


# Competency Assessment on Gram Stain Examination and Interpretation Among Medical Laboratory Professionals Working in Selected Hospitals of Addis Ababa, Ethiopia

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**Background:** Laboratory test results are the cornerstone for patient diagnosis and treatment. Gram staining is a classic laboratory test method used to differentiate between bacteria. Competence assessment can help identify gaps and provide suggestions to academics, researchers, and policymakers to address competency gaps. In Ethiopia, there is no evidence of competency assessment by medical laboratory professionals using the Gram-staining technique.

**Objective:** To assess the competency of medical laboratory professionals on Gram stain examination and interpretation in selected hospitals of Addis Ababa, Ethiopia.

**Methods:** A cross-sectional study was conducted to assess the competency of medical laboratory professionals on Gram stain examination and interpretation from September 2015 to December 2017.

**Results:** Of 190 participants, 55 (28.9%) participants scored low knowledge, 131 (68.9%) scored medium knowledge, and only 4 (2.1%) respondents scored high knowledge. From the study variables, education level, supervision by regional or federal government bodies, and training about Gram staining were significantly associated with the knowledge level of study participants. Forty eight (25.3%), 78 (41%), and 64 (33.7%) participants scored low, medium, and high skill level, respectively, from a total of 190 participants. From skill level analysis, hospital type, microscope type, and availability of health information resources were significantly associated with skill levels. There were 44 observations (4%) with major errors and 321 observations (28%) with very major errors from all 1140 observations. Of all observations, 321 (28.2%) reported without grading, 39 observations (3.4%) reported gram-positive bacteria as gram-negative bacteria, and 15 observations (1.4%) reported gram-negative bacteria as gram-positive bacteria.

**Conclusion:** The current study found that most medical laboratory professionals work without supervision or refresher training in Gram stain examination and interpretation. Hence, medical laboratory professionals' knowledge and skill levels are unsatisfactory. Regular competence assessments, training, and follow-up are necessary to improve the professional competence in medical laboratories.

**Keywords:** knowledge, skill, competence, gram stain and medical laboratory professionals

## Background

Laboratory tests are the cornerstone of patient diagnosis and treatment.<sup>1</sup> Gram staining is one of the oldest laboratory methods 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.<sup>2-4</sup>

Gram staining is routinely used as requested for clinical specimens submitted for smear and culture. It is used to characterize any specimen.<sup>5-7</sup> Gram staining provides immediate information regarding the presence or absence of bacterial infection and the morphotypes of the bacteria involved in the infection. This can be the baseline for treatment initiation and initial choice of antibiotic therapy.<sup>8,9</sup>

In Ethiopia, most medical laboratories in healthcare facilities cannot create microbial cultures to diagnose microbial infections. This is because of a lack of skilled manpower, infrastructure, cost of culture diagnostic materials, and other issues.<sup>10</sup> Hence, Gram staining is the method of choice for the detection of microorganisms in most healthcare facilities, as microbial culture is impossible or not accessible in most of these facilities.

Despite its importance, interpretation of Gram staining is difficult for medical laboratory professionals because such an interpretation requires multiple observations and judgment that comes with years of experience.<sup>11</sup> For Gram stain interpretation, a minor error may report the presence of a polymorph when not present, a major error may report the presence of an organism not present or fail to report the presence of 1+ to 2+ polymorphs, and a very major error may fail to report the presence of an organism and fail to report the presence of 3+ to 4+ polymorphs.<sup>8,12</sup>

According to CLIA '88 (Clinical Laboratory Improvement Amendments), medical laboratory personnel must be assessed for competency semiannually during the first year of employment and annually thereafter.<sup>13-15</sup> However, some challenges that clinical laboratories face today include the design and implementation of competency assessment programs.<sup>13</sup>

In Africa, including Ethiopia, there is no accessible mandated law on how the competencies of medical laboratory professionals should be assessed.

Scientific evidence from competency assessments of medical laboratory professionals can deliver important information for planning continuous professional development, training, and supervision. It also provides important evidence to higher institutions to better align their training and produce knowledgeable and skilled manpower. Generally, study in this area would be valuable for medical laboratory professional associations, higher institutions, government bodies, stakeholders, participating hospitals, and other healthcare facilities by showing addressable competency gaps in Gram stain examination and interpretation. This study assessed the competency of medical laboratory professionals on Gram stain examination and interpretation in the study area and deliver the scientific evidence that needs to be addressed by the responsible bodies.

## Materials and Methods

### Study Design, Study Period and Study Area

This cross-sectional study was conducted between September 2015 and December 2017 in Addis Ababa, Ethiopia. Addis Ababa is the capital city of Ethiopia and has 42 hospitals, of which 30 hospitals are private. The study was conducted in ten public hospitals (All Africa Leprosy Rehabilitation and Training Center (ALERT), St. Paul's Hospital Millennium Medical College, Amanuel Mental Hospital, Tikur Anbessa Specialized Hospital, Minilik II Hospital, Zewditu Memorial Hospital, Yekatit 12 Hospital Medical College, Gandhi Memorial Hospital, Tirunesh Beijing Hospital and Ras Desta Damtew Memorial Hospital), two uniformed hospitals (Armed Force Hospital and Police Hospital), and eight private hospitals (Kadisco General Hospital, St. Gebreal General Hospital, Addis Hiwot General Hospital, Amin General Hospital, Hayat General Hospital, Myung Sung Christian Medical Center, Yerer Primary Hospital, and Tzna General Hospital).

### Source Population

All medical laboratory professionals working in medical laboratories of hospitals in Addis Ababa, Ethiopia.

### Study Population

Medical laboratory professionals working in the laboratories of the selected hospitals in Addis Ababa fulfilled the inclusion criteria.

### Sample Size Determination

A preliminary assessment of the number of medical laboratory professionals in each hospital was performed prior to data collection. On average, there were five medical laboratory professionals in private hospitals, and the total number of

medical laboratory professionals working in private hospitals in Addis Ababa was approximately 150. In Government hospitals, approximately 331 medical laboratory professionals participated in the preliminary assessment. The total number of medical laboratory professionals working in hospitals in Addis Ababa is approximately 481. Nancy et al included 278 participants in a computer-based competence assessment study on Gram stains in the US. Similar studies in Ethiopia by Ayalew et al on malaria and by Hailemariam et al on TB included 80 and 81 participants, respectively.<sup>16–18</sup> We benchmarked the aforementioned studies and included 190 medical laboratory personnel.

## Sampling Methods

The sampling method used in this study was purposive sampling. Government and private hospitals in Addis Ababa were selected for this study.

## Data Collection Tools and Procedure

Six Gram-stained panel slides and 15 structured knowledge questions with background questionnaires were used in this study. One slide from the graded and interpreted stained slides was provided to each participant, and each participant was graded and interpreted on one stained slide within five minutes. Participants were also provided with 15 structured knowledge questions with a background questionnaire to fill in their responses within 20 minutes. The microscopes used for the study were checked for proper functionality by senior medical laboratory personnel at each laboratory and by the principal investigator. The principal investigator administered the data collection procedure.

## Panel Slide Preparation, Distribution and Quality Assurance

Bacteriology laboratory experts at the Armauer Hansen Research Institute (AHRI) and the principal investigator prepared and validated Gram-stained slides. Both patient samples and samples from the American Type Culture Collection (ATCC) were used to prepare Gram-stained panel slides. The known bacterial strains (ATCC) were subcultured on blood agar. Gram-stained panel slides were prepared from blood agar, and no organism panel slides were prepared from patient samples. All slides were stained with Gram stain. Experts interpreted the prepared Gram stain smears using investigative criteria for the presence or absence of Gram stain findings (bacteria, yeasts, and cells) and quantification of findings (few, moderate, and many).<sup>5,12,13</sup> Experts have validated the Gram stain interpretation agreement with the culture growth on blood agar. The validated Gram stain panel slide interpretation agreed with the culture growth. For this study, six gram-stained panel slide types were used, and the test panels were prepared in the AHRI Bacteriology Laboratory. The panel slides that were prepared included gram-positive cocci in the cluster *Staphylococcus aureus* (*S. aureus*), gram-positive cocci in the chain *Streptococcus pyogenes* (*S. pyogenes*), gram-negative diplococci *Neisseria gonorrhoea* (*N. gonorrhoeae*), gram-positive diplococci *Streptococcus pneumoniae* (*S. pneumoniae*) and gram-positive rods *Listeria monocytogenes* (*L. monocytogenes*) from ATCC, and no microorganisms with many pus cells from the patient samples. The slides were graded as few, moderate, or many with different interpretations. Next to preparation and validation, the principal investigator arranged, packed, and distributed slides to the participants.

Data quality was ensured through the use of standardized data collection materials, pretesting of the questionnaires, and intensive supervision during data collection.

## Data Entry and Analysis

Data were entered into RED Cap<sup>19</sup> and analyzed using the R software.<sup>20</sup> Bloom's cut-off point was used to measure respondents' knowledge and skills. Percent of correct responses to a set of 15 knowledge questions was graded as follows: 59% or below (8/15) as low, 60–80% (9–12/15) as medium, and above 80% (>12/15) as high knowledge level. Similarly, percentage of correct responses to a set of six skill-related questions was graded as follows: those who performed correctly in three or below 59% (3/6) as low, 60–80% (3–4.8/6) as medium, and above 80% (>4.8/6) as high skill level.<sup>21</sup> Statistical significance was determined based on a two-sided P-value < 0.05. The median, very major error, major error, minor error, maximum score, minimum score, and other errors were analyzed

for skill tests. For the 15 knowledge questions, analyses were also performed for the median, maximum, and minimum scores of exam answers.

## Operational Definitions

Gram stain interpretation minor error: report the presence of polymorphs when not present.<sup>5</sup>

Gram stain interpretation major error: report the presence of an organism not present or failure to report the presence of 1+ to 2+ polymorphs.<sup>5</sup>

Gram stain interpretation very major error: failure to report the presence of an organism and failure to report the presence of 3+ to 4+ polymorphs.<sup>5</sup>

Competency: The ability to carry out the total performance responsibilities of the given practitioner's generic position or the combined knowledge and skill factors necessary to fulfill work obligations adequately.<sup>13</sup>

## Results

### Sociodemographic and Other Characteristics of Respondents

A total of 190 medical laboratory professionals were included in this study. The median age of the respondents was 28 years, with an interquartile (IQR) range of eight years. The median experience of the participants was five years, with an IQR range of six years, while the median experience in the bacteriology laboratory was 0.5 years with IQR range of 3 years. Most of the study respondents were males, 103 (55.1%), and majority of the respondents, 98 (52.1%), had a first degree. One hundred thirty-eight (75%) participants were from government hospitals and most participants, 142 (78.5%), were from government higher institutions (Table 1).

**Table 1** Sociodemographic Characteristics of Medical Laboratory Professionals Working in Selected Hospitals of Addis Ababa, Ethiopia, 2017

Background Variables	Characteristics	Number	Percent (%)
Age in years (n <sup>2</sup> =186)	19–25	37	19.9%
	26–30	83	44.6%
	31–35	30	16.1%
	>=36	36	19.4%
	Unknown	4	2%
Sex (n =187)	Male	103	55.1%
	Female	84	44.9%
	Unknown	3	1.6
Hospital type (n =184)	Government	138	75%
	Private	40	21.7%
	Uniformed <sup>b</sup>	6	3.2%
	Unknown	6	3.1
Higher Institution type (n =181)	Government	142	78.5%
	Private	39	21.5%
	Unknown	9	4.7

(Continued)

**Table 1** (Continued).

Background Variables	Characteristics	Number	Percent (%)
Education level (n =188)	Masters	16	8.5%
	First degree	98	52.1%
	Diploma	74	39.4%
	Unknown	2	1

Notes: <sup>a</sup>n or N= number of study subjects. <sup>b</sup>Uniformed =Armed force and Police hospitals.

Eighty-one (42.6%) of the study participants were using a similar type of microscope. During the study period, most of the participants were working in hematology, 29 (15.4%), microbiology, 25 (13.3%), phlebotomy, 23 (12.2%), clinical chemistry, 24 (12.77%) and parasitology, 20 (10.6%) laboratory sections. The majority of the study participants, 139 (73.16%), were working without supervision on Gram stain examination and interpretation from regional or federal institutions. One hundred sixty-two (85.7%) of the respondents were working without in service training on Gram stain examination and interpretation. Most of the study participants, 123 (65.4%), accessed health information resources and most of respondents, 102 (68%), used these health information resources sometimes (Table 2).

**Table 2** Medical Laboratory Professionals Work Settings in Selected Hospitals of Addis Ababa, Ethiopia, 2017

Background Variables	Characteristics	Number	Percent (%)
Microscope type (n =190)	Tensido	3	1.6%
	Labomed	13	6.8%
	HumaScop	22	11.6%
	LEICA	14	7.4%
	Ecoline	4	2.1%
	Olympus	81	42.6%
	PrimoStar	53	27.9%
Working department (n = 188)	Parasitology	20	10.6%
	Serology	9	4.8%
	Clinical Chemistry	24	12.8%
	Phlebotomy	23	12.2%
	Microbiology	25	13.3%
	Management	7	3.7%
	All section	18	9.6%
	Blood Bank	10	5.3%
	Haematology	29	15.4%
	Urine Analysis	2	1.1%
	Others	21	11.2%
	Unknown	2	1

(Continued)

**Table 2** (Continued).

Background Variables	Characteristics	Number	Percent (%)
Supervision (n =190)	Yes	51	26.8%
	No	139	73.2%
Training on Gram stain (n =189)	Yes	27	14.3%
	No	162	85.7%
	Unknown	1	0.5
Health information resources (n =188)	Yes	123	65.4%
	No	65	34.6%
	Unknown	2	1
Frequency of health information used as resources	Always	26	17.3%
	Sometimes	102	68%
	Never	22	14.7%

## Knowledge of Medical Laboratory Professionals on Gram Stain Examination and Interpretation

For the theoretical knowledge questions, the minimum score was 3 (20%) and maximum score was 13 (86.7%). Of 190 participants, 55 (28.9%) participants scored with low knowledge level, 131 (68.9%) scored medium knowledge, and only 4 (2.1%) respondents were scored high knowledge.

Analysis was carried out to examine the association between different factors and the knowledge of study participants regarding Gram stain examination and interpretation. Education level, supervision by regional or federal government bodies, and training about Gram staining were significantly associated with the knowledge level of study participants. Of the four study participants with high knowledge level, 3 (75%) of them had second degrees (MSc) and one (25%) had a first degree level. The respondents with diplomas did not have a high level of knowledge (Table 3).

**Table 3** Knowledge Levels of the Medical Laboratory Professionals on Gram Stain Examination and Interpretation Associated with Background Characteristics in Hospitals in Addis Ababa, Ethiopia, 2017

Independent Variables	Characteristic	Knowledge Level			Total N (%)	$\chi^2$ <sup>C</sup>	P value
		Low N (%)	Medium N (%)	High N (%)			
Age	19–25	11(5.9%)	26(13.98%)	0(0%)	37(19.89%)	2.9	0.8
	26–30	26(13.98%)	54(29%)	3(1.6%)	83(44.6%)		
	31–35	9(4.84%)	21(11.3%)	0(0%)	30(16.1%)		
	>=36	9(4.84%)	26(13.98%)	1(0.5%)	36(19.4%)		
	Total N (%)	55(29.6%)	127(68.3%)	4(2.2%)	186		
Sex	Male	26 (13.9%)	73(39.1%)	4 (2.1%)	103 (55.1%)	4.4	0.1
	Female	28 (14.9%)	56(29.95%)	0(0%)	84(44.9%)		
	Total N (%)	54 (28.9%)	129(68.9%)	4 (2.1%)	187		

(Continued)

Table 3 (Continued).

Independent Variables	Characteristic	Knowledge Level			Total N (%)	$\chi^2$ C	P value
		Low N (%)	Medium N (%)	High N (%)			
Hospital Type	Government	42(22.8%)	92(50%)	4 (2.2%)	138(75%)	3.1	0.5
	Private	10(5.4%)	30(16.3%)	0(0%)	40(21.7%)		
	Uniformed	3 (1.6%)	3(1.6%)	0(0%)	6(3.3%)		
	Total N (%)	55(29.9%)	125(67.9%)	4(2.2%)	184		
Higher Institution Type	Government	47(25.9%)	92(50.8%)	3(1.7%)	142(78.5%)	3.4	0.2
	Private	8(4.4%)	31(17.1%)	0(0%)	39(21.6%)		
	Total N (%)	55(30.4%)	123(67.96%)	3 (1.7%)	181		
Microscope Type	Tensido	0(0%)	3(1.6%)	0(0%)	3(1.6%)	7.2	0.8
	Labomed	3(1.6%)	10(5.3%)	0(0%)	13(6.8%)		
	HumaScop	6(3.2%)	16(8.4%)	0(0%)	22(11.6%)		
	LEICA	3(1.6%)	11(5.8%)	0(0%)	14(7.4%)		
	Ecoline	0(0%)	4(2.1%)	0(0%)	4(2.1%)		
	Olympus	24(12.6%)	55(28.9%)	2(1.1%)	81(42.6%)		
	PrimoStar	19(10%)	32(16.8%)	2 (1.1%)	53(27.9%)		
	Total N (%)	55(28.9%)	131(68.9%)	4(2.1%)	190		
Education level	Masters	2 (1.1%)	11(5.9%)	3(1.6%)	16(8.5%)	24.6	0.0000006
	First Degree	29(15.4%)	68(36.2%)	1(0.5%)	98(52.1%)		
	Diploma	23(12.2%)	51(27.1%)	0(0%)	74(39.36%)		
	Total N (%)	54(28.7%)	130(69.2%)	4 (2.1%)	188		
Working Department	Parasitology	6(3.2%)	14(7.5%)	0(0%)	20(10.6%)	14.8	0.79
	Serology	3(1.6%)	5(2.7%)	1(0.5%)	9(4.8%)		
	Clinical Chemistry	6(3.2%)	17(9%)	1(0.5%)	24(12.8%)		
	Phlebotomy	6(3.2%)	17(9%)	0(0%)	23(12.2%)		
	Microbiology	9(4.8%)	14(7.5%)	2(1.1%)	25(13.3%)		
	Management	1(0.5%)	6(3.2%)	0(0%)	7(3.7%)		
	All section	5(2.7%)	13(6.9%)	0(0%)	18(9.6%)		
	Blood Bank	2(1.1%)	8(4.3%)	0(0%)	10(5.3%)		
	Haematology	10(5.3%)	19(10.1%)	0(0%)	29(15.4%)		
	Urine Analysis	1(0.5%)	1(0.5%)	0(0%)	2(1.1%)		
	Others	5(2.7%)	16(8.5%)	0(0%)	21(11.2%)		
	Total N (%)	54(28.7%)	130(69.2%)	4(2.1%)	188		

(Continued)

**Table 3** (Continued).

Independent Variables	Characteristic	Knowledge Level			Total N (%)	$\chi^2$	P value
		Low N (%)	Medium N (%)	High N (%)			
Supervision By Federal (Regional) institution	Yes	14(7.4%)	33(17.4%)	4(2.1%)	51(26.8%)	11.1	0.004
	No	41(21.6%)	98(51.6%)	0(0%)	139(73.2%)		
	Total N (%)	55(28.9%)	131(68.9%)	4(2.1%)	190		
Training on Gram stain	Yes	6(3.2%)	18(9.5%)	3(1.6%)	27(14.3%)	2.1%	0.002
	No	49(25.9%)	112(59.3%)	1(0.5%)	162(85.7%)		
	Total N (%)	55(29.1%)	130(68.8%)	4(2.1%)	189		
Health Information Resources	Yes	35(18.6%)	84(44.7%)	4(2.1%)	123(65.4%)	2.2	0.3
	No	19(10.1%)	46(24.5%)	0(0%)	65(34.6%)		
	Total N (%)	52(28.7%)	130(69.1%)	4(2.1%)	190		
Frequency of health information used as resources	Always	8(5.3%)	16(10.7%)	2(1.3%)	26(17.3%)	3.9	0.4
	Sometimes	29(19.3%)	71(47.3%)	2(1.3%)	102(68%)		
	Never	5(3.3%)	17(11.3%)	0(0%)	22(14.7%)		
	Total N (%)	42(28%)	104(69%)	4(2.7%)	150		

Notes:  $\chi^2$  = Chi square.

Among study respondents with low knowledge, most of the respondents 41(74.6%) with low knowledge were those who had no supervision. From all study participants, there were 41(21.6%) participants who had no supervision and low knowledge. Of all high knowledge participants, the majority 3(75%) had training on Gram staining, whereas of all participants with low knowledge levels, most of them 49(89.1%) had no training on Gram staining. Of all study participants, 25.9% had no training in Gram staining and had low knowledge (Table 3).

For this study, female participants, participants working in private hospitals, who had learned in private higher institutions, had used some microscope types (Tensido, Labomed, HumaScop, LEICA, and Ecoline) and participants who had no access to health information resources did not have a high level of knowledge (Table 3).

### Skill of Medical Laboratory Professionals on Gram Stain Examination and Interpretation

Of the 190 participants administered with six skill related tests, the maximum score was 6 (100%) and minimum score was 0 (0%). Forty-eight (25.3%), 78 (41%), and 64 (33.7%) participants had low, medium, and high scores, respectively. Hospital type, microscope type, and availability of health information resources were significantly associated with skill levels ( $p = 0.0009$ ,  $0.04$ , and  $0.01$ , respectively). However, the other independent variables did not show a statistically significant association with the skill levels of the respondents (Table 4).

All study subjects from uniformed hospitals had high skill levels in Gram stain examination and interpretation. Among participants with low skill level and medium skill levels, most of the respondents were from government hospitals (43(89.6%) and 52(70.3%), respectively). Participants who were using microscope-type Labomed and Ecoline had no low skill in Gram stain examination and interpretation. However, participants who were using microscope type Tensido had no high skill level in Gram stain examination and interpretation. Of the respondents who had access to health information resources, most of them scored medium and high skill levels on Gram stain examination and interpretation (49 (39.9%) and 50 (40.7%), respectively). Furthermore, the majority of those who had no access to health information resources on Gram stain scored low and medium skill levels on Gram stain examination and interpretation (23 (35.4%) and 28 (43.1%), respectively) (Table 4).



**Table 4** Skill Level of the Medical Laboratory Professionals on Gram Stain Examination and Interpretation Associated with Background Characteristics in Hospitals in Addis Ababa, Ethiopia, 2017

Independent Variable	Characteristic	Skill Level			Total N (%)	$\chi^2$	P value
		Low N (%)	Medium N (%)	High N (%)			
Age	19–25	9(4.8%)	15(8.1%)	13(6.9%)	37(19.9%)	3.4	0.8
	26–30	23(12.4%)	34(18.3%)	26(13.9%)	83(44.6%)		
	31–35	4(2.2%)	14(7.5%)	12(6.45%)	30(16.1%)		
	>=36	11(5.9%)	15(8.1%)	10(5.4%)	36(19.4%)		
	Total N (%)	47(25.3%)	78(41.9%)	61(32.8%)	186		
Sex	Male	23(12.3%)	41(21.9%)	39(20.9%)	103(55.1%)	1.47	0.5
	Female	23(12.3%)	36(19.3%)	25(13.4%)	84(44.9%)		
	Total N (%)	46(24.6%)	77(41.2%)	64(34.2%)	187		
Hospital Type	Government	43(23.4%)	52(28.3%)	43(23.4%)	138(75%)	18.7	0.0009
	Private	5(2.7%)	22(11.9%)	13(7.1%)	40(21.7%)		
	Uniformed	0(0%)	0(0%)	6(3.3%)	6(3.3%)		
	Total N (%)	48(26.1%)	74(40.2%)	62(33.7%)	184		
Higher Institution Type	Government	38(20.9%)	58(32%)	46(25%)	142(78%)	0.54	0.8
	Private	9(4.9%)	15(8.3%)	15(8.3%)	39(21.6%)		
	Total N (%)	47(25.9%)	73(40%)	61(33.7%)	181		
Microscope Type	Tensido	2(1%)	1(0.5%)	0(0%)	3(1.6%)	21.6	0.04
	Labomed	0(0%)	4(2%)	9(4.7%)	13(6.8%)		
	HumaScop	6(3.2%)	11(5.8%)	5(2.6%)	22(11.6%)		
	LEICA	1(0.53%)	9(4.7%)	4(2%)	14(7.4%)		
	Ecoline	0(0%)	3(1.6%)	1(0.5%)	4(2%)		
	Olympus	21(11%)	30(15.8%)	30(15.8%)	81(42.6%)		
	PrimoStar	18(9.5%)	20(10.5%)	15(7.9%)	53(27.9%)		
	Total N (%)	48(25.3%)	78(41.1%)	64(33.7%)	190		
Education level	Masters	4(2.1%)	4(2.1%)	8(4.3%)	16(8.5%)	4.2	0.4
	First Degree	24(12.8%)	46(24.5%)	28(14.9%)	98(52.13%)		
	Diploma	19(10.1%)	28(14.9%)	27(14.4%)	74(39.4%)		
	Total N (%)	47(25%)	78(41.5%)	63(33.5%)	188		

(Continued)

**Table 4** (Continued).

Independent Variable	Characteristic	Skill Level			Total N (%)	$\chi^2$	P value
		Low N (%)	Medium N (%)	High N (%)			
Working Department	Parasitology	3(1.6%)	11(5.9%)	6(3.2%)	20(10.6%)	25.5	0.2
	Serology	3(1.6%)	1(0.5%)	5(2.7%)	9(4.8%)		
	Clinical Chemistry	7(3.7%)	11(5.9%)	6(3.2%)	24(12.8%)		
	Phlebotomy	5(2.7%)	14(7.5%)	4(2.1%)	23(12.2%)		
	Microbiology	5(2.7%)	9(4.8%)	11(5.9%)	25(13.3%)		
	Management	3(1.6%)	4(2.1%)	0(0%)	7(3.7%)		
	All section	4(2.1%)	6(3.2%)	8(4.3%)	18(9.6%)		
	Blood Bank	1(0.5%)	2(1.1%)	7(3.7%)	10(5.3%)		
	Haematology	8(4.3%)	9(4.8%)	12(6.4%)	29(15.4%)		
	Urine Analysis	1(0.5%)	1(0.5%)	0(0%)	2(1.1%)		
	Others	7(3.7%)	9(4.8%)	5(2.7%)	21(11.2%)		
	Total N (%)	47(25%)	77(40.9%)	64(34.1%)	188		
Supervision By Federal (Regional) institution	Yes	10(5.3%)	20(10.5%)	21(11%)	51(26.8%)	2.10	0.3
	No	38(20%)	58(30.5%)	43(22.6%)	139(3.1%)		
	Total N (%)	48(25.3%)	78(41.05%)	64(33.7%)	190		
Training on Gram stain	Yes	7(3.7%)	13(6.9%)	7(3.7%)	27(14.3%)	1.01	0.6
	No	41(21.7%)	64(33.9%)	57(30.2%)	162(85.7%)		
	Total N (%)	48(25.4%)	77(40.7%)	64(33.9%)	189		
Health Information resources	Yes	24(12.8%)	49(26.1%)	50(26.6%)	123(65.4%)	8.95	0.01
	No	23(12.2%)	28(14.9%)	14(7.5%)	65(34.6%)		
	Total N (%)	47(25%)	77(40.9%)	64(34.1%)	188		
Frequency of health information used as resources	Always	7(4.7%)	6(4%)	13(8.7%)	26(17.3%)	5.6	0.2
	Sometimes	17(11.3%)	47(31.3%)	38(25.3%)	102(68%)		
	Never	6(4%)	9(6%)	7(4.7%)	22(14.7%)		
	Total N (%)	30(20%)	62(41.3%)	58(38.7%)	150		

## Errors of Medical Laboratory Professionals on Gram Stain Examination and Interpretation

There were 44 observations (4%) with major errors and 321 observations (28%) with very major errors from all 1140 observations. Of all observations, 321 (28.2%) reported without grading, 39 observations (3.4%) reported gram-positive bacteria as gram-negative bacteria, and 15 observations (1.4%) reported gram-negative bacteria as gram-positive bacteria. One hundred forty-eight observations (12.9%) also reported without grading with gram-positive bacteria reported as gram-negative bacteria and forty-three observations (3.7%) reported without grading with gram-negative bacteria reported as gram-positive bacteria (Table 5).

**Table 5** The Level of the Error of the Medical Laboratory Professionals on Gram Stain Examination and Interpretation for Skill Test in Hospitals in Addis Ababa, Ethiopia, 2017

Error Type p	Slides Code des Cod	Characteristics	Number	Percent
Errors	GSS-01	Very major error	15	7.9%
		No error	175	92.1%
	GSS-02	Very major error	22	11.6%
		No error	168	88.4%
	GSS-03	Very major error	20	10.5%
		No error	170	89.5%
	GSS-04	Very major error	131	68.9%
		No error	59	31.1%
	GSS-05	Very major error	32	16.8%
		No error	158	83.2%
	GSS-06	Major error	44	23.1%
		Very major error	101	53.1%
		No error	45	23.7%
	All (N=190×6=1140)	Major error	44	4%
		Very major error	321	28%
No error		775	68%	
Other Error Types	GSS-01	No grading	63	33.2%
		Gram positive reported as Gram negative	19	10%
		Both	59	31%
		No Error	49	25.9%
	GSS-02	No grading	70	36.8%
		Gram positive reported as Gram negative	15	7.9%
		Both	60	31.6%
		No Error	45	23.7%
	GSS-03	No grading	96	50.5%
		No grading and Gram positive reported as Gram negative	14	7.4%
		No Error	80	42.1%
	GSS-04	No grading	26	13.7%
		Gram positive reported as Gram negative	5	2.6%
		Both	15	7.9%
		No Error	144	75.8%

(Continued)

**Table 5** (Continued).

Error Type p	Slides Code des Cod	Characteristics	Number	Percent
	GSS-05	No grading	62	32.6%
		Gram negative reported as Gram positive	15	7.9%
		Both	43	22.7%
		No Error	70	36.8%
	GSS-06	No grading	4	2.1%
		No error	186	97.9%
	All(N=190×6=1140)	No grading	321	28.2%
		Gram positive reported as Gram negative	39	3.4%
		No grading and Gram positive reported as Gram negative	148	12.9%
		Gram negative reported as Gram positive	15	1.4%
		No grading and Gram negative reported as Gram positive	43	3.7%
		No error	574	50.4%

## Association of Knowledge and Skill Level of Medical Laboratory Professionals on Gram Stain Examination and Interpretation

There was a statistically significant association between the knowledge and skill level of the study participants on the Gram stain examination and interpretation ( $p = 0.006$ ). Those who had a high knowledge level scored a high skill level on the Gram stain examination and interpretation. Of all study participants, 21 (11%) scored low knowledge and skill levels, 60 (31.6%) had medium knowledge level and medium skill levels, and 4 (2.1%) had high knowledge and skill levels (Table 6).

**Table 6** Association of Knowledge and Skill Level of the Medical Laboratory Professionals on Gram Stain Examination and Interpretation in Hospitals in Addis Ababa, Ethiopia, 2017

Knowledge Vs Skill Level	Low Skill Level	Medium Skill Level	High Skill Level	Total	$\chi^2$	P value
Low knowledge Level	21 (11%)	18 (9.5%)	16 (8.4%)	55 (28.9%)	14.6	0.006
Medium knowledge level	27 (14.2%)	60 (31.6%)	44 (23.2%)	131 (68.9%)		
High knowledge level	0 (0%)	0 (0%)	4 (2.1%)	4 (2%)		
Total	48 (25.3%)	78 (41%)	64 (33.7%)	190		

## Discussion

To the best of our knowledge, this study is the first to assess the competence of medical laboratory professionals in Gram stain examination and interpretation using both knowledge and skill tests. Although no similar studies have been published to compare our findings, we found that most of the study participants were working without supervision and refresher training on Gram stain examination and interpretation. However, according to CLIA' 88, medical laboratory professionals must be assessed for competence semi-annually during the first year of employment and annually thereafter.<sup>14</sup> Our results showed that the competence of medical laboratory professionals in Gram stain examination and interpretation lacks attention from the responsible bodies. A survey conducted in Canada by Desjardins et al also indicated that the most common competency issue requiring remediation was associated with gram staining and interpretation.<sup>22</sup>

The findings of this study revealed that very few participants had high levels of knowledge. This may be because of a lack of supervision and training. However, a study in the US using computer-based Gram stain competency assessment on 278 study subjects reported that overall users correctly identified approximately 90% of the Gram stain items.<sup>16</sup> This may be because of appropriate supervision, training, and access to an advanced laboratory setting.<sup>1</sup>

We found that educational level, supervision, and training on Gram staining had statistically significant associations with the knowledge level of the study participants. Generally, the study showed that education level, training and supervision have an impact on the Gram staining knowledge level of medical laboratory professionals.

Despite the fact that most of the participants were working without training, we found that training had an impact on the Gram stain knowledge level of study participants. However, in this study, training did not affect participants' skill levels. A previous study in the US reported that training and in-service education were correlated with the skills of medical laboratory professionals in interpreting Gram stained smears. It showed that those who had training had a high-level skill level compared to those who had no training in Gram stain interpretation.<sup>23</sup> The difference from the US study may be due to different factors that affect the knowledge and skill level of medical laboratory professionals in Gram stain interpretation. Moreover, our panels of slides were prepared from live bacterial cultures, unlike the US one, which could have its own impact on the difference.

The study results showed that hospital type, microscope type, and health information resource availability affected the skill level of participants in Gram stain interpretation. In this study, subjects from uniform hospitals had high skills in Gram stain examination and interpretation. In our study, the skill levels of the study participants were different at different sites and were significantly associated with the type of hospital. Similarly, a study conducted in the US indicated that the Gram stain error rate varied significantly depending on the site.<sup>24</sup>

Our results also indicate that health information resource availability can improve the skills of medical laboratory professionals in Gram stain examination and interpretation. However, if health information resources are not available, it leads to poor skills in Gram stain examination and interpretation.

In our study, we found that major errors, very major errors, gram-positive bacteria were reported as gram-negative bacteria and gram-negative bacteria as gram-positive bacteria. Similarly, in Ethiopia, a study conducted in 12 hospital laboratories participated in the national PT program from six cycles in 2012 and 2013, which had high PT failure rates from 20 test parameters. In this study Gram staining failure rate was 88.9%.<sup>25</sup> However, a study conducted in the US, from 5885 observations read as gram-positive cocci, six (0.1%) had gram-negative organisms by culture, and from 1959 read as gram-negative bacilli, 25 (1.3%) had gram positive organisms by culture.<sup>24</sup>

## Conclusion

There were misinterpretations of the Gram stain results, including major errors, very major errors, reporting gram-positive bacteria as gram-negative bacteria, and gram-negative bacteria as gram-positive bacteria. This lack of proper Gram stain examination and interpretation could lead to misdiagnosis and mistreatment of patients, which could cause drug resistance, resource wastage, suffering, and the death of patients.<sup>15,16,26</sup>

## Recommendation

- Practice focused training is needed to reduce the error rates of Gram stain examination and interpretation and to improve knowledge and skill level of medical laboratory professionals on Gram stain examination and interpretation.
- Supervision by regional or federal institutions and stakeholders' is necessary to improve the knowledge and skill of medical laboratory professionals on Gram stain examination and interpretation.
- Improving and encouraging access to health information resources is important to improve the knowledge and skill levels on Gram stain examination and interpretation.

## Availability of data and materials

Data supporting the findings of this study are available from the Armauer Hansen Research Institute. However, restrictions apply to the availability of these data, which were used under license in the current study and thus are not publicly available. However, the data are available from the authors upon reasonable request and with permission from the Armauer Hansen Research Institute.

## Ethical approval

This study was approved by the Departmental Research and Ethics Review Committee (DRERC) of the Department of Medical Laboratory Sciences, College of Health Sciences, Addis Ababa University, and the Armauer Hansen Research Institute Ethics Review Committee. This study was conducted in accordance with the ethical guidelines. Written informed consent was obtained from all participants.

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## Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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## Disclosure

The authors declare that they have no competing interests in this work.

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