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Original Research Article

Cytomorphological evaluation of ascitic and pleural fluid with special reference to cell block method

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ABSTRACT

Introduction: The cytological examination of serous effusions helps in diagnosis, staging and prognostication of patients with various malignancies. Combined analysis of Conventional smear and Cell block methods increases the diagnostic value of serous effusions.

Objectives: Of this study is to evaluate the cytomorphological features of ascitic and pleural fluids by the Conventional smear (CS) technique and the Cell block (CB) method, to compare them and also to assess the utility of their combined approach in the diagnosis of malignancy.

Materials and Methods: It is a cross sectional study, 110 specimens (64 ascitic and 46 pleural fluids) were subjected to evaluation by both CS and CB methods over a period of one year. Cellularity, architecture patterns, morphological features, nuclear & cytoplasmic details and also yield for malignancy were compared. Chi-square and p-value were calculated.

Results: Cell block method provided higher cellularity, better architectural patterns and additional yield for malignancy as compared to Conventional smear method. Chi square -19.0275, p- value is 0.000776. The p value is p<0.05, it is statistically significant.

Conclusion: The present study shows that it is advisable to routinely make CBs in every cases that were suspicious for malignancy by CS method.

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1. Introduction

Cytological examination of effusions is one of the commonly performed investigations. It has been universally recognised as the most important diagnostic tool in the recognition of malignant tumours in effusions. The cytodiagnosis by CS method have low sensitivity due to overcrowding of cells, cell loss and different laboratory processing methods. Therefore CB technique is usually recommended along with CS method for diagnosis of neoplastic effusions.¹

The CB technique is one of the oldest methods for the evaluation of effusions. The CB preparation concentrates the cells in a limited field without loss of cellular material and tissue architecture. The advantage of CB is the availability of multiple sections of the same material. From CB, we can go for further ancillary techniques to diagnose malignancy causing effusions. Cell Block is of particular value in effusion samples subjected to immunohistochemical staining (IHC) to elucidate the occult primary site of tumour. Besides primary diagnosis, IHC test on cell block is also useful for prognostication.² The aim of this study is to evaluate the cytomorphological features of ascitic and pleural fluids by the CS and the CB method, to compare them and also to assess the utility of their

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combined approach in the diagnosis of malignancy. Very few literature available about this topic which prompt us to do the study.

2. Materials and Methods

The present study was conducted on 110 patients, after taking written informed consent, who underwent pleural and peritoneal tapping for the cytological diagnosis of effusions by conventional and cell block methods. It is a hospital based cross sectional study. A total of 110 specimens (64 ascitic and 46 pleural fluids) were received in the Cytology section, Department of Pathology, Gauhati Medical College and Hospital. Both sexes and all age groups, from May 2020 to April 2021 were included in the study. Clotted fluid samples, time between collection and processing more than one hour were excluded from our study.

Each fresh fluid specimen was divided into two equal parts. One part was subjected to the CS cytology technique and the other part was subjected to CB technique. In CS technique, the sample was centrifuged at 2500 rpm for 15 minutes. Two thin smears were prepared from the sediment. One was stained with the May- Grunwald-Giemsa (MGG) stain and other was stained with Papanicolaou (PAP) stain. In CB method, the fluid sample after centrifugation, the supernatant was poured off and one drop of pooled plasma and two drops of thromboplastin (1:2 ratio) were added to the sediment and centrifuged again at 2500 rpm for 3 minutes for proper aggregation of the sediments. Then 10% formalin is added slowly by the side of the tube and allowed to be fixed for 8 hours. The clot is then wrapped in filter paper and processed in histokinette as part of routine paraffin embedded section preparation. Paraffin embedded cell block sections were stained by Hematoxylin and Eosin stain. Sections were mounted by DPX mountant and examined under microscope.³

The CS and CB sections were examined separately for cellularity, architectural patterns and morphology (cytoplasmic and nuclear details) to come at a cytological diagnosis for each case and the findings of each case were compared. The smears were categorised as Inadequate, Benign, Atypical, Suspicious and Malignant pleural and peritoneal effusion. Yield for malignancy were identified by both the CS and the CB method. Data was entered into Microsoft excel 2007 sheet and analysed. Chi- square test and p-value (<0.05) were calculated. Assessment and comparison between both the CS and CB methods were carried out.

3. Result and Observations

Out of total 110 specimens studied, 46(41.82%) were pleural fluid, 64(58.18%) were ascitic fluid.

3.1. Distributions of fluid

Table 1: Type of fluids

Ascitic	64(58.18%)
Pleural	46(41.82%)
Total	110(100%)

Table 2: Sex distribution

Total no of patient	Male	Female
110	44(40%)	66(60%)

3.2. Age distribution

Table 3: Age distributions

Age group (in years)	Patient no.
13-22	2 (2%)
23-32	11 (10%)
33-42	12 (11%)
43-52	28 (25%)
53-62	19 (17%)
63-72	13 (12%)
73-82	15(14%)
83-92	10(9%)

Most commonly affected age group is 43-52 year group and least affected group were 13-22 year group. In Ascitic fluids, the number of transudates are 34 and exudates are 30. In Pleural fluids, the number of transudates are 25 and exudates are 21.

Table 4: Distribution of fluids according to their nature

Fluids	Transudate	Exudate
Ascitic	34	30
Pleural	25	21

Amongst the total number of specimens studied, males predominantly had pleural effusion and females had ascitic effusion. Most common primary site in malignant ascitic effusion was found to be ovary, whereas the most common primary site in malignant pleural effusion was found to be lung.

By the CB method, an additional yield of fifteen (15) more malignant effusions i.e., 13.64% more cases were detected as malignant [or we can say discrepancy rate is 13.64% between CS and CB method]. These samples

Table 5: Comparison of various findings in CS and CB method

No	Diagnostic Category	CS Method	CB Method
1	Malignancy	14	29
2	Suspicious for malignancy	16	1
3	Benign	75	77
4	Atypical	3	2
5	Inadequate/Non Diagnostic	2	1
	Total	110	110

Discrepancy ratio- 13.64%
Chi square -19.0275, p- value is 0.000776. The p value is $p < 0.05$, it is statistically significant.

Table 6: Various type of malignancy in pleural and ascitic fluid in CB method

Malignancy type	Pleural fluid	Ascitic fluid
Adenocarcinoma	9	10
Squamous	2	5
Lymphoma	1	0
Leukemia	1	0
Urothelial cell malignancy	0	1

Table 7: Type of different benign causes of effusions in CB method

Category of non-neoplastic effusion	Total	Percentage
Reactive mesothelial cells	37	48.05
Granulomatous	22	28.58
Infective nature	18	23.37
Total	77	100

were reported as either suspicious for malignancy or benign samples.

Out of 29 malignant effusions (pleural and ascitic effusion), 16 were female patients and 13 were male patients. Male is to female ratio was 1: 1.23

4. Discussion

The present study “Cytomorphological evaluation of ascitic and pleural fluid with special reference to cell block method” was carried out in the Department of Pathology of Gauhati Medical College and Hospital, Guwahati for a period of one year from May 2020 to April 2021. A total of 110 effusion samples from Ascitic and Pleural cavities were collected.

The cytological study of serous effusions is one of the most definitive diagnostic tool for diagnosis of malignancy. It also helps in staging and prognosis of various malignancies.

Occurrence of malignant ascitic and pleural effusions is one of the most important features of various malignancies mainly ovarian and lung malignancies in this study.

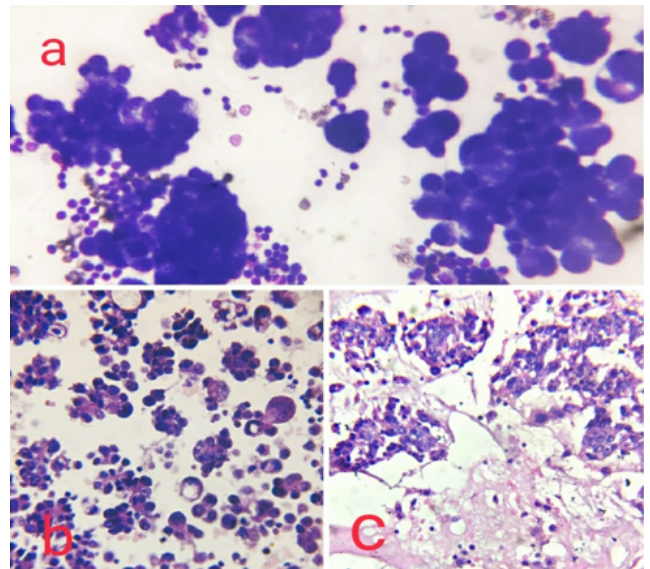


Fig. 1: a: Microphotograph of Metastatic adenocarcinoma from ovary. Conventional smear (10x40), MGG stain; b,c: Cell block preparation of metastatic adenocarcinoma from ovary (10x40), H&E stain.

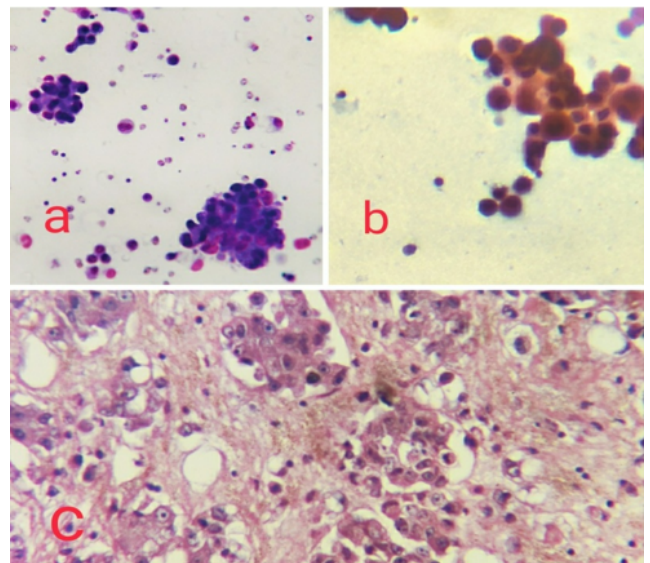


Fig. 2: a: Microphotograph of Metastatic adenocarcinoma from lung, Conventional smear (10x40), MGG stain; b: Metastatic adenocarcinoma from lung, Conventional smear (10x40), PAP stain; c: Cell block preparation of metastatic adenocarcinoma from lung (10x40), H&E stain.

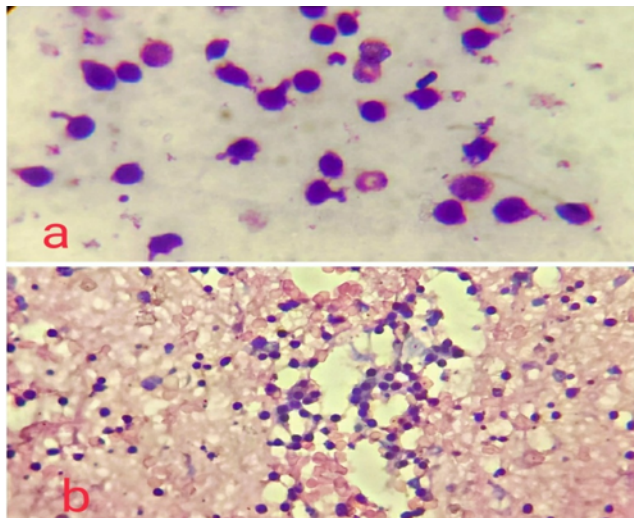


Fig. 3: **a:** Microphotograph of T-cell ALL of pleural fluid, Conventional smear preparation (10x40), MGG stain; **b:** Cell block preparation of T-cell all of pleural fluid (10x40), H&E stain

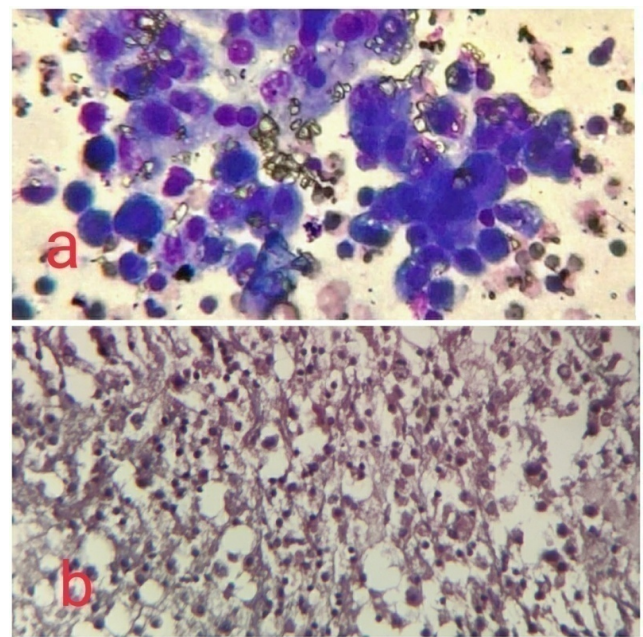


Fig. 5: **a:** Microphotograph of ascitic fluid infiltration by urothelial malignancy, Conventional smear preparation, (10x40), MGG stain; **b:** Ascitic fluid infiltration by urothelial malignancy, Cell block preparation (10x40), H&E stain.

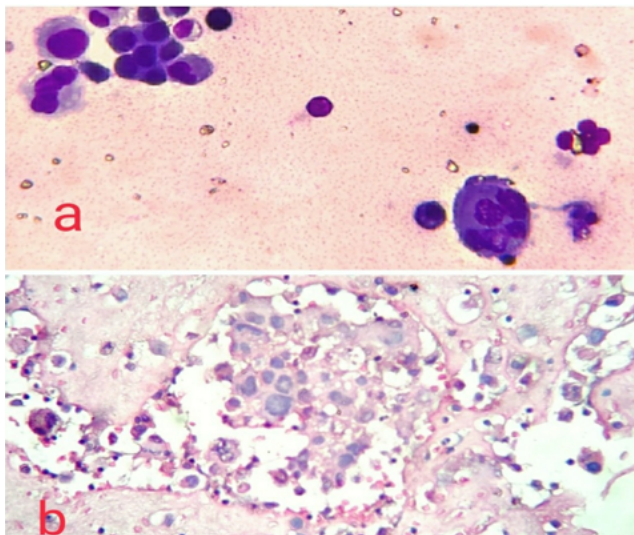


Fig. 4: **a:** Microphotograph of ascitic fluid infiltration from Pleomorphic giant cell adenocarcinoma of Pancreas, Conventional smear preparation (10x40), MGG stain; **b:** Ascitic fluid infiltration from Pleomorphic giant cell adenocarcinoma of pancreas, Cell block preparation (10x40), H&E stain

4.1. Age group distribution

Age of patients in our study ranged from 13-92 years, maximum samples were in the age group of 43-52 years. Our study was similar to Priyanka R et al,⁴ where the most commonly affected age group was 41-50 years.

4.2. Sample size distribution

The present study was done in Gauhati Medical College and Hospital and includes 110 effusion samples from ascitic and

Table 8: Showing comparison of our study with other studies carried out by different study groups on distribution of age

Priyanka R et al ⁴	41-50 years
Present study	43-52

pleural cavities. Our study was similar to Santwani P.M et al⁵ and Khan et al⁶ where the sample sizes were 150 and 75 respectively.

Table 9: Showing comparison of our study with other studies carried out by different study groups on distribution of sample size

Santwani P.M et al ⁵	150
Khan et al ⁶	75
Present study	110

4.3. Gender distribution

In our study, we found out that 40% cases were males and 60% were females indicating a higher prevalence of effusions in females. Our study was similar to Sumedha D et al² where prevalence of effusions was also more in females. Table 10

4.4. Diagnostic yield comparison

In our study, the increase in yield of malignancy was 13.64%, which was similar to the studies done by Flint et al,⁷ Santwani P.M et al⁵ and Sumedha D et al,² where the

Table 10: Showing comparison of our study with other studies carried out by different study groups on distribution of gender

	Male	Female
Sumedha D et al ²	23 (54%)	27 (46%)
Present study	44(40%)	66(60%)

Table 11: Comparison of increase in yield of malignancy by CB method of our study with other studies done by different study groups

	Flint et al ⁷	Bhanvadia Viral M	Sumedha D et al ²	Present study
Increase In Malignancy Yield	9%	10%	15.38%	13.64%

Table 12: Comparison of cytodiagnosis of effusions of our study with other studies (CS only)

S. No	Study	No of cases	Specimen	Malignant	Suspicious	Benign	Atypical	Inadequate
1	Dekker and Bupp, ⁸	173	Pleural, Ascitic and pericardial effusion	28	8	128	9	0
2	Thapar M et al.	190	Pleural, Ascitic and pericardial effusion	70	0	120	0	0
3	Present study	110	Pleural and ascitic fluid	14	16	75	3	2

Table 13: Comparison of cytodiagnosis of effusions of our study with other studies (CB only)

S. No.	Study	No of cases	Specimen	Malignant	Suspicious	Benign	Atypical	Inadequate
1	Sumedha D et al, ²	50	Pleural and Ascitic effusion	24	6	20	0	0
2	Kulkarni MB et al ³	38	Pleural, Ascitic and Pericardial effusion	23	0	15	0	0
3	Present study	110	Pleural and ascitic fluid	29	1	77	2	1

increase in yield of malignancies were 9%, 10% and 15.38% respectively. Table 11

In our study by CS method, 14 cases were found to be malignant, 16 were suspicious, 75 were benign, 3 were atypical and 2 were found to be inadequate. Our study was similar to Dekker and Bupp,⁸ where 28 cases were found to be malignant, 8 suspicious, 128 benign and 9 were atypical cases. Our study was also similar to Thapar M et al⁹ where 70 cases were found to be malignant and 120 benign cases. Table 12

In our study, 29 cases were found to be malignant, 1 was suspicious, 77 were benign, 2 were atypical and 1 case was found to be inadequate. Our study was similar to Sumedha D et al,² where 24 cases were found to be malignant, 6 were suspicious and 20 were benign. Our study was also similar to Kulkarni MB et al,³ where 23 cases were found to be malignant and 15 cases were found to be benign. Table 13

5. Conclusion

The present study shows that it is advisable to routinely make cell blocks in every atypical, suspicious and malignant cases found by conventional smear method. Cell block method is an easily available, better diagnostic tool for diagnosis of malignant effusions. Ancillary techniques can

also be used in cell block method for diagnosing specific type of malignancy and in evaluation of primary site.

6. Conflict of Interest

The authors declare no relevant conflicts of interest.


7. Source of Funding

None.

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