# Knowledge, Attitude and Practice with Regard to Gram stain, as a Preliminary Test for Clinical Diagnosis among the Internees and Postgraduates, in a Tertiary Care Teaching Hospital

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#### ABSTRACT

## BACKGROUND

Hans Christian Gram, a Danish pathologist originally devised the Gram staining method in 1882 for identification of Gram positive and Gram-negative bacteria and is the gold standard method for preliminary detection before inoculating for culture and sensitivity for better treatment outcome. In the present era, use of irrational antimicrobials has led to antimicrobial resistance which can be resolved by routine usage of Gram stain.

## METHODS

This is a cross sectional study of 2 months duration (May 2020 - June 2020) with 172 subjects. Institutional ethical clearance and pharmaco-vigilance program of India permission was obtained. All interns and post-graduates who were available at the time of interview, and who gave written informed consent were included in the study. Those who did not give written informed consent and those who were absent or unavailable were excluded. Pre tested questionnaire was validated using Cronbach's alpha for internal consistency and was filled voluntarily. Data was collected, compiled and analysed for results.

## RESULTS

Among 172 subjects who participated in the study, 99 (57 %) are females and 73 (43 %) are males. 88 (51 %) are post-graduates and 84 (49 %) are interns. 100 % had knowledge about Gram stain, 47 (27 %) participants knew about procedure timing as 5 minutes for Gram staining and 26 (15 %) participants thought that test can be done at bedside. 90 (52 %) knew that test can be done for all specimens. 169 (98 %) had knowledge on the Gram stain which could help for provisional diagnosis. 119 (69 %) agreed that test was very cost effective. 28 (16 %) agreed that Gram stain was a better test for provisional diagnosis. 63 (36.6 %) were practicing regularly and 10 (6 %) were practicing Gram stain for appropriate antimicrobial use.

## CONCLUSIONS

The present study concluded 100 % of the study participants had knowledge with regard to Gram staining, 27 % knew that the time taken was 5 minutes to perform test. 15 % knew that it can be done at bedside. 63 (36.6 %) were practicing regularly and 10 (6 %) were practicing as initial step for all clinical confirmations for appropriate antimicrobial treatment for better outcome.

## **KEYWORDS**

Gram Stain, Preliminary Test, Antimicrobial Stewardship, Evidence Based Medicine, Better Clinical Approach

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## BACKGROUND

Hans Christian Gram, the Danish pathologist during 1882, discovered Gram staining technique while working on infected lung tissue from cadavers, used for more than 145 years. He found that some bacteria after staining had retained stain preferentially and identified as Gram-positive and Gram-negative, which lead to the golden era for microbiology in clinical diagnosis. He thought that this method of staining was imperfect, but until today it is the gold standard in differentiating microorganisms for all clinical specimens.1 Gram staining procedure is the most costeffective initial approach procedure in assessment of community-acquired respiratory infections, which help in directing patients for hospitalisation.<sup>2</sup> Present generation of the health care professionals are facing more of antibioticresistant strains called superbugs as evolving mutant forms, due to irrational use of antibiotics. A simple procedure like Gram stain would help in rapid preliminary evidence-based medicine to authenticity in clinical diagnosis, advising culture and sensitivity for all clinical samples which would help to prescribe appropriate antibiotics.<sup>3,4</sup>

A lot of additions have been made as guidelines in identification reading the morphology and of microorganisms in the past for the sake of nonmicrobiologist to identify, based on Gram staining for early detection.<sup>5</sup> Therefore, the present need for this study is to test the knowledge, attitude and practice on Gram staining procedure by internees and postgraduates as a tool for better clinical diagnosis and treatment approach, and to improve the quality of life, who suffer from infections and to reduce time of hospital stay. Development of antibiotic policy in hospitals and use of culture sensitivity pattern in treatment plays a major role in decreasing mortality and morbidity.

We wanted to assess the knowledge, attitude and practice with regard to Gram staining, among internees and postgraduates.

## METHODS

It's a cross-sectional, observational hospital-based study conducted for a period of 2 months from May 2020 – June 2020. This study was carried at Santhiram Medical College, Nandyal, Andhra Pradesh.

Before starting the study, Institutional Ethical Committee permission was obtained reference no: IEC/2020/52. This study was briefed well in advance to the field staff and trained to collect data separately, by taking precautions and standardise the information collection process in order to maximize reliability and minimize the bias. All internees and post-graduates who gave written informed consent were included and rest were excluded from the study. A pilot study was conducted before starting the main study. The pre-tested KAP (Knowledge, Attitude and Practices) questionnaire was given to all the subjects to fill voluntarily, which took 10 minutes to complete. Questionnaire was validated using Cronbach's alpha for internal consistency using SPSS version 24. The values obtained  $\alpha \ge 0.7$  were considered after running the software and rest were eliminated.

#### Sampling and Sample Size

Using convenient sampling we have chosen 172 subjects, 84 were internees and 88 were postgraduates from all departments who were willing to participate in the study. The subjects were enrolled using simple random sampling method.

#### **Data Collection and Statistical Analysis**

Collected data was compiled and cleansing was done to eliminate data duplication. Data was imported from Excel spread sheet to data view of SPSS version 24 and was subjected for analysis. Final results were tabulated and statistically analysed to obtain frequency tables along with their percentages.

#### RESULTS

Variable	Count	Percent		
	Gender			
Females	99	57		
Males	73	43		
Total	172	100		
Designation				
Internees	84	48.8		
Post Graduates	88	51.2		
Total	172	100		
Table 1 Distribution of Frequency Table				

for Gender and Designation

Variable	Count	Percent			
Knowledge on Gram staining?					
Yes	172	100			
No	0	0			
Time ta	Time taken for Gram staining?				
3 minutes	13	7.6			
5 minutes	47	27.3			
10 minutes	85	49.4			
15 minutes	9	5.2			
20 minutes	18	10.5			
Total	172	100			
Gram staining is done at?					
Microbiology Lab	134	77.9			
Pathology Lab	8	4.7			
Bed side Ward Lab	30	17.4			
Total	172	100			
Which Specimens are subjected For Gram staining?					
Urine	7	4.1			
Blood	32	18.6			
Exudates	29	16.9			
Fluid taps	14	8.1			
All	90	52.3			
Total	172	100			
Will Gram staining help in Early Diagnosis?					
Yes	169	98.3			
No	3	1.7			
Total	172	100			
What is the Cost for Gram staining?					
Costly	3	1.7			
Less cost	119	69.2			
No Idea	50	29.1			
Total	172	100			
Table 2. Knowledge on Gram Staining					

Out of 172 subjects, 99 (57 %) were females and 73 (43 %) were males. 88 (51 %) were post-graduates, and 84 (49 %) were interns (Table 1). 100 % had knowledge of Gram stain. 47 (27 %) knew the time taken is 5 mins, 26 (15 %) thought test could be done at bedside lab which is attached adjacent to wards. 90 (52 %) knew Gram stain is done for all specimens. 169 (98 %) knew that Gram stain could help in provisional diagnosis, and 119 (69 %) agreed it is low cost (Table 2). 162 (94 %) had an attitude to advice Gram stain and 98 (57 %) felt, needed for all specimens (Table 3). 63 (36.6 %) were practicing regularly, but only 10 (6 %) followed practicing for all clinical specimens. 28 (16 %) had agreed for Gram stain, as better test for provisional diagnosis (Table 4).

Do you wish to advice Gram staining?				
Yes	162	94.2		
No	10	5.8		
Total	172	100		
Are you willing to subject these following specimens for Gram staining?				
For All Specimens				
Yes	98	57.0		
No	74	43.0		
Blood				
Yes	34	19.8		
No	138	80.2		
Exudates				
Yes	26	15.1		
No	146	84.9		
Fluid taps				
Yes	14	8.1		
No	158	91.9		
Table 3. Attitude to Advice Gram Staining				

How often do you advice Gram staining?				
	Count	Percent		
Regularly	73	42.4		
Occasionally	99	57.6		
Total	172	100		
For All Specimens				
Yes	10	5.8		
No	162	94.2		
Total	172	100		
Which is the Better test advised?				
Culture	95	55.2		
Gram stain	28	16.3		
ELISA	29	16.9		
PCR	20	11.6		
Total	172	100		
Table 4. Practices Regarding Gram Staining				

## DISCUSSION

Among 172 subjects who participated in the study, 99 (57 %) were females and 73 (43 %) were males. In this present study female participation is more compared to males, which is exactly same compared to the workshop study conducted by Matthew S. on medical students on competency in comprehension and memorisation on Gram stain technique in the year 2016.<sup>6</sup> Among subjects participated 88 (51 %) were post-graduates, and 84 (49 %) were interns. All the participants in the study had 100 % knowledge of Gram stain. 47 (27 %) of them knew the time taken is 5 minutes to perform the test and 26 (15 %) had knowledge on Gram stain test could be done at the bedside lab attached. 90 (52 %) study subjects knew that Gram stain is done for all specimens and the same was observed in one of the studies

by Yunusa Thairu et. al, in his study concluded that all clinical specimens considered to be infectious are subjected to Gram staining either directly or indirectly help to assess preliminary diagnosis and antimicrobial therapy selected after collecting culture specimens and before final identification of the microorganism.<sup>7</sup> 169 (98 %) knew that Gram stain could help in provisional diagnosis, and 119 (69 %) agreed it is low cost, Becerra SC, et. al, in their study concluded that Gram staining results in a low cost and timeefficient technique for identifying bacteria,<sup>8</sup> and effective in rapid diagnosis, Tomohiro Taniguchi et. al, in their study concluded that Gram staining has reduced overuse of antimicrobials and reduced the cost of drugs by half for clinical conditions.<sup>9</sup>

Therefore, Gram staining is the most opted test and is low cost which helps the internees and postgraduates to speed up for early diagnosis and also lessens the financial burden on the patients and also reduces the hospital stay time to improve quality of life. Even Richard B. Thomson, in his study during 2016 described that clinicians were mostly depending on Gram staining at bedside, which was performed by interns for preliminary rapid diagnosis for all clinical specimens like urine, CSF (Cerebro-Spinal Fluid), and all infective specimens and results were showing more than compared to other investigations.<sup>10</sup> Gram staining is the only test considered to be important for certain clinical specimens such as urine, CSF, vaginal smears and certain exudates for diagnosing for microorganisms which are the leading cause of undiagnosed asymptomatic infections like asymptomatic bacteriuria, in bacterial meningitis, etc. 162 (94 %) had an attitude to advice Gram stain, and 98 (57 %) felt, needed for all specimens. One of the studies conducted by Enver Vardar et. al, has compared Gram stain with pap smear in the diagnosis of bacterial vaginosis, found that Gram stain was no less than to pap smear and which required more time for reporting than Gram staining. In another study conducted by Adriaanse AH et. al, concluded that the Gram staining has great significance in rapid intrapartum screening for maternal carriership of group B streptococci in women during labour which helps in reduction of postpartum infection leading to puerperal sepsis.

Subjects in this study were having attitude for advising Gram staining but only 57 % were agreeing Gram staining for all clinical specimens. 28 (16.3 %) of the participants were practicing regularly to advice Gram staining, but only 10 (6 %) followed practicing for all specimens. This shows that it is very less percentage of the participants who had complete knowledge on Gram staining but practically they are not following to advice regularly for clinical specimens. Only 6 % of the participants in this study were advising Gram staining test for all the clinical specimens, who shows very less percentage considering the test as least important and therefore it is the time for regular implantation of more of awareness programs and need to conduct regular workshops on Gram staining to all internees and postgraduates. 28 (16.3 %) thought Gram stain is better to test for clinical diagnosis. In this present study though 16.3 % agreed that Gram stain is better test for clinical diagnosis and the same was proved by Thairu Y et. al, titled laboratory perspective of Gram staining and its significance in

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investigations of infectious diseases, finally concluded that Gram stain is considered most important and most significant as lab diagnosis in undiagnosed infections.<sup>7</sup>

Therefore, this study on knowledge, attitude and practice regarding Gram stain, in allowing internees and postgraduates in achieving good approach to clinical diagnosis in patient care area with regard to usage of appropriate antimicrobials to achieve better treatment outcome by reducing irrational antimicrobial usage, following evidence-based medicine, to improve quality of life and improving patient turn over time. The same Gram stain importance on knowledge and perception for better clinical approach was studied by W J Steinbach et. al, in their study concluded that internees or house staff, physicians should receive formal rigorous training in the interpretation of the Gram stain and confident use of clinical microbiological knowledge may allow greater precision in clinical diagnosis to reduce irrational use of antimicrobials to decrease antimicrobial resistance in this present era.<sup>11</sup>

## Limitations

Interns and postgraduates participated less in number and the outcome of the study is limited to one institution. Needs a greater participation from students, interns and postgraduates and also all clinicians from all institutions across the nations.

## CONCLUSIONS

100 % knew about Gram stain; only 27 % knew the time taken for performing Gram stain is 5 minutes and 15 % thought it could be done at the bedside. Though 94 % had an attitude to advice, 16.3 % were advising Gram staining regularly. Only 6 % were advising Gram staining for all clinical specimens for better diagnosis and treatment outcome. Interns and postgraduates are in the need of regular training with regard to procedure and interpretation of the Gram staining for early diagnosis and to stop irrational use of antimicrobials and to follow proper antimicrobial protocol for better quality of life and to improve patient turn over time.

## Recommendations

- All clinicians should adopt Gram staining for all infective pathology at the bedside before starting antibiotics.<sup>7</sup>
- Gram staining should be performed for all infective cases by internees and postgraduates, as part of the skill and curriculum and need regular workshops.<sup>6,11</sup>
- Irrational use of antibiotics should be stopped thereby helping in reducing antimicrobial resistance.<sup>12</sup>
- Follow evidence-based medicine.<sup>7</sup>
- Detect asymptomatic bacteriuria in antenatal mothers for early detection of UTI's.<sup>13</sup>
- Gram stain helps in early diagnosis of bacterial vaginosis in women which is 97 % sensitive on par with Pap smear which is 93 %.<sup>14</sup>

- Helps in rapid intra-partum maternal screening for group B *Streptococcus* carrier state.<sup>15</sup>
- Should be recommended as part of skill development in the curriculum based medical education.

Data sharing statement provided by the authors is available with the full text of this article at jebmh.com.

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