Journal of Medical Diagnostic Methods
ISSN: 2168-9784
OPEN ACCESS Freely available online
Research Article

Assessment of Gram Stain Competency and Associated Factors among Laboratory Professional in Clinical Laboratories at Mekelle City Health Facility

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ABSTRACT

Background: Gram stains are initially used as a pre-analytical indicator of specimen quality and acceptability for culture. It also gives the clinician preliminary information regarding the nature of potential pathogens present in the patient specimen and thus serve to guide empirical therapy. This procedure is still considered a high-complexity procedure by the Clinical Laboratory Improvement Amendments (CLIA) program. The manual nature of the staining process and the subjectivity of Gram stain interpretation contribute to the incidence of errors.

Methodology: A cross-sectional study was conducted from January 2020 to March 2020 on 77 different health facilities present at Mekelle City. Convenient sampling method was used to collect qualitative data from 148 medical laboratory professional through a structured self-administered questionnaire and panel slide at on-site assessment of their performance. Question and panel slide was used to assess knowledge and skill of laboratory professionals respectively. Panel slides were prepared from American Type Culture Collection by using known bacterial strains and no organism slides were prepared from patient sample. Bloom's cut off point was used to describe the knowledge and practical skills of the respondents.

Result: Among 155 eligible medical laboratory professionals, 148 (95.5%) of them participated. Ninety Five (95) (64.2%) and One hundred twenty seven (127) (85.2%) participants had low knowledge and skill level respectively. The level of skills of medical laboratory professional had significant associated with educational level, accreditation status of health facility and training and also Education level, accreditation status of health facility, higher institution type and sex had significant association with knowledge level of study participant about gram stain ($p<0.05$).

Conclusion: The present study showed that the majority of medical laboratory professional had low knowledge and skill in gram stain examinations. Attention should be given to develop training strategies that can improve laboratory professional knowledge and skill level. This could be achieved through pre service and in service training and also giving adequate emphasis to gram stain related practical training, continuous follow up and regular competence assessment (supervision).

Keywords: Professional competence; Clinical performance; Gram stain

INTRODUCTION

Background

Gram staining is one of the laboratory test methods which are named after the Danish bacteriologist Hans Christian Gram who originally devised it in 1882 and published in 1884 to discriminate between *Pneumococci* and *Klebsiella pneumoniae* bacteria in lung tissue. Gram stain is a classic biological protocol that is still actively used to differentiate bacteria into two possible classifications: gram-positive cells, in which the primary stain is retained and gram-negative cells, in which the primary stain is lost [1-3].

Gram stains are initially used as a pre-analytical indicator of specimen quality and acceptability for culture. They also give the clinician preliminary information regarding the nature of potential pathogens present in the patient specimen and thus serve to guide empirical therapy. Although the gram stain has

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Received: June 09, 2020; **Accepted:** July 27, 2020; **Published:** August 03, 2020

Citation: Miruts F, Molla H, Hassen F, Hailu K, Lemlem L (2020) Assessment of Gram Stain Competency and Associated Factors among Laboratory Professional in Clinical Laboratories at Mekelle City Health Facility. J Med Diagn Meth 9:296. doi: 10.35248/2168-9784.2020.9.296

been the staple of clinical microbiology laboratories for over a century, it is still considered a high-complexity procedure by the Clinical Laboratory Improvement Amendments (CLIA) program. The manual nature of the staining process and the subjectivity of gram stain interpretation contribute to the incidence of errors [4-7]. Inappropriate specimen sampling, specimen processing, smear preparation, and prior antibiotic therapy are all factors that can have an adverse impact on gram stain result.

Gram stains remain the cornerstone of diagnostic testing in the microbiology laboratory for the guidance of empirical treatment prior to availability of culture results. Incorrectly interpreted gram stains may adversely impact patient care, and yet there are no comprehensive studies that have evaluated the reliability of the technique and there are no established standards for performance. In this study, clinical microbiology laboratories at four major tertiary medical care centers evaluated gram stain error rates across all non-blood specimen types by using standardized criteria [8].

In Ethiopia, most medical laboratories in health care facilities-cannot perform microbial culture for the diagnosis of microorganisms. Microbial culture for the diagnosis of microorganisms is a limited service. This was due to lack of skilled-man power, infrastructure, or cost of culture diagnostic materials and other issues [9]. Hence, gram stain is a method of choice for the diagnosis of microorganisms in most health care facilities, as microbial culture is impossible or not accessible in most of these facilities.

Competency defines the ability to carry out the total performance responsibilities of the given practitioner's generic position or competency as the combined knowledge and skill factors necessary to fulfill work obligations adequately. In other words, competency is the ability to carry out a specific task within given parameters of control. The competency required for clinical laboratory personnel reflects performance in many dimensions such as knowledge, intelligence, technical skills, problem solving abilities, interpersonal skills, and skills in oral and written expression [10-12].

In Ethiopia, even though gram stain is the method of choice for diagnosis of microorganisms to guide initial choice of antibiotic therapy as stated above, there was a gap between higher institutions to produce medical laboratory professionals with usable knowledge and practical skills [13], lack of health information resources [14], no evidence of refresher training and supervision on gram stain examination and interpretation, low level but progressive Strengthening Laboratory Management Towards Accreditation (SLMTA) practice [15,16], and lack of Continuing Professional Development (CPD) program. Hence, due to these gaps gram stain examination and interpretation by medical laboratory professionals is low quality and inconsistent. Lack of proper gram stains examination and interpretation could lead clinicians to miss diagnose and miss treat patients, which could cause drug resistance, resource-wastage, and suffering and possibly death of patients and others [6,17,18].

In Ethiopia, a study conducted on 12 hospital laboratories participated on the national PT program from 6 cycles of 2012 and 2013 GC, gram stain had high PT failure rates from 20 test parameters. In this study gram stain failure rate were 88.9% [19]. So, this study indicates there is high knowledge and skill gap on gram stain interpretation in Ethiopia including study area in particular.

In Africa including in Ethiopia there were also limited accessible studies in the scientific literature that reported competency of medical laboratory professionals on gram stain examination and interpretation. This indicates that the area was not enough examined by both researchers and responsible bodies for continuing professional development and quality medical laboratory services. Therefore, there was lack of scientific evidence on competency of medical laboratory professionals on gram stain examination and interpretation in Ethiopia and in the study area in particular.

Competency assessment it is a part of problem analysis and becomes a key tool in identifying errors through quality assurance process and also preventing from recurring. Competency assessment procedures can help to identify problems occurring in the technical aspects of laboratory practice and assess performance deficiencies before they develop into major problems. Competency assessment is also an opportunity to provide continuing education and performance feedback to employees and to document valuable objective information for performance evaluations. It should and can be used as a positive experience that helps to ensure that employees and employers can perform assigned tasks. The result of this study will contribute in provision of concrete data about the gram stain competency assessment and associated factors on clinical laboratories in the study area. It will also enable to design strategies for enhancing patient safety and reduce laboratory error because of significant laboratory errors have an impact on patient outcome, in some cases leading to erroneous medical interventions. In addition to this result will be useful for employee to evaluate own strength and weakness to perform required tasks, and also encourages employees to read and review carefully of laboratory policies and procedures.

MATERIAL AND METHODS

Study area

The study was conducted at Mekelle City, Tigray, North Ethiopia. Mekelle formerly the capital of Enderta Awraja in Tigray, is currently the capital city of Tigray National Regional state. It is located around 780 Kilometres north of the Ethiopian capital Addis Ababa, with an elevation of 2,254 metres (7,395 ft) above sea level. Administratively, Mekelle is considered a special zone, which is divided into seven sub-cities. Mekelle is the economic, cultural, and political hub of northern Ethiopia. Based on the 2007 Census conducted by the Central Statistical Agency of Ethiopia (CSA), this town has a total population of 215,914 people (104,925 men and 110,989 women). This city has 77 health facility, from those 62 are private health facility (4 primary hospital, 4 lower, 20 medium, 34 higher/speciality clinic), one uniformed hospital and 14 are governmental health facility those are 9 health center (Quiha Health Center, Aynalem Health Center, Kasech Health Center, Mekelle Health Center, Adi Shendehun Health Center, Adiha Health Center, Semen Health Center, Serawat and Lachi Health Center) , two primary hospital (HEWO and Adihaki), two general hospital (Mekelle and Quiha) and one Federal Hospital (Ayder Comprehencive Specialized Hospital). All health facility that is present at this city was included on the study.

Study design and period

A cross-sectional study was conducted from January 2020 to March 2020.

Population

Source population: All medical laboratory professionals who were working in medical laboratories of health facilities at Mekelle city, Ethiopia.

Study population: All medical laboratory professionals who were working medical laboratories of health facility in Mekelle City and fulfilled the eligibility criteria.

Eligibility criteria: All laboratory professionals working permanently in Mekelle City health facility during study period was eligible for the study.

Sample size determination and sampling techniques

The number of private and governmental health facility at Mekelle City were 77, from those 62 were private health facility (4 primary hospital, 4 lower, 20 medium, 34 higher/speciality clinic), one uniformed hospital and 14 are governmental health facility (Federal and Tigray Administration Regional Bureau), of those are 9 health center. The total number of medical laboratory professional who was working at governmental and private health facility was around 140 and 62 respectively. This makes a total of 202 professionals. Of those 17, 16, 7 and 7 laboratory, this makes 47 professional was excluded due to those are on education, break, training and also part time worker during the study period respectively, so total number of medical laboratory professional that are working permanently at government and private health facility during the study period was 155, those are included in the study to increase the reliability and accuracy of the study result. Preliminary assessment was done to know about the number of medical laboratory professionals in each health facility before data collection. After preliminary assessment there were 155 laboratory professional present at the whole health facility that are present at Mekelle City. Among those 95.5% (148) of them fully participated. The remaining 4.5% were not willing to participate and lack of time was the most frequent reason given not to participate. Of those 80, 63 and 5 medical laboratory professional who are from private, governmental and uniformed health facility respectively.

Sampling techniques

All laboratories professional who are working permanently at Mekelle Health Facility was included using convenience sampling technique.

Data collection tools and procedure

Data on socio demographic characteristics and 25 knowledge assessment question of study subject were collected by self-administered structured questioner. The questionnaire was developed based on reviewing different literatures. 7 panel slides were prepared to assess skill of laboratory professionals. Stained and graded slide were provided to each participant and then each participant was graded when interpreting one stained slide within five minutes. Participants were provided 25 structured knowledge questions with background questionnaire to fill their response within 30 minutes and also monitored the participant while filling the questionnaire so as to not use reading material and discuss with their friends. Functionality of microscope was checked by senior laboratory professional by using control slide that are prepared from ATCC. The data from gram stained slide identification and 25 structured knowledge questions with background questionnaire from each study participant were collected by the researcher.

Data management and data quality assurance

The following precautions were considered so as to assure the quality of the data. The data were collected by the researcher. By using standard data collection material, preparing of quality control from ATCC for checking of microscope functionality before assessing the performance of laboratory personnel and pre testing of the questioner's data at wukro general hospital, checking the completeness of questionnaires. Based on the findings of the pre-test, some questions were modified. Furthermore data were checked during entry into the computer before analysis. After preparation of panel slide from ATCC, all slides were stained by gram staining procedure. Then senior microbiologist were validated the gram stain interpretation agreement with the culture growth on blood agar and also interpreted the prepared gram stain smears using investigative criteria for the presence or absence of gram stain findings (bacteria, yeasts and cells) and also quantification of findings (few, moderate and many).

Data analysis and presentation

Data obtained was checked for completeness and qualitative data from the questionnaires coded and entered into SPSS version 23 data analysis. Descriptive statistics were used to summarize socio demographic characteristics of study participant. The univariate analysis such as percentage and frequency distribution of different characteristics of the questionnaire was analyzed. Bivariate analysis was used to see the association of independent with the dependent variable. A chi square model was employed to determine associations with dependent and independent variables. Percent of correct response to a set of 25 knowledge questions were graded as follows: 59% or below (14.7/25) as low, 60%-80% (15-20/25) as medium and above 80% (>20/25): as high knowledge level. Similarly, percent of correct responses to a set of 7 skill related questions were graded as follows: those who perform correctly in 59% or below (4.13/7) as low, 60%-80% (4.2-5.6/7) as medium and above 80% (>5.6/7) as high skill level (34). Major error, minor error, very major error, maximum score, minimum score were analyzed for skill tests and also participants grouped in to low, medium and high skill level. For 25 knowledge questions the analyses were performed for maximum score and minimum score of questioner answers and also classify participants to low, medium and high knowledge level. p-value of <0.05 was considered statistical significant. Participants' knowledge about gram stain was assessed using 25 questions. Define gram stain, difference between cell structure of gram positive and gram negative bacteria, important of decolourization in gram staining, why is the gram stain considered a differential stain and important of knowing gram positive or gram negative bacteria. Those questions had a coded in spss (minimum correct response coded by 'Yes' and wrong or don't know response had a code of 'No'). The rest had single answers, so giving a value as a choice question.

RESULTS

Socio-demographic characteristics of the study participant

During study time, there were 77 health facilities; of them 62 were private, 14 governmental and also one uniformed health facility.

Among 62 health facilities, 58 of them was clinics, the rest one was primary hospitals and also among 14 governmental health facility, 9 was health center, one specialized hospital, two primary hospitals and two referral hospitals.

Of 155 medical laboratory professional, 95.5% (148) of them fully participated. The remaining 4.5% were not willing to participate and lack of time was the most frequent reason given not to participate. Of which 78 (52.7%) were males. regarding education, 67 (45.3%), 73 (49.3%) and 8 (5.4%) were degree, diploma and masters holder respectively. Fifty nine (39.9%) of the respondents were in the age group of 26-30 years. The present study indicates Most of study participants 107 (72.3%) had work experience of ≤ 9 years. Nearly half, 81 (54.7%) participants were from private health facility and seventy two (46.2%) respondents was who graduate from Government University. Most 109 (73.6%) of respondent were not participate in EQA and among study participants most of them 106 (71.6%) were not get regular competency assessment (supervision). Of respondent 142 (95.9%) were not get training related to gram stain. During the study time, 109 (73.6%) respondent were from health facilities that are not participate on accreditation program (Table 1).

Resource access of study participants

Out of all study participants, there was 102 (68.9%) laboratory professional who have health information resource in their laboratory section. 53 (35.8%) of study participants used Olympus Microscope (Table 2).

Knowledge of medical laboratory professional

For the 25 theoretical knowledge questions administered to 148 participants, minimum score was 0 (0%) and maximum score was 24 (96%). A number of medical laboratory professionals who score 0 of 25 knowledge assessment question were 7 (4.7%). Among participants, the knowledge score was low for 95 (64.2%), medium for 49 (33.1%) and high for 4 (2.7%). From study respondents with low knowledge level, most of the respondents 59 (62.1%) with low knowledge level were those who had diploma. Analyses were carried out to examine the association between different factors and knowledge of study participants on gram stain examination. Education level, accreditation status of health facility, higher institution type and sex had statistically significant association with knowledge level of study participants (p value<0.005). Out of 148 study participant four respondents had high knowledge level, among those two of them had degree holder and one had diploma and masters holder (Table 3).

Out of four study participants who had high knowledge level, three of them were from health facility that had star 4 level of accreditation, the rest one was from a heath facility that are not participate on accreditation. About 79 (83.2%) of respondents who had low knowledge level was from health facility that are not participate on accreditation. Of respondents with high knowledge level (4), 3 (75%) of them were respondents who graduate from government university and also all respondents who graduates in private college had low knowledge level. As the present study indicates among low and high knowledge level participants, 55 (57.9%) and 3 (75%) was female and male respectively (Table 3).

Age, microscope type, health facility type, health information resources, EQA participation, training, competency assessment and experience had no significant association with knowledge level of medical laboratory professionals on Gram stain examination (Table 3).

Table 1: Socio demographic characteristics of medical laboratory professionals in Mekelle City Health Facility, Tigray, Ethiopia 2020 (N=148).

Variable characteristics		Number	Percent
Sex	Male	78	52.7
	Female	70	47.3
Age in years	≤ 25	27	18.2
	26-30	59	39.9
	31-35	36	24.3
	≥ 36	26	17.6
Experience in years	0-9	107	72.3
	19-Oct	35	23.6
	≥ 20	6	4.1
Educational level	Diploma	73	49.3
	Degree	67	45.3
	Masters	8	5.4
Health facility type	Government	62	41.9
	Private	81	54.7
	Uniformed	5	3.4
EQA	Yes	39	26.4
	No	109	73.6
Current accreditation status	Not participated	109	73.6
	Star 0	4	2.7
	Star 1	5	3.4
	Star 2	11	7.4
	Star 4	19	12.8
Training related to gram stain	Yes	6	4.1
	No	142	95.9
Competency assessment (supervision)	Yes	42	28.4
	No	106	71.6
Institution type	Government college	59	39.9
	Government university	72	48.6
	Private college	17	11.5
Over all total	-	148	100

Table 2: Resource access of study participants at Mekelle City Health Facility, Tigray, Ethiopia 2020.

Variable characteristics		Number	Percent
Health information resource	Yes	102	68.9
	No	46	31.1
	Total	148	100
Types of microscope	Olympus	53	35.8
	Novel	40	27
	Primo star	21	14.2
	Gemmy and heuer	19	12.8
	Other	15	10.1
	Total	148	100

Response of medical laboratory professional on each theoretical knowledge question about Gram Stain

Out of all participants, 110 (74.3%) respondents score correct answer on question of what is a primary stain in gram stain technique, and also 124 (83.8%) respondents score incorrect answer on the question of what would happen if iodine were missed during gram staining procedure (Table 4).

The level of the skills of the medical laboratory professional on gram stain examination

Among all respondents assessed for skills in gram stain examination, 127 (85.2%), 19 (12.8%) and 2 (1.3%) participants scored low, medium and high skill level respectively. The level of skills of medical laboratory professional varied according to educational level, accreditation status and training ($p<0.05$) (Table 5).

During study time, regarding to educational level among respondents with low skill level, most of the respondents were diploma and degree level 71 (55.9%) and 52 (40.9%) respectively. Of respondents who had low skill level, 101 (79.5%) was from health facility that are not participate on accreditations. In addition to this participants from accreditation level of stare 0 and 1 health facility had no high skill level but respondents with high skill level was those are from accreditation level 2 and 4 health facility. Among all respondents, 120 (81.1%) participants incorrectly report

Table 3: Knowledge of medical laboratory professional related to background characteristics in Mekelle City Health Facility, Tigray, Ethiopia, 2020 (n=148).

Variable characteristics		Knowledge level			Total	X^2	P-value
		Low No (%)	Medium No (%)	High No (%)			
Age	<25	26 (17.6%)	1 (0.7%)	0 (0%)	27 (18.2%)	5.359	0.499
	26-30	47 (31.8%)	11 (7.4%)	1 (0.7%)	59 (39.9%)		
	31-35	31 (20.9%)	4 (2.7%)	1(0.7)	36 (24.3%)		
	≥ 36	23 (15.5%)	3 (2%)	0 (0%)	26 (17.6%)		
Sex	Male	40 (27%)	35 (23.6%)	3 (2%)	78 (52.7%)	11.971	0.003
	Female	55 (37.2%)	14 (9.5%)	1(0.7%)	70 (47.3%)		
Health facility type	Government	33 (53.2%)	27 (43.5%)	3 (2%)	62 (41.9%)	5.827	0.212
	Private	58 (71.6)	21 (25.9)	1 (1.2%)	80 (54.1%)		
	Uniformed	4 (80%)	1 (20%)	0	5 (3.4)		
Higher institution type	Government college	43 (29.1%)	15 (10.1%)	1 (0.7%)	59 (39.9%)	19.078	0.001
	Government university	35 (23.6%)	34 (22.9%)	3 (2%)	72 (48.6%)		
	Private college	17 (11.5%)	0 (0%)	0 (0%)	17 (11.5%)		
Experience in years	0-9	73 (49.3%)	32 (21.6%)	2 (1.4%)	107 (72.3%)	3.957	0.412
	19-Oct	19 (12.8%)	14 (9.5%)	2 (1.4%)	35 (23.6%)		
	≥ 20	3 (2%)	3 (2%)	0 (0%)	6 (4.1%)		
Educational level	Diploma	59 (39.9%)	13 (8.8%)	1 (0.7%)	73 (49.3%)	27.169	0
	Degree	36 (24.3%)	29 (19.6%)	2 (1.4%)	67 (45.3%)		
	Masters	0 (0%)	7 (4.7%)	1 (0.7%)	8 (5.4%)		
EQA	Yes	22 (14.9%)	16 (10.8%)	1 (0.7%)	39 (26.4%)	1.506	0.471
	No	73 (49.3%)	33 (22.3%)	3 (2%)	109 (73.6%)		
Current accreditation status	Not participated	79 (53.4)	28 (18.9%)	1 (0.7%)	108	22.381	0.004
	Star 0	2 (1.4%)	2 (1.4%)	0	4 (2.7%)		
	Star 1	4 (2.7%)	1 (0.7%)	0	5 (3.4%)		
	Star 2	6 (4.1%)	5 (3.4%)	0	11 (7.4%)		
	Star 4	4 (2.7%)	13 (8.8%)	3 (2%)	20 (13.5%)		
Training	Yes	2 (1.4%)	3 (2%)	1 (0.7%)	6 (4.1%)	5.978	0.05
	No	93 (62.8%)	46 (31.1%)	3 (2%)	142 (95.9%)		
Competency assessment	Yes	28 (18.9%)	13 (8.8%)	1 (0.7%)	42 (28.4%)	0.161	0.923
	No	67 (45.3%)	36 (24.3%)	3 (2%)	106 (71.6%)		
Health information resource	Yes	59 (39.9%)	39 (26.4%)	4 (2.7%)	102 (68.9%)	6.469	0.039
	No	36 (24.3%)	10 (6.8%)	0	46 (31.1%)		
Types of microscope	Olympus	33 (22.3%)	18 (12.2%)	2(1.4%)	53 (35.8%)	11.624	0.071
	Novel	26 (17.6%)	13 (8.8%)	1 (0.7%)	40 (27%)		
	Primo star	8 (5.4%)	12 (8.1%)	1 (0.7%)	21 (14.2%)		
	Gemmy and heuer	15 (10.1%)	4 (2.7%)	0 (0%)	19 (12.8%)		
	Other	13 (8.8%)	2 (1.4%)	0 (0%)	15 (10.1%)		
Over all total		95 (64.2%)	49 (33.1%)	4 (2.7%)	148 (100%)	-	-

Table 4: Participants' knowledge about gram stain among medical laboratory professionals at Mekelle City, Tigray, Ethiopia, 2020 (n=148).

No	Item	Yes (Correct)	No (Incorrect)
1	What is gram stain?	67 (45.3%)	81 (54.7%)
2	Write at least one gram positive bacteria	79 (53.4%)	69 (46.6%)
3	What is the difference between cell structure of gram positive and gram negative bacteria?	28 (18.9%)	120 (81.1%)
4	Why is the decolourization important in gram staining?	39 (26.4%)	109 (73.6%)
5	What are the possible cause of false gram reaction results?	108 (73%)	30 (27%)
6	Using gram staining method, what is the appearance of gram positive bacteria	41(27.7%)	107(72.3%)
7	Using gram staining method, what is the appearance of gram negative bacteria	88 (59.5%)	60 (40.5%)
8	Which bacteria's are decolorized by acetone alcohol and contain a counter stain colour?	90 (60.8%)	58 (39.2%)
9	Which bacteria resist decolourization and retain the colour of the primary stain?	90 (60.8%)	58 (39.2%)
10	What is a counter stain in gram stain technique?	106 (71.6%)	42 (28.4%)
11	What is a primary stain in gram stain technique?	110 (74.3%)	38 (25.7%)
12	Write one possible morphologies of bacteria	77 (52%)	71 (48%)
13	What would happen if iodine were missed during gram staining?	24 (16.2%)	124 (83.8%)
14	What is the order of reagents used in the Gram stain	82 (55.4%)	66 (44.6%)
15	Why we use heat fixative after smearing the sample before we use reagent to stain bacteria?	80 (54.1%)	68 (45.9%)
16	Write typical morphology of *Staphylococcus aureus*	51 (34.5%)	97 (65.5%)
17	Why is the gram stain considered a differential stain?	12 (8.1%)	136 (91.9%)
18	What does gram positive cocci in pairs mean (suggestion/interpretation)?	57 (38.5%)	91 (61.5%)
19	What does gram positive cocci in cluster mean (suggestion/interpretation)?	51 (34.5%)	97 (65.5%)
20	Why is it important to know gram positive or gram negative?	68 (45.9%)	80 (54.1%)
21	Write a counter stain other than safranin be used in gram staining	21 (14.2%)	127 (85.8%)
22	Write a primary stain other than crystal violet be used in gram staining	20 (13.5%)	128 (86.5%)
23	What happens if you reverse crystal violet and safranin stains?	28 (18.9%)	120 (81.1%)
24	Is safranin basic or acidic dye?	77 (52%)	71 (48%)
25	Write at least one gram negative bacteria	59 (39.9%)	41 (60.1%)

Table 5: Skill of medical laboratory professional on each slides about gram stain.

Item	Correct	Incorrect
Gss 1	66 (44.6%)	82 (55.4%)
Gss 2	49 (33.1%)	99 (66.9%)
Gss 3	70 (47.3%)	78 (52.7%)
Gss 4	52 (35.1%)	96 (64.5%)
Gss 5	87 (58.8%)	61 (41.2%)
Gss 6	28 (18.9%)	120 (81.1%)
Gss 7	64 (43.2%)	84 (56.8%)

gram stain slide 6 (gram positive cocci) as gram negative bacteria and also 87 (58.8%) participants correctly report gram stain slide 5 (gram negative rod) (Table 5).

Out of high skill level participants, all 2 (100%) had gram stain training certificate whereas of all participants with low knowledge level, almost all of them 124 (97.6%) had no training on gram stain. Among all study participants, 83.8% had no training certificate on gram stain and had low knowledge level. Of respondents with low knowledge level were a respondents who get one 2 (33.3%) and two 1 (16.7%) gram stain training certificate (Table 6).

Errors of medical laboratory professionals on gram stain examination

During the study time, results from a total of (148 × 7=1036) observation, 437 (42.2%)observation with major error and 308 (29.7%) observation with very major error. 249 observations (24%) reported gram positive bacteria as gram negative bacteria and 106 observations (10.2%) reported gram negative bacteria as gram positive bacteria (Table 7).

About 55.4% of laboratory professionals have reported negative slides (slide 1) as false positive (gram positive and gram negative). 49 (33.1%) of the participants correctly reported the bacteria of slide 2. Slide 3 contains gram positive rod (many), 70 (47.3%) of the participants correctly reported the bacteria. Similarly 52 (35.1%) of the participants correctly reported the bacteria of slide 4. Out of respondents 87 (58.8%), 28 (18.9%), and 64 (43.2%) correctly reported bacteria of slide 5, 6 and 7 respectively (Table 7).

Performance of laboratory personnel to correctly identify gram stain slides

The overall sensitivity, specificity, PPV and NPV of laboratory professionals in gram stain examination were 80.7% and 44.6%, 89.7% and 27.8% respectively (Table 8).

Association of knowledge and skill level of medical laboratory professionals on gram stain examination

There was statistically significant association between knowledge and skill level of study participants on gram stain examination ($p<0.05$). Those who had low knowledge level scored low skill level on gram stain examination but they had medium skill level 3 (2%). Among all study participants 92 (62.2%) scored low knowledge level and low skill level, but among high knowledge level participants half of them had low skill level, 15 (10.1%) had medium knowledge level and medium skill level and 1 (0.7%) was with high knowledge and skill level (Table 9).

Table 6: The level of the skills of the medical laboratory professional on gram stain examination in Mekelle City Health Facility, Tigray, Ethiopia, 2020.

Variable characteristics		Skill level			Total	x2	p-value
		Low No (%)	Medium No (%)	High No (%)			
Age	<25	26 (17.6%)	1 (0.7%)	0 (0%)	27 (18.2%)	5.359	0.499
	26-30	47 (31.8%)	11 (7.4%)	1 (0.7%)	59 (39.9%)		
	31-35	31 (20.9%)	4 (2.7%)	1 (0.7%)	36 (24.3%)		
	≥ 36	23 (15.5%)	3 (2%)	0 (0%)	26 (17.6%)		
Sex	Male	62 (41.9%)	14 (9.5%)	2 (1.4%)	78 (52.7%)	5.919	0.052
	Female	65 (43.9%)	5 (3.4%)	0 (0%)	70 (47.3%)		
Health facility type	Government	50 (79.4%)	11 (17.5%)	2 (3.2%)	63 (42.6%)	4.438	0.232
	Private	73 (91.2%)	7 (8.8%)	0 (0%)	80 (54.1%)		
	Uniformed	4 (80%)	1 (20%)	0 (0%)	5 (3.4%)		
Higher institution type	Government college	56 (37.8%)	3 (2%)	0 (0%)	59 (39.9%)	13.945	0.007
	Government university	54 (36.5%)	16 (10.8%)	2 (1.4%)	72 (48.6%)		
	Private college	17 (11.5%)	0 (0%)	0 (0%)	17 (11.5%)		
Experience in years	0-9	92 (62.2%)	14 (9.5%)	1 (0.7%)	107 (72.3%)	0.938	0.919
	19-Oct	30 (20.3%)	4 (3.1%)	1 (0.7%)	35 (23.6%)		
	≥ 20	5 (3.4%)	1 (0.7%)	0 (0%)	6 (4.1%)		
Educational level	Diploma	71 (55.9%)	2 (10.5%)	0 (0%)	73 (49.3%)	21.907	0
	Degree	52 (40.9%)	13 (68.4%)	2 (2.9%)	67 (45.3%)		
	Masters	4 (3.1%)	4 (3.1%)	0 (0%)	8 (5.4%)		
EQA	Yes	31 (20.9%)	7 (4.7%)	1 (0.7%)	39 (26.4%)	1.901	0.387
	No	96 (64.9%)	12 (8.1%)	1 (0.7%)	109 (73.6%)		
Current accreditation status	Not participated	101 (79.5%)	8 (5.4%)	0 (0%)	109 (73.6%)	28.191	0
	Star 0	4 (3.1%)	0 (0%)	0 (0%)	4 (3.1%)		
	Star 1	4 (3.1%)	1 (0.7%)	0 (0%)	5 (3.4%)		
	Star 2	8 (5.4%)	2 (1.4%)	1 (0.7%)	11 (7.4%)		
	Star 4	10 (6.8%)	8 (5.4%)	1 (0.7%)	19 (12.8%)		
Training	Yes	3 (2%)	1 (0.7%)	2 (1.4%)	6 (4.1%)	48.339	0
	No	124 (83.8%)	18 (94.7%)	0 (0%)	142 (95.9%)		
If yes, how many times/certificate	One	2 (1.4%)	0 (0%)	1 (0.7%)	3 (2%)	-	-
	Two	1 (0.7%)	0 (0%)	1 (0.7%)	2 (1.4%)	-	-
	Four	0 (0%)	1 (0.7%)	0 (0%)	1 (0.7%)	-	-
	Total	3 (2%)	1 (0.7%)	2 (1.4%)	6 (4.1%)	-	-
Competency assessment (supervision)	Yes	36 (24.3%)	5 (3.4%)	1 (0.7%)	42 (28.4%)	0.5	0.779
	No	91 (61.5%)	14 (9.5%)	1 (0.7%)	106 (71.6%)		
Health information resource	Yes	83 (56.1%)	17 (11.5%)	2 (1.4%)	102 (68.9%)	5.403	0.067
	No	44 (29.7%)	2 (1.4%)	0 (0%)	46 (31.1%)		
	Total	127 (85.8%)	19 (12.8%)	2 (1.4%)	148 (100%)		
Types of microscope	Olympus	43 (29.1)	8 (5.4%)	2 (1.4%)	53 (35.8%)	7.763	0.256
	Novel	37 (25%)	3 (2%)	0 (0%)	40 (27%)		
	Primo star	16 (10.8%)	5 (3.4%)	0 (0%)	21 (14.2%)		
	Gemmy and heuer	18 (12.2%)	1 (0.7%)	0 (0%)	19 (12.8%)		
	Other	13 (8.8%)	2 (1.4%)	0 (0%)	15 (10.1%)		
Over all total		127 (85.8%)	19 (12.8%)	2 (1.4%)	148 (100%)	-	-

Table 7: The skill level of medical laboratory professionals on gram stain examination at Mekelle City Health Facility, Tigray, Ethiopia 2020.

Slide number	Gram reaction				Total	Characteristics			Error type		
	No	Pos	Neg	Both		No. of participants correctly report Morphology of bacteria	No. of participants correctly report Density of bacteria	Other cells (PMNS)	Minor	Major	V major
GSS 1	66	48	34	0	148	-	-	11	-	82	137
GSS 2	25	49	73	1	148	64	54	1	1	73	25
GSS 3	28	70	47	3	148	75	64	0	0	47	28
GSS 4	37	59	52	0	148	38	28	0	0	59	37
GSS 5	14	47	87	0	148	79	19	1	1	47	14
GSS 6	26	28	92	2	148	57	44	1	1	92	26
GSS 7	41	64	37	6	148	35	21	1	1	37	41
Total	237	365	422	12	1036	-	-	-	4	437	308

Table 8: Performance of laboratory personnel to correctly identify gram stain slides, Mekelle, Tigray, Ethiopia, 2020.

		Known result		Total	Sensitivity	Specificity	PPV	NPV
	-	Pos	Neg					
Participant reader	Pos	717	82	799	80.70%	44.60%	89.70%	27.80%
	Neg	171	66	237				
	Total	888	148	1036				

Table 9: Association of knowledge and skill level of medical laboratory professionals on gram stain examination at Mekelle City, Tigray, Ethiopia.

	Skill level				
	-	Low	Medium	High	Total	p-value
Knowledge level	Low	92 (62.2%)	3 (2%)	0	95	0
	Medium	33 (22.3%)	15 (10.1%)	1 (0.7%)	49	
	High	2 (1.4%)	1 (0.7%)	1 (0.7%)	4	
	Total	127	19	2	148	

DISCUSSION

The goal of competency assessment is to improve the laboratory's performance by identifying areas requiring education and/or training of the staff and thus ensuring patient safety. Ongoing competency assessment outside initial training is not as universally implemented and, in some cases where it exists, may be limited in scope and intensity. Performing competency assessment as the need arises is a reactive rather than a proactive approach. The goal should be to detect problems before they happen. Ongoing assessment needs to be incorporated into a laboratory's quality management system. Quality laboratory testing is an essential building block of the clinical diagnosis scheme, infectious disease surveillance, and the development of public health policy. Following good laboratory practices leads to reliable and accurate test results, which in turn fosters good patient care and promotes a positive attitude toward testing from providers' and patients' perspectives.

In this study, we used both knowledge and skill tests to assess competence of medical laboratory professionals and associated factors on gram stain examination. This study showed Among 148 participants, the knowledge score was low for 95 (63.8%), medium for 49 (32.9%) and high for 4 (2.7%) and also the skill score was low for 127 (85.2%), medium for 19 (12.8%) and high for 2 (1.4%) so, this finding revealed that participants with low knowledge level were higher than the study conducted at Adis Abeba, regarding to skill level, as study at Adis Abeba revealed that participants with high skill level was 33.7% , so this indicate the knowledge and skill level of participants was better than our studies this may be due to all participants was from hospitals (33) but in both studies the participants with high knowledge level were very few. This may be due to lack of regular competency assessment, training and also lack of attention from responsible body. When we see skill test of our study, out of all participants 13.4% and 21.4% respondents correctly identify gram negative and gram positive respectively this may be due to lack of proper funding, adequate training for laboratory workers and systematic management of work. A study in the U.S showed that participants score of gram negative (88%) and gram positive (90%), which was higher than our study so, this finding was it may be due to adequate training, supervision, high infrastructure of laboratory facility and also advanced technology.

Our study indicate that from 25 knowledge assessment questions the mean score was 11.07 (range 0%-96%), mean score was lower than the finding in USA in 2014 by Goodyear N [18]. This may be due to continuous training, concerning body giving an attention on this, enough funding budget to improve quality and also advanced set up.

Analyses were carried out to examine the association between different factors and knowledge of study participants on gram stain examination. So our study found sex, higher institution type, education level and accreditation status had statistically significant association with knowledge level of study participants. Level of knowledge of participants regarding to educational level was low for diploma holder than degree and master's holder. So

concerning body should give education opportunity to medical laboratory professional in order to upgrade themselves. From study respondents with low knowledge level, most of the respondents 59 (62.1%) with low knowledge level were those who had diploma, this number was higher than the study conducted at Adis Abeba. This may be due to most study participants were diploma level or it may be due to most participants was from private health facility. Of respondents with high knowledge level, all of them were respondents who graduate from government institution this was similar with the study conducted at Adis Abeba. Both study results indicate may be due to responsible body not give attention on private higher institution. When we compare government institution (university and college), 3 (75%) of them was from government university this may be due to responsible body gives attention on government university rather than college. In addition to this when we compare government college and private college, all respondents who graduates in private college had low knowledge and skill level but not participants from government college, which had medium and high knowledge level and also medium skill level. This may be due to lack of attention of responsible body on private higher institution.

This study showed that out of four study participants who had high knowledge level, three of them were from health facility that had star 4 level of accreditation, the rest one was from a heath facility that are not participate on accreditation and also of the low level respondents about 79 (73.1%) of respondents who had low knowledge level was from health facility that are not participate on accreditation but only 4 (4.2%) respondents who were from health facility that had level 4 accreditation so, this indicates participation on accreditation has an impact on knowledge of medical laboratory professional.

Among 148 respondents assessed for skills in gram stain examination, one hundred twenty seven (127) (85.2%), nineteen (19) (12.8%) and 2 (1.3%) participants scored low, medium and high skill level respectively in addition to this high and low score for skill was 6 (85.7%) and 0 (0%) from all 7 skill tests. The level of skills of medical laboratory professional varied according to educational level, accreditation status and training. During study time, regarding to educational level among respondents with low skill level, most of the respondents were diploma and degree level 71 (55.9%) and 52 (40.9%) respectively. Participants who were diploma and masters level had no high skill level. Of respondents who had low skill level, 101 (79.5%) and also no high skill level was from health facility that are not participate on accreditations in addition to this accreditation level of stare 0, 1 had no high skill level. From study participants with high skill level, all respondents 2 (100%) were those were from accreditation level star 2 and 4 health facility. Although related literature was not accessed, our study showed that participation on accreditation and also increase step of accreditation had an impact on skill of medical laboratory professional.

This study showed that among all study participants, 83.8% had no training on gram stain and had low knowledge level. From all study participants with high skill level, all of them had gram stain training certificate. Of respondents who get training certificate with low skill level were a respondents who get one 2 (1.4%) and two 1 (0.7%) training certificate. Participants who get four training certificate had no low skill on gram stain examination. The study done in US by using gram stained smear of known culture result, accuracy of identification of *Pseudomonas aeruginosa* was 60% but after 1-2 year of in service training it improved progressively to over 80%. So this study indicates if there is continuous training it can be improve our skill level. Generally our study showed that this area had lack of attention from concerning body. Doing competency assessment definitely improve the quality of the laboratory services provided using gram staining methods.

LIMITATION

Too little literature was available and shortage of recently conducted studies are some of the limitation.

CONCLUSION

The present study showed that the majority of medical laboratory professional had low knowledge and skill in gram stain examinations this may be due to most participants had no get training, were from heath facility that are not participate on accreditation, diploma level and also it may be due to the professionals focus on other laboratory technique.

As the present study indicates, higher institution type, sex, accreditation status of health facility and educational level affect knowledge level of medical laboratory professional on gram stain examination. Educational level, accreditation status of health facility and training were affect skill level of medical laboratory professional on gram stain examination. As our study showed most participant with low skill level were working without training this may effect on gram stain examination. Even though this study did not show association of microscope type with skill level, the study conducted at Adis Abeba indicates microscope type correlated with skill of medical laboratory professionals on gram-stain examination.

Knowledge level was found to be associated with skill level of medical laboratory professionals on gram stain examination and interpretation. Participants who had low knowledge can have low skill on gram stain. Educational level, accreditation status, training, higher institution type and sex were factors that are affect skill and knowledge of study participant about gram stain. Generally, our study indicates the competency level (skill and knowledge) of medical laboratory professional that are present at Mekelle City had low on gram stain examination this may be due to lack of training, continuous professional development, supervision, attention by responsible body and also professionals focus on other laboratory technique.

Our study showed 73.1% of participant who comes from non-accredited health facility had low knowledge level.

ETHICAL CONSIDERATIONS

This study was ethically approved by the "Departmental Research and Ethical Review Committee" (DRERC) of the Department of Medical Laboratory Science. Permission letter was written from Tigray regional health biro. All the collected data was kept confidentially by using codes instead of any personal identifiers. It is also cleared that participation fully based on the willingness of participants using written consent.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ACKNOWLEDGMENTS

I would like to thank to Haftay Haileslase for guiding how to implement my thesis and also for all laboratory professional those are voluntary to participate on this research.

REFERENCES

1. Adwan G, Shtayah A, Adwan K, Al-Sheboul S, Othman S. Prevalence and molecular characterization of *P. aeruginosa* isolates in the west bank-palestine for ESBLs, MBLs and integrons. J Appl Life Sci Int. 2016;8:1-11.
2. Rawat D, Nair D. Extended-spectrum β-lactamases in gram negative bacteria. J Glob Infect Dis. 2010;2:263-274.
3. Wilhelm MJ, Sheffield JB, Sharifian GM, Wu Y, Spahr C, Gonella G, et al. Gram's stain does not cross the bacterial cytoplasmic membrane. ACS Chem Biol. 2015;10:1711-1717.
4. Nagendra S, Bourbeau P, Brecher S, Dunne M, LaRocco M, Doern G. Sampling variability in the microbiological evaluation of expectorated sputa and endotracheal aspirates. J Clin Microbiol 2001;39:2344-2347.
5. Munson E, Block T, Basile J, Hryciuk JE, Schell RF. Mechanisms to assess gram stain interpretation proficiency of technologists at satellite laboratories. J Clin Microbiol. 2007;45:3754-3758.
6. Rand KH, Tillan M. Errors in interpretation of gram stains from positive blood cultures. Am J Clin Pathol. 2006;126:686-690.
7. Church DL, Don JC, Unger B. Effects of restructuring on the performance of microbiology laboratories in Alberta. Arch Pathol Laboratory Med. 2000;124:357-361.
8. Samuel LP, Balada LJM, Harrington A, Cavagnolo R. Multicenter assessment of gram stain error rates. J Clin Microbiol. 2016;54:1442-1447.
9. Dagnaw AM, Tiruneh BZ. Most laboratory tests used in the diagnosis of tuberculosis in Ethiopia are based on direct microscopy: a systematic review. Health Care. 2014;3:4.
10. Woodcock S, Fine G, McClure K, Unger B, Rizzo PP. The role of standards and training in preparing for accreditation. Am J Clin Pathol. 2010;134:388-392.
11. Parikh RP. Competency assessment for medical laboratory practitioners and existing rules and regulations. J Health Occupations Education. 2000;14:6.
12. der Van VC, Schuwirth L, Scheele F, Driessen E, Hodges B. The assessment of professional competence: building blocks for theory development. Best Pract Res Clin Obstet Gynaecol. 2010;24:703-719.
13. Fonjungo PN, Kebede Y, Arneson W, Tefera D, Yimer K, Kinde S, et al. Preservice laboratory education strengthening enhances sustainable laboratory workforce in Ethiopia. Hum Resour Health. 2013;11:56.
14. Andualem M, Kebede G, Kumie A. Information needs and seeking behaviour among health professionals working at public hospital and health centres in Bahir Dar, Ethiopia. BMC Health Serv Res. 2013;13:534.
15. Hiwotu TM, Ayana G, Mulugeta A, Kassa GB, Kebede Y, Fonjungo PF, et al. Laboratory system strengthening and quality improvement in Ethiopia. Afr J Lab Med. 2014;3:1-6.
16. Sisay A, Mindaye T, Tesfaye A, Abera E, Desale A. Assessing the outcome of Strengthening Laboratory Management Towards Accreditation (SLMTA) on laboratory quality management system in city government of Addis Ababa, Ethiopia. Pan Afr Med J. 2015;20:314.
17. Desjardins M, Fleming CA. Competency assessment of microbiology medical laboratory technologists in Ontario, Canada. J Clin Microbiol. 2014;52:2940-2945.
18. Goodyear N, Kim S, Reeves M, Astion ML. A 2-year study of gram stain competency assessment in 40 clinical laboratories. Ame JClin Pathol. 2006;125:28-33.
19. Gurmessa A, Sisay A. Proficiency test feedback utilization at government hospitals laboratory, Addis Ababa, Ethiopia. J Med Diagn Meth. 2017;6:1-8.