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QUALITY ASSESSMENT OF SPUTUM GRAM STAIN IN RELATION TO SPUTUM CULTURE FOR LOWER RESPIRATORY TRACT INFECTIONS IN A TERTIARY CARE CENTRE KANPUR

Deepak Sameer Bind[1*], Jayesh Pandey[2], R Sujatha Arunagiri[3]

[1*]Assistant Professor, Department of Microbiology KMC Medical College & Hospital Maharajganj, U.P deepaksameer0007@gmail.com
[2]Associate Professor, Department of Microbiology KMC Medical College & Hospital Maharajganj, U.P, jayesh1102@gmail.com
[3]Principal and Professor, Department of Microbiology Rama Medical College Hospital and Research Centre, Mandhana Kanpur drsujatha1523@gmail.com

***Corresponding Author:** Dr. Deepak Sameer Bind
*Assistant Professor Department of Microbiology, KMC Medical College & Hospital Maharajganj, U.P. Email ID: deepaksameer0007@gmail.com,

Abstract

Background: Lower respiratory tract infections (LRTIs) are among the most common infectious disease and responsible for the cause of morbidity and mortality worldwide. Microscopic examination of sputum is the most commonly followed method in the laboratory for diagnosing lower respiratory tract infections (LRTI).

Aim and Objectives:- Quality Assessment of Sputum Gram Stain in Relation to Sputum Culture for lower Respiratory Tract Infections in a Tertiary Care Centre Kanpur.

Material & Methods: - This study was a Cross- sectional study carried out at the Department of Microbiology, Rama Medical College Hospital & Research College Kanpur, and Uttar Pradesh, India from January 2021 to December 2021. All 150 sputum samples were collected from patients in our hospital during the study period and processed in the central laboratory. Repeated sputum samples from the same patient and samples received from pediatric age group were excluded from this study. Samples were evaluated by gross appearance and subjectively categorized into mucus (mucus strands present) and watery (saliva present).

Results: - In this study total 150 sputum samples, 105 (70%) samples were accepted according to modified Bartlett's screening system and 45 (30%) samples were in the not acceptable category. Among acceptable category, 68(64.76%) samples were showed culture positivity. Among non-acceptable category, 11(24.44%) samples were showed culture positivity. The most common organism isolated was Klebsiella spp 29(42%), followed by Pseudomonas spp 23 (33.82%), Staphylococcus aureus 9(13.23%), Enterobacter spp 1(1.4%), Escherichia coli 1(1.4%), Citrobacter spp 1(1.4%) and Acinitobacter spp 1(1.4%). Streptococcus pyogenes 3(4.4%). Klebsiella spp. was the commonest isolated organism followed by Pseudomonas aeruginosa, Staphylococcus aureus and Streptococcus pyogenes.

Conclusion: - Sputum quality assessment is a useful tool for recommended to receive good quality of sputum and do initial sputum screening for diagnosing clinically relevant lower respiratory tract infections.

Keywords: - LRTI, Modified Bartlett's Criteria, Klebsiella, Pseudomonas spp etc.

Introduction:

Lower respiratory tract infections (LRTIs) are among the most common infectious disease and responsible for the cause of morbidity and mortality worldwide. Microscopic examination of sputum is the most commonly followed method in the laboratory for diagnosing lower respiratory tract infections (LRTI) [1]. One of the most important uses of the Gram stain is to evaluate the quality of expectorated sputum received for routine bacteriological culture. An acceptable sample yields less than 10 squamous epithelial cells per low power field [2]. The simplest and least expensive sample for the diagnosis of lower respiratory infections is expectorated sputum. The utility of this approach is the subject of controversy, as the sample is contaminated by or pharyngeal flora as it passes through the mouth. When the sample is collected carefully, it can provide useful information for initial therapy of community acquired pneumonia [3]. Sputum Gram's stain and culture are traditionally recommended procedures for routine diagnosis of LRTIs. But some physicians feel that definite diagnosis of LRTIs depends upon the properly performed sputum Gram's stain and microscopically examination according to the correct guidelines [1]. When significant oropharynges l contamination is evidenced in the cellular content of Gram stained sputum smears, the second sample representing lower respiratory tract must be collected [4]. The microbiology laboratory must use objective criteria by Gram stain screening for purulence before inoculation into culture media [5]. Unless microscopic examination is routinely included, half of all microbiological information rendered on sputum samples is meaningless and subject to misinterpretation of culture results. Hence culture must be guided by microscopic findings. When there is no correlation between culture and smear, the culture report may not indicate the etiology of lower respiratory tract infection. The present study was designed microscopically examination of Gram stained sputum smears and the sputum culture in patients with LRTIs.

Material and Methods:

This study was a Cross- sectional study carried out at the Department of Microbiology, Rama Medical College Hospital & Research College Kanpur, and Uttar Pradesh, India from January 2021 to December 2021. All 150 sputum samples were collected from patients in our hospital during the study period and processed in the central laboratory. Repeated sputum samples from the same patient and samples received from pediatric age group were excluded from this study. Samples were evaluated by gross appearance and subjectively categorized into mucus (mucus strands present) and watery (saliva present) [6].

The neutrophils (pus cells) and epithelial cells were observed under Microscope in 20-30 low power fields and average number of epithelial cells and pus cells calculated. Then the total score of epithelial cells and pus cells arrived at. The final score value of less than or equal to 0 is indicated a salivary contamination of sputum sample or lake of active inflammation (non- acceptable sputum sample). The final score of 1 and above was indicated an acceptable sputum sample (**Table:-1** According to the Modified Bartlett's Criteria)

Table:-1 Modified Bartlett's Criteria.

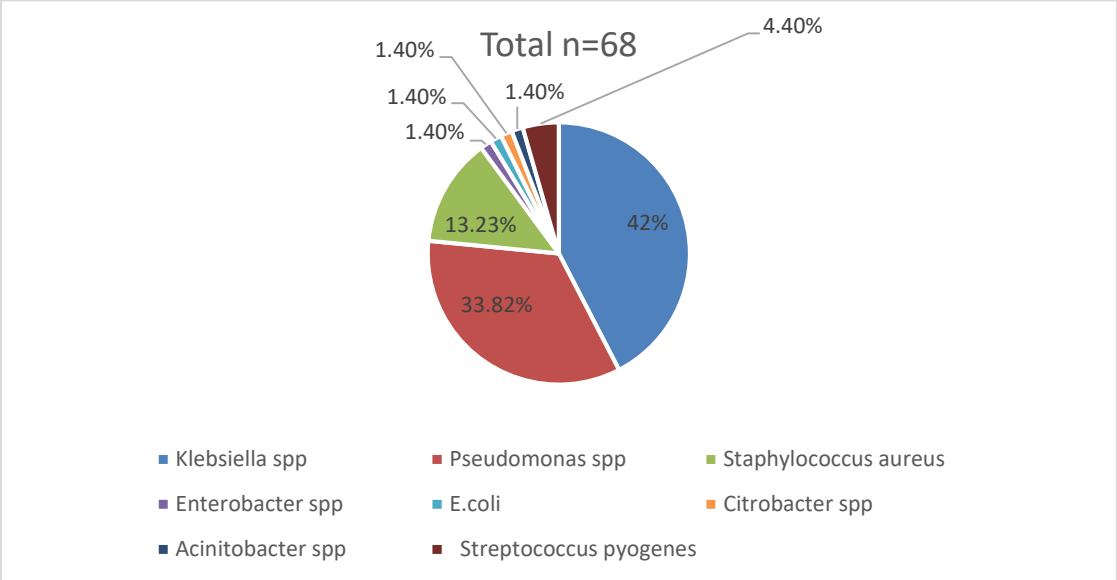
	CRITERIA	**SCORE**
	<10 Neutrophils / 10x Field	0

Neutrophils (Pus Cell) Count (Score A)	10-25 Neutrophils / 10x Field	+1
	>25 Neutrophils / 10x Field	+2
Macroscopic (Score B)	Mucoid, Mucopurulent, Purulent or blood Stained	+1
Squamous Epithelial Cell Count (Score C)	<10 Squamous Epithelial Cell / 10x Field	0
	10-25 Squamous Epithelial Cell / 10x Field	-1
	>25 Squamous Epithelial Cell / 10x Field	-2

The total score is Calculated using: - TOTAL SCORE = SCORE A + SCORE B + SCORE C
Each specimen was first mixed with an applicator swab and then inoculated on to blood agar, CLED agar and MacConkey agar plates and a smear was prepared for Gram staining. Each stained smear was examined microscopically under low power, oil immersion and the cellular components were evaluated. All the samples were processed regardless of the appearance of the stained smears. Organisms were identified by standard protocols and antibiotic susceptibility of recommended drugs was performed by Kirby-Buer disc diffusion method. Viridians group *streptococci*, *CoNS*, *Diphtheroids* and some Neisseria species were considered as normal respiratory flora [5].

Results:
In this study total 150 sputum samples, 105 (70%) samples were accepted according to modified Bartlett's screening system and 45 (30%) samples were in the not acceptable category. Among acceptable category, 68(64.76%) samples were showed culture positivity. Among non-acceptable category, 11(24.44%) samples were showed culture positivity. The most common organism isolated was *Klebsiella* spp 29(42%), followed by *Pseudomonas* spp 23 (33.82%), *Staphylococcus aureus* 9(13.23%), *Enterobacter* spp 1(1.4%), *Escherichia coli* 1(1.4%), *Citrobacter* spp 1(1.4%) and *Acinitobacter* spp 1(1.4%). *Streptococcus pyogenes* 3(4.4%). *Klebsiella* spp. was the commonest isolated organism followed by *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Streptococcus pyogenes.*

Fig 1:- Organism Isolates



Discussion:

Microscopic examination of sputum is the most commonly used method in laboratories for diagnosing lower respiratory tract infections (LRTIs) [1]. Clinicians require rapid, simple, inexpensive, and readily available tests that assist in prescribing appropriate medications for LRTIs. When a potential pathogen is isolated from a sputum sample, it is often difficult to determine whether it is the true etiological agent or merely represents pharyngeal contamination. The extent of pharyngeal contamination can be assessed by evaluating the relative number of squamous epithelial cells in the sample.

In 1974, Bartlett proposed that the quality of sputum samples should be assessed based on the relative concentration of polymorph nuclear neutrophils, squamous epithelial cells, and mucus in Gram-stained smears [7]. Although sputum culture has been criticized for its lack of sensitivity and specificity, comparability of positive sputum culture results with trans-tracheal aspiration has been demonstrated when the sputum samples are microscopically adequate [8]. Miraj J et al. initially reported that 21% of their specimens were unacceptable [9]. Archana et al. showed that 72 (55.4%) samples were acceptable based on Bartlett's screening system, while 58 (44.6%) samples were categorized as unacceptable. Among the acceptable samples, 64 (78.05%) showed culture positivity, whereas among the non-acceptable samples, 18 (21.95%) showed culture positivity. *Klebsiella pneumoniae* (31.71%) was the most commonly isolated organism, followed by *Pseudomonas aeruginosa* (14.63%) and *Staphylococcus aureus* (13.41%) [1]. Gorica Popova et al. reported that 132 (63.2%) samples showed culture positivity, of which *Streptococcus pneumoniae* 48 (36.4%) was the most commonly isolated organism, followed by *Moraxella catarrhalis* 22 (16.7%) and *Haemophilus influenzae* 21 (15.9%). Among the non-acceptable samples, 185 (14.5%) were culture positive, with the most commonly isolated organisms being *Escherichia coli*, *Staphylococcus aureus*, and others, accounting for 64 (34.6%), 54 (29.2%), and 28 (15.1%), respectively [10].

In the present study, a total of 150 sputum samples were analyzed. Of these, 105 (70%) samples were considered acceptable according to the modified Bartlett's screening system, while 45 (30%) were categorized as unacceptable. Among the acceptable samples, 68 (64.76%) showed culture positivity, whereas among the non-acceptable samples, 11 (24.44%) showed culture positivity.

The most commonly isolated organism was *Klebsiella* spp. 29 (42%), followed by *Pseudomonas* spp. 23 (33.82%), *Staphylococcus aureus* 9 (13.23%), *Enterobacter* spp. 1 (1.4%), *Escherichia coli* 1 (1.4%), *Citrobacter* spp. 1 (1.4%), *Acinetobacter* spp. 1 (1.4%), and *Streptococcus pyogenes* 3 (4.4%). Overall, *Klebsiella* spp. was the most commonly isolated organism, followed by *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus pyogenes*.

Conclusion:

Sputum quality assessment is a useful tool for recommended to receive good quality of sputum and do initial sputum screening for diagnosing clinically relevant lower respiratory tract infections.

References:

1. Archana, Keshav Kumar Bimal et al. A Study on Analysis of the Sputum Gram Staining and Culture in Patients with Lower Respiratory Tract Infections Attending a Tertiary Care Hospital International Journal of Health and Clinical Research, 2021; 4 (1):288-291.
2. Forbes BA, Sahm DF, Weissfeld AS. *Bailey and Scott's Diagnostic Microbiology*; 12th edn. Missouri: Mosby; 2007.
3. Koneman EW, Allen SD, Janda WM, Schrekenberger PC. Color Atlas and Text book of Diagnostic Microbiology; 6th edn. Philadelphia: *JB Lippencott*; 2006.
4. Howard H Mizrachi and Paul N. Valenstein 1, 2. 1987 Randomized Trial Interpreting Sputum Quality in a Clinical Laboratory. *J. Clin. Microbiol*. Vol 25. No.12.
5. Joseph R Lentino and David A. Lucks. Non value of sputum culture in the management of lower respiratory tract infections. *J. Clin. Microbiol*. 1987; Vol 25. No 5.

6. Bindu Nair, Jenny Stapp, Lynn Stapp, Linda Bugni, Jill Van Dalfsen, and Jane L. Burns. Utility of Gram staining for evaluation of the quality of cystic fibrosis sputum samples. *J. Clin. Microbiol*. 2002; Vol 40. No 8.
7. Howard H Mizrachi and Paul N. Valenstein 1, 2. Randomized Trial Interpreting Sputum Quality in a Clinical Laboratory. *J. Clin. Microbiol*. 1987; Vol 25. No.12.
8. N Arora, MK Daga, R Mahajan, S Krishna Prakash and N Gupta. 2001. Microbiol Pattern of acute infective exacerbation of chronic obstructive airway disease in a hospital based study. *Indian J chest dis allied sci* 2001; 43: 157-62.
9. Mariraj j, et al Sputum Gram Stain Assessment in Relation to Sputum Culture for Respiratory Tract Infections in a Tertiary Care Hospital. Journal of Clinical and Diagnostic Research. 2011; Vol-5(8): 1699-1700.
10. Gorica Popova et al Sputum Quality Assessment Regarding Sputum Culture for Diagnosing Lower Respiratory Tract Infections in Children Open Access Macedonian Journal of Medical Sciences. 2019; 30: 7(12):1926-1930.