blood agar (BA), whereas Mac Conkey supported only 93% of growth. In another study by Nehar *et al*., (2020), it was seen that both HiCrome UTI agar supported 100% bacterial growth, while MAC agar yielded 133 (91.72%) bacterial growth out of 145 total isolates.

The rate of matching with the standard colour for bacterial isolates on HiCrome UTI agar medium is shown in Table (3). It is evident that the colony colour of all isolates of *Klebsiella spp., Enterococcus spp., Staphylococcus aureus, Acinetobacter spp.* and *Proteus spp*. matched 100% with the standard colony colour on HiCrome UTI agar medium. While, out of 258 isolates of *E. coli*, 03 (01.16 %) did not match with the standard colony colour because of absence of β – galactosidase activity in that strain by Bose, (2015). According to Sharmin *et al*., (2010), the high prevalence of E. coli in UTI cases may be due to its existence as a normal flora in the large intestine and colonization in the perineal area. However, HiCrome UTI agar failed to produce expected colony colors for 3 (03.33%) of the E. coli. This behavior of certain bacterial species on chromogenic agar media has also been reported by other investigators (Nehar *et al*., 2020).

Table 4 shows the antimicrobial susceptibility rate found in Staphylococcus aureus, Pseudomonas spp., Enterococcus spp., and total Enterobacteriaceae. The sensitivity patterns of S. aureus to the following antibiotics: gentamicin, tetracycline, penicillin, and levofloxacin were 12 (85.71%), 11 (78.57%), 9 (64.28%), and 7 (7.00%), respectively. Gentamicin, Amoxycillin/clavulanate, Streptomycin, Cloxacillin, Erythromycin, Chloramphenicol, Cotrimoxazole, Tetracycline, Penicillin, Ciprofloxacin, Ofloxacin, Levofloxacin, Ceftriaxone, Amoxycillin, and vancomycin have been reported with sensitivity patterns of 92.4%, 63.0%, 44.2%, 35.8%, 52.4%, 61.9%, 15.5%, and 31.2%, according to Onwubiko et al. (2011). The sensitivity patterns of Pseudomonas spp. to the following antibiotics: piperacillin-tazobacta, amikacin, ceftazidime-avibactam, and gentamicin were 62 (87.32%), 43 (60.56%), 35 (49.29%), and 21 (29.57%), respectively. Javiya et al. (2008) studied combination drugs like ticarcillin + clavulanic acid, piperacillin + tazobactum, cefoperazone + sulbactum, cefotaxime + sulbactum, ceftriaxome + sulbactum, and monotherapy with amikacin, which showed higher sensitivity to Pseudomonas infections. Enterococcus spp. was sensitive to Penicillin 79 (94.94%), Ampicillin 68 (80.95%), Vancomycin 57 (67.85%), and Tetracycline 46

(54.76%). Similar study by Kraszewska et al. (2022). Enterobacteriaceae showed high sensitivity to Meropenem (84.25%), Ceftazidime (67.68%), Piperacillin-Tazobactam (55.80%), and Ampicillin-Sulbactam (41.44%), like those studied by Mulla et al. (2011).

CONCLUSION

Advanced diagnostic tools, like molecular methods, might not be affordable to all and need technical skills to perform. The overall findings of our study suggest that HiCrome UTI Medium supports the growth of all uropathogens. Furthermore, it provided the correct and rapid identification of organisms based on their color morphology. Therefore, it should be incorporated into routine laboratory work to process urinary cultures. DST using HiCrome media helps with the concurrent confirmation of bacterial species. It may be suggested for use as the main urine culture medium by clinical microbiology laboratories. The use of HiCrome UTI Medium minimizes the workload of a laboratory and helps treating clinicians initiate treatment with the right drug and dose at the right time for the right patient.

REFERENCES

Akter, M.L., Haque, R., and Salam, M.A. (2014). Comparative evaluation of chromogenic agar medium and conventional culture system for isolation and presumptive identification of uropathogens. *Pak J Med Sci*. 30(5):1033 1038. http://dx.doi.org/10.12669/pjms.305.5243

Ali, N., Hussain, W., Mirza, I.F., Niazi, A.A., Rubab, F., Salim, S., et al. (2023). Diagnostic accuracy of chromagar orientation for identification of uropatthogens. *Pak Armed Forces Med J*. 73 (1): 17-20. https://doi.org/10.51253/pafmj.v73i1.7453

Bajoria1, A., Kaore, N.M., Kaur, S., and Gautam, S.K. (2019). Utility of chromogenic media against conventional culture techniques for isolation of bacterial uropathogens in resource poor settings. *National Journal of Laboratory Medicine*. 8(1):5-8. DOI: 10.7860/NJLM/2019/39997:2331

Bose, S. (2015). The role of chromogenic replica system for isolation & identification of uropathogens in the era of molecular diagnostics. *British Journal of Medicine & Medical Research*, 8(1): 76-81. DOI:10.9734/bpi/nidpr/v1

Bouslah, Z., Mahjoub, R., Allagui, Z., Dabboussi, M., Zouaoui, M., Bibi, A. et al. (2020). Contribution of the Uriselect and the CPS ID chromogenic media to the isolation and identification of urinary tract infections bacteria. *Ann Biol Clin (Paris).*78(1):47-53. DOI: 10.1684/abc.2019.1512

Harris, M., and Fasolino, T. (2022). New and emerging technologies for the diagnosis of urinary tract infections. *J Lab Med.* 46(1): 3–15. https://doi.org/10.1515/labmed-2021-0085

Jadhav, P.A., and Roushani, S.B. (2016). HiCrome UTI agar: An alternative to conventional media for presumptive identification and isolation of uropathogens. *International Journal of Scientific Research*. 5(12): 20-23.

Javiya VA, Ghatak SB, Patel KR, Patel JA. Antibiotic susceptibility patterns of Pseudomonas aeruginosa at a tertiary care hospital in Gujarat, India. Indian J Pharmacol. 2008 Oct;40(5):230-4. doi: 10.4103/0253-7613.44156. PMID: 20040963; PMCID: PMC2792624.

Jain, C., and Nikita. (2023). Evaluation of chromogenic agar media for isolation, identification and direct antibiotic susceptibility testing of uropathogens. *International Journal of Pharmaceutical Research & Allied Sciences*. 12(2):7-12. https://doi.org/10.51847/Kd4vp42v9B

Karah, N., Rafei, R., Elamin, W., Ghazy, A., Abbara, A., Hamze, M. et al. (2020). Guideline for urine culture and biochemical identification of bacterial urinary pathogens in low-resource settings. *Diagnostics*. 10, 832. doi:10.3390/diagnostics10100832

Khalid, M. (2021). Comparison of chromogenic (HiCrome urinary tract infection agar) medium with cysteine lactose electrolyte deficient agar in a resource-limited setting. *International Journal of Applied and Basic Medical Research.* 11(1): 9-13. DOI: 10.4103/ijabmr.IJABMR_306_19

Kraszewska Z, Skowron K, Kwiecińska-Piróg J, Grudlewska-Buda K, Przekwas J, Wiktorczyk-Kapischke N, Wałecka-Zacharska E, Gospodarek-Komkowska E. Antibiotic Resistance of Enterococcus spp. Isolated from the Urine of Patients Hospitalized in the University Hospital in North-Central Poland, 2016-2021. Antibiotics (Basel). 2022 Dec 3;11(12):1749. doi: 10.3390/antibiotics11121749. PMID: 36551406; PMCID: PMC9774570.

Moyo, S.J., Aboud, S., Kasubi, M., Lyamuya, E.F., and Maselle, S.Y. (2010). Antimicrobial resistance among producers and non-producers of extended spectrum beta-lactamases in urinary isolates at a tertiary Hospital in Tanzania. *BMC Research Notes*.3:348. DOI:10.1186/1756-0500-3-348

Mulla S, Charan J, Panvala T. Antibiotic sensitivity of Enterobacteriaceae at a tertiary care center in India. Chron Young Sci 2011; 2:214-8.

Nahar, S.F., Hasan, M.B., Aktar, R., and Naznin, M. (2020). An assessment of chromogenic agar medium and conventional culture system for the isolation of uropathogens. *Scholars Journal of Applied Medical Sciences*. 8(1): 114-119. DOI:10.36347/sjams. 2020.v08i01.022

Sachu, A., and Samuel, A.K. (2022). Evaluation of chromogenic agar medium, can it be a suitable alternative to conventional culture system for identification of uropathogens. *Iran J Microbiol*. 14(6): 825–831. DOI: 10.18502/ijm. v14i6.11257

Shafiyabi1, S., Krishna, S., Jeer, M., Pavithra. and Divya. (2014). Trends in antibiotic resistance pattern among escherichia coli isolates from patients with urinary tract infection in tertiary care hospital, Bellary. *International Journal of Pharmaceutical Sciences Review and Research.* 24(2):43-49. www.globalresearchonline.net

Sharmin, S., Alamgir, F., Begum, F., and Jaigirdar, M.Q. H. (2010). Use of Chromogenic Agar Media for Identification of Uropathogen. *Bangladesh J Med Microbiol*. 04(01): 18-23. DOI: https://doi.org/10.3329/bjmm.v4i1.8464

Stefaniuk, E.M. (2018). The usefulness of chromogenic media for qualitative and semi-quantitative diagnostic of urinary tract infections. *Polish Journal of Microbiology.* 67(2): 213–218. DOI: 10.21307/pjm-2018-031

Onwubiko NE, Nasiru MS. Antibiotic sensitivity pattern of Staphylococcus aureus from clinical isolates in a tertiary health institution in Kano, Northwestern Nigeria. Pan Afr Med J. 2011; 8:4. doi: 10.4314/pamj. v8i1.71050. Epub 2011 Jan 26. PMID: 22121413; PMCID: PMC3201603.