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COMPARATIVE STUDY OF CONVENTIONAL SMEARS AND CELL BLOCKS IN PLEURAL AND PERITONEAL FLUIDS CYTODIAGNOSIS

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Conflicts of Interest: Nil

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Abstract:

Introduction: Cytologic techniques have been universally the most important diagnostic tool in the recognition of malignant tumors in effusions. Cytodiagnosis by conventional smears (CS) has got lower sensitivity due to cell overcrowding, cell loss and different laboratory methods. Multiple sections can be obtained from the same material for special stains and immunohistochemistry in cell blocks (CB). Efficacy of cytodiagnosis is increased in CB method. This study was carried out with aim to study the morphological features of CS and CB technique and to assess the utility and sensitivity of CS and CB technique in cytodiagnosis of pleural and peritoneal fluids effusions.

Materials and Methods: Present study was a Hospital based Obervational study done for two years at a Tertiary Care Centre. All 189 fresh, received sample of effusion was divided into two equal parts and subjected for cytological evaluation of CS and CB technique.

Results: 42.3% cases belonged to age group 41-60 years, 66.1% were male. Cases of pleural effusions were 127 and of peritoneal effusions were 62. Out of 09 cases, diagnosed as suspicious of malignancy on CS, 07 cases were categorized as benign and 02 cases were labelled as malignant on CB. Diagnostic Accuracy is more for CB as compared to that of CS. **Conclusion:** Combined approach CB in conjunction with CS should be used in suspicious for malignancy cases.

Keywords: Cytodiagnosis, Conventional smear, Cell Block, Peritoneal fluid, Pleural fluid

Introduction:

"By the defects we know the talents, by the expectations we may know the rules, by studying pathology we construct a model of health."

Laurence Miller

In absence of disease, the parietal and the visceral layers of the mesothelium are separated by a thin layer of lubricating fluid that facilitates the movements of the two serous membranes against each other.

Under pathologic circumstances, two leaflets of the serous membrane may be separated from each other either because of the presence of air or fluids within the body cavity. The presence of fluid other than blood constitutes an effusion, which in the abdomen is called ascitis. Other types of effusions are chylous and pseudochylous effusions.

The purpose of cytological investigation is to determine the presence or absence of tumorcells; many other conditions can also be identified.

Cytologic techniques have been universally recognized as the most important diagnostic tool in the recognition of malignant tumors in effusions. The diagnosis of cancer in a pleural, pericardial, or peritoneal fluid is of capital importance for the patient and the attending physician or surgeon¹ and is well accepted. Positive diagnosis is often considered as a definitive diagnosis. ² It is important not only in diagnosis of malignant lesions, but also helps in staging and prognosis and also gives the information regarding infectious as well as non-infectious conditions of the membranes. ^{3,4}To differentiate between reactive mesothelial cells and malignant cells is the diagnostic problem in conventional smear (CS) method. ^{2,5,6}Cytodiagnosis by CS has got lower sensitivity due to cell overcrowding, cell loss and different laboratory methods. ^{2,5-7}

Preparation of cell blocks (CB) from residual sediment is often of great diagnostic value in the recognition of morphology and origin of the tumor and in application of special stains or other analytical procedures¹. It gives better architectural pattern, morphologic features and help to differentiate mesothelial cells from malignant cells. Multiple sections can be obtained from the same material for special stains and immunohistochemistry (IHC). 8,9,10 It provides less cellular dispersal, which permits easier microscopic observation.Possibility of storing slides for retrospective studies is possible for CB. Storage of the CS is a practical problem. 11

Thus the efficacy of cytodiagnosis is increased in CB method.

This study was carried out:

1. To study the morphological features of CS and CB technique.

2. To assess the utility and sensitivity of CS and CB technique in cytodiagnosis of pleural and peritoneal fluids effusions.

Materials and methods:

Present study was a Hospital based Obervational study done for two years at a Tertiary Care Centre. Patient's clinical history, examination findings and investigations were noted.

All pleural and peritoneal fluids, of admitted patients in the study hospital, of both the sexes and all age group (irrespective of clinical diagnosis), and received for cytology in the Department of Pathology was studied.

Fresh, received sample of effusion was divided into two equal parts and subjected for cytological evaluation of CS and CB technique.

For CS method, one part was centrifuged at 2500rpm for 15minutes. Supernatant fluid was discarded and minimum two smears were prepared. One smear was air dried for May-Grunwald Giemsa (MGG) staining and other was immediately fixed in 95% alcohol for Papanicolaou (PAP) staining.

For CB method, a clot was formed by Plasma-Thromboplastin, fixed in buffered formalin andprocessed in histokinette as part of routine paraffin section histopathology. Haematoxylin and eosin (H&E) staining was done. Special staining and IHC was done wherever required.

Slides were studied for morphology considering cellularity, predominant cell type, architecture and background and the diagnosis of benign (acute or chronic inflammatory) or malignant lesion or suspicious of malignancy was reported accordingly. Final diagnosis of negative or positive for malignancy was made considering history, clinical examination, radiological and cyto-histological findings.

Results:

The current study was carried out for a period of two years on 189cases of pleural and peritoneal fluids.

Cellular yield of CB was better due to the advantage of concentrating the cells. Cell morphology was distinct on both the techniques. But it was clearer on CB, distinguishing suspicious cases of CS to the definitive one. Similarly, architecture was well defined on both the methods. But was more typically seen on CB sections. Background was found to be obscured on CS while it was clear on CB.

80 (42.3%) of the cases belonged to age group 41-60 years, while only 2 (1.1%) cases were above 80 years of age. As regards gender, 125 (66.1%) cases were males and

64 (33.9%) cases were females, cases of pleural effusions were 127 and of peritoneal effusions were 62. [Figure 1]

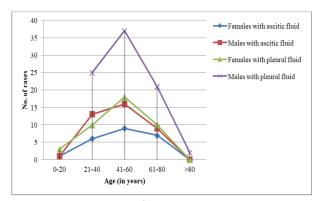


Figure 1: Distribution of cases according to age, sex and type of fluid

Total 160 cases were diagnosed as benign and 20 cases were found to be malignant on CS. Out of 09 cases, diagnosed as suspicious of malignancy on CS, 07 cases were categorized as benign and 02 cases were labelled as malignant on CB. [Table 1]

Table 1: Diagnostic comparison of cases based on CS and CB (n=189)

Conventional smear	Cell block		Total
	Benign	Malignant	<u> </u>
Benign	160	00	160
Suspicious	07	02	09
Malignant	00	20	20
Total	167	22	189

Chi-square = 173.9; df = 2; p = 0.0000001

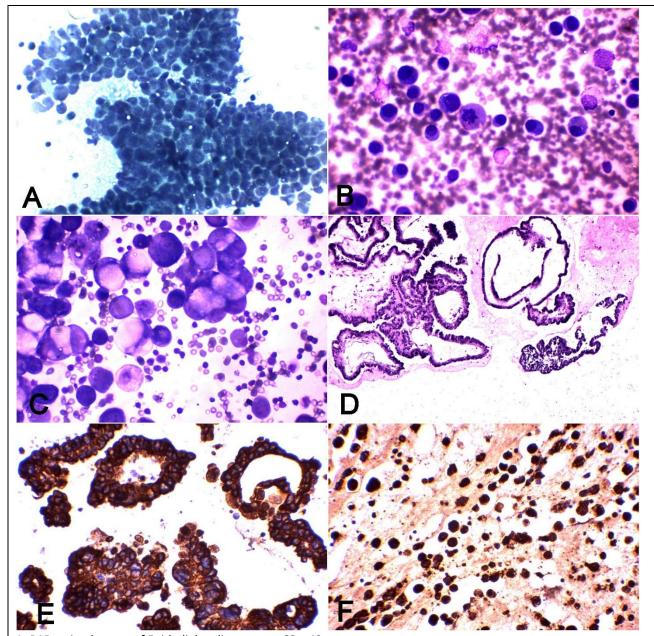
p-value < 0.001, so it is highly significant

Considering all the criteria for making final diagnosis, as mentioned earlier, out of total 189 cases, 166 (87.8%) cases were labelled as negative for malignancy and 23 (12.2%) cases as positive for malignancy.

The specificity and Positive predictive value of CB is 100% as compared to that of CS (98.75% and 90.90% respectively). Also, Diagnostic Accuracy is more for CB as compared to that of CS [Table 2]

Table 2: Comparison of CS and CB technique

Parameter	's	CS	СВ	
Sensitivity		86.95%	95.65%	
Specificity		98.75%	100%	
Positive	Predictive	90.90%	100%	
Value				
Negative	Predictive	98.14%	99.40%	
Value				
Diagnostic Accuracy		97.28%	99.47%	



- A: PAP stained smear of Epithelial malignancy on CS 40x
- B: MGG stained smear of suspicious of malignancy on CS 40x
- C: MGG stained smear of adenocarcinoma on CS 40x
- D: H&E stained section of adenocarcinoma on CB -10x
- E: Pancytokeratin positive adenocarcinoma showing membranous staining on CB (40x)
- F: Vimentin positive mesothelioma showing nuclear staining on CB (40x)

Figure 3: Microphotographs of stained smears on CS and CB

Discussion:

Cytological examination of serous effusions is of paramount importance in diagnostic, therapeutic and prognostic implications.

Te present study is comparable to different studies in literature, with respect to various parameters like most

common age group, number of males, number of females, number of pleural fluid and number of ascitic fluid studied. In 2015, Poorana P¹⁵, studied 120 cases, out of which 75 (62.5%) cases were male and found ascitic fluids out numbering with 48.33% cases.Study done by ThaparM et al⁵ on total 120 cases, also showed maximum cases of

pleural effusion (48.33%).In 2006, Khan N et al ¹⁶ studied 58 cases, with 55.17% of pleural fluid.

Present study included total 189 cases, out of which 160 (84.7%) cases were labeled as benign, 20 (10.6%) cases as malignant and remaining 09 (4.85%) cases were suspicious of malignancy, on CS. This is comparable with the studies done by BansodeS et al¹², Bhavandia VM et al⁶, Bodele AK et al⁴, Sujathan K et al⁷ and Takagi F et al¹⁷ in terms of diagnostic distribution done of the cases based on CS examination.

In the present study, 167 (88.4%) cases were labeled as benign, 22 (11.6%) cases as malignant and no case was found to be suspicious of malignancy, on CB. Thus, present study is comparable with the studies done by Bhavandia VM et al 6 , Shivkumarswami U et al 3 and Bodele AK et al 4 in terms of diagnostic distribution done of the cases based on CB

In present study, accuracy of CB was 99.47%. Increased accuracy was also noted by BansodeS et al 12 (97%), ThaperM et al 5 (85.72%) and Ceelen GH 18 (89%), when compared to CS.

Conclusion:

CS technique is easy and quick method for cytodiagnosis of body fluids, but background gets obscured and may affect the diagnosis.

CB technique is simple, reproducible and uses routine laboratory reagents and processing, but is time consuming. It offers advantage like concentrating all the cellular material and increasing cellular yield. It eliminates the suspicious for malignancy category giving more definitive diagnosis and hence, increase diagnostic yield. Multiple sections of the same material can be processed for IHC and also for special stains, if required. It increases sensitivity and accuracy of final diagnosis.

Hence, combined approach CB in conjunction with CS should be used in suspicious for malignancy cases.

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