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Original Research Article

Correlation of Cytology and Bacteriology of Effusions – A Diagnostic Clue

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Abstract

Body fluids like pleural, ascetic/ peritoneal, synovial and cerebrospinal fluids are usually sterile, but they can get infected by different microbes, thus leading to life threatening infections. This study was aimed to identify the prevalence of common bacterial isolates in effusions; determine the antimicrobial susceptibility pattern; correlate the microbiological and cytological imprints of effusions. This is a cross sectional and prospective study conducted between January and December 2018 in a tertiary care teaching hospital among the samples received for pathological and microbiological investigations. Of the 267 different body fluids samples processed, 127 (47.6%) were found to have inflammatory cells, 98 (36.7%) with non infectious cells and 42 (15.7%) with malignant cells in cytology. Of the 127 inflammatory samples, 42 samples supported with bacterial growth. The most common pathogens isolated were *Klebsiella pneumoniae* (n=12;28.6%) followed by non fermenting gram negative bacilli, *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and coagulase negative staphylococci. The gram negative isolates were relatively resistant to cephalosporins and quinolones; whereas gram positive isolates resistant to quinolones and aminoglycosides. Knowledge of the cytological investigations, bacteriological and antimicrobial resistant profile of sterile body fluids is necessary for clinicians for appropriate patient management.

Keywords: Body fluids; cellular microscopy; antibiotic resistance.

INTRODUCTION

Effusions are body fluids collecting from the cavity or joints which may be exudates or transudates based on whether it is an inflammatory process i.e. infective (septic) or non-inflammatory. Effusions are the most common causes for infective synovitis and arthritis, empyema, purulent pericarditis, ascites and peritonitis [1].

The cytological diagnosis of effusions has been discussed in medical literature for almost for hundred years. Cytological examinations of body fluids have increasingly gained acceptance in clinical medicine, to such an extent of a clinical clue that often considered as a definitive test. Availability of such early information helps the clinician to initiate early and more specific treatment and reduced lengths of stay of the patients in the hospital with less adverse effects [2]. There is a need for periodic analysis of the local geographical bacteriological profile and antibiotic susceptibility pattern of organisms isolated and the results to be correlated with cytology which needs to be communicated to the clinician at the earliest.

Hence it is important to optimize the culture techniques for isolation and identification of specific etiological agents. Microbial growth is scanty because of less number of pathogens as well as prior administration of empirical antibiotics before collecting samples [1]. Early detection and rapid identification of microorganisms are crucial for the appropriate management.

The increased risk of bacterial infection is further compounded by rising the trends of antibiotic resistance that are commonly implicated organisms all over the world [3]. This is particularly true in the case of members of Enterobactericeae group like *E. coli* and *Klebsiella* spp and non-fermenter group of bacteria such as *Pseudomonas* spp and *Acinetobacter* spp. To keep all these in mind this investigation was aimed to study the cytological profile of body fluids, identify the prevalence of bacterial isolates; analyze the

antimicrobial susceptibility pattern, and correlate among cytological and bacteriological imprints.

MATERIAL AND METHODS

Study Design

This is a cross-sectional prospective study; conducted in a tertiary care teaching hospital among samples; between January and December 2018. A descriptive research design was utilized in the current study which focused on collecting, processing the body fluids cytologically and bacteriologically of effusions in body cavities without trying to make interference. Institutional Ethical Committee clearance was obtained (CMCH&RC/IEC-101/05/12/2017).

Inclusion Criteria

All sterile body fluids received for cytological and bacteriological (aerobic culture and sensitivity) from different clinical departments.

Exclusion Criteria: All blood samples

Collection of Sample and Processing

The samples collected were pleural, ascitic, synovial, cerebrospinal fluids, pericardial and bile. All the eligible samples were processed cytologically and bacteriologically for determining inflammatory and malignant cells, culture and sensitivity respectively.

Cytology

Smears of body fluids were fixed and stained with Hematoxilin Eosin and Papanicolaou and examined microscopically for their cellular content and classification.

Bacteriology

All the samples were inoculated on Nutrient, Blood and MacConkey's agar plates and incubated aerobically at 37°C for 24 to 48hours. The colonies were further subjected to standard microbiological techniques (Gram staining, colony characteristics and biochemical properties). Antimicrobial sensitivity testing was carried out as per Clinical Laboratory standard institute (CLSI) guidelines. *Staphylococcus aureus* (ATCC 25923), *E. coli* (ATCC 25922) and *P. aeruginosa* (ATCC 27853) were used as quality control throughout the study. All the data were statistically analyzed.

RESULTS AND DISCUSSION

Out of 267 body fluid samples, 127 were considered as infective/ inflammatory, 98 (non inflammatory) and 42 as malignant based on the cellularity (Table 1). The pleural fluids showed maximum data in infective diagnosis and all the 127 infective/ inflammatory samples were further subjected for bacteriological analysis.

Table-1: Type of body fluid and clinical diagnosis

Specimen	No. of cases (n= 267)	Clinical diagnosis		
		Infective/ Inflammatory (n=127)	Unknown/ Non-inflammatory (n=98)	Suspicious of malignancy/ Malignancy (n=42)
Pleural fluid	114 (42.7)	56 (44.1)	41 (41.8)	17 (40.5)
Peritoneal fluid	81 (30.3)	39 (30.7)	25 (25.5)	17 (40.5)
Synovial fluid	49 (18.4)	27 (21.3)	16 (16.3)	6 (14.3)
CSF	16 (6.0)	3 (2.3)	11 (11.2)	2 (4.7)
Pericardial fluid	7 (2.6)	2 (2.6)	5 (5.1)	00

[Figure in parenthesis denoted percentages]

Cytological study of body fluid is a complete diagnostic modality. The information provided by body fluid analysis serves several functions. First, it assists the clinician in formulating and pointing out the etiology of effusion and list of differential diagnoses. Second, it allows one to follow the results of therapy and prognosis. Third it aids the Microbiologist in definitive diagnosis and thereby gives a clue regarding the organism (definite pathogen/ probable pathogen/ coloniser) [1, 3].

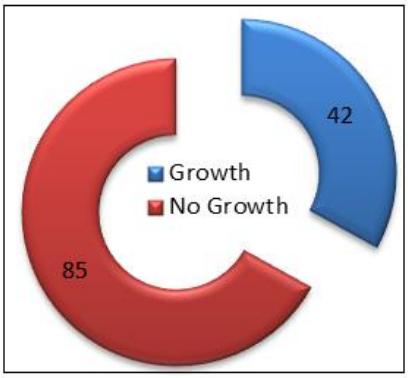

Fig-1: Correlation of infective cells with bacterial growth (n=127)

Among the 127 samples, the bacterial growth was found predominant in peritoneal fluid with 45.3% followed by pleural and synovial fluids. No growth was found among pericardial and CSF (Table-2).

Table-2: Type of specimen and positives

Specimen type (n=127)	Positive to bacterial growth (n=42)
Pleural (56; 44.1%)	18 (42.8%)
Pericardial (2; 1.6%)	-
Peritoneal (39; 30.7%)	19 (45.3%)
Synovial (27; 21.3%)	5 (11.9%)
CSF (3; 2.3%)	-

Pleural fluid was the most common sample submitted followed by peritoneal, synovial, CSF and finally pericardial fluid. Most of the study literature supports ascitic fluid being the most common body fluid submitted for microbiological analysis. The overall prevalence of bacteria in the present study is around 33.1% (n=42/ 127) which is comparative with the study done with all the samples received in Brazil (14.1%) [4]. This might be due to prior exposure to antibiotics, emergence of non infectious conditions like malignancy.

The isolation of *Klebsiella pneumoniae* dominated with 12 isolates followed non fermenting GNB, *E. coli* etc. The most common organism isolated from pleural, peritoneal and synovial fluids were *Klebsiella pneumonia*, *E. coli* and non fermenting GNB respectively (Table-3).

Table-3: Bacteriological profile of body fluids

Organism isolated	Pleural	Peritoneal	Synovial
Klebsiella pneumoniae (n=12)	6 (50)	5 (41.7)	1 (8.3)
Non-fermenting GNB (n=11)	5 (45.5)	4 (36.4)	2 (18.2)
E. coli (n=8)	2 (25)	5 (62.5)	1 (12.5)
Pseudomonas aeruginosa (n=4)	2 (50)	2 (50)	-
Staphylococcus aureus (n=3)	2 (66.7)	1 (33.3)	-
CONS (n=4)	1 (25)	2 (50)	1 (25)

[Figure in parenthesis denoted percentages]

The findings of the current study are in contrast to the previous studies [5, 6] where *E. coli* is the predominant organism but in our study *K. pneumoniae* predominated. The emergence of aerobic GNB and the presence of poly bacterial isolates in pleural fluid is alarming which requisites the use of aggressive antimicrobial therapy to curtail the infection.

The most common organism isolated from ascetic fluid was *E coli* and *K. pneumoniae* followed by non fermenting GNB which is correlated with literature [5-8]. The most common organism isolated from synovial fluid was NF GNB followed by *E. coli*, *Klebsiella* and CONS. This is in contrast to the observations of previous studies [9, 10] where gram positive organism like *Staphylococcus aureus* predominates followed by members of Enterobacteriaceae and non fermenters.

In the current study, gram positive isolates were sensitive to linezolid, vancomycin and teicoplanin and resistant to ciprofloxacin, ampicillin and aminoglycosides. Among gram negative isolates carbapenems, aminoglycosides and few cases to quinolones showed sentiive whereas nearly 70% of the isolates were relatively resistant to β-lactamases, β-lactamase-lactamases inhibitors and cephalosporins. This is in accordance with the study done in different parts of the world [9, 10].

The reported spectrum of microorganisms responsible for body fluid infection is varied, and is modified by introduction of antibiotics, patient specific factors such as surgical procedures, trauma or underlying conditions or by methodological factors namely the proper specimen collection, transport and culture. For these reasons, several studies have found discordant results in the spectrum of pathogens causing these infections [1].

Antimicrobial resistance is increasing to a larger extent which in turn increases the morbidity of the patient, length of hospital stay, risk of acquiring more infections, co-morbid conditions may aggravate etc.

CONCLUSION

This is the first kind of correlation study that provides the anticipated figure for the clinicians. Cytologic association of body fluids with microbiology will help in identifying the definite pathogen or coloniser. The infections of sterile fluid are usually associated with high morbidity which could be prevented by early initiation of appropriate therapy. Regular monitoring of prevalent pathogenic organisms and their sensitivity are essential as this will help in formulating the hospital antibiotic policy thereby preventing indiscriminate use of antibiotics and the restrain the development of super bug.

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