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Original Research Article

Evaluation of Cell Block Techniques in the Cytodiagnosis of Body Fluids in Meenakshi Medical College and Research Institute, Kanchipuram

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Abstract
Background: *Preparation of conventional smear is simpler and less time bound but it has many disadvantages causing difficulties in definitive diagnosis in fine needle aspiration materials and fluids. But cell block techniques are particularly useful in positive for malignancy and suspicious of malignancy cases. Once diagnosed as malignant in cell blocks, this can be taken as definitive diagnosis as a biopsy specimen. This helps in early diagnosis, treatment and better prognosis of patient. But cell block has minor disadvantages too e.g. needs more material and time.*
Aim: *To study cell block preparation of all the body fluids with conventional cytology smear.*
Material and methods: This prospective study conducted in Meenakshi Medical College and Research Institute, a tertiary care hospital which includes fresh body fluids and urine samples of 80 patients with relevant clinical details of both sexes and all ages. Very scanty fluid samples were rejected. Samples were processed on both cell block technique and conventional smear method. For cell block preparation agar gel method was used with 10% buffered formalin used for fixation.
Results: *80 body fluid specimens obtained and evaluated on cell block and conventional smear method, of which ascitic fluid was 16%, Pleural fluid was 13%, peritoneal fluid was 18% and other body fluids were 51%. Most of the patients were in age group of 41 to 60 years. Conventional smear preparation showed accuracy of 75% and cell block technique showed 92% accuracy.*
Conclusion: *Cell block along with conventional smear increases the accuracy of cytology fluid diagnosis. Cell block techniques provide higher cellularity, better tissue architecture, patterns of malignant cells and use of immunohistochemistry to identify primary site of malignancy over conventional smear method.*
Keywords: *Cell block, conventional smear, Agar-gel method of cell block preparation.*

Introduction

Cytological examination of bodily fluids has gained immerse acceptance in the clinical medicine to such extent that a positive diagnosis is considered definitive diagnosis and paramount important to diagnosis the type of malignancy and its primary site[1]. It helps in early diagnosis and treatment and ultimately increases patient survival rate[2, 3].

Conventional fluid sediment cytology smear is simpler procedure than that of cell block techniques. Conventional techniques has lower sensitivity due to overcrowding of cells, loss of cellularity during steps with loss of cellular architecture, increase number of inflammatory cells and obscuring factors and less number of diagnostic cells. These factors lead to considerable amount of difficulties to make a proper diagnosis on conventional smear[4, 5]. Though cell block technique is one of the oldest techniques but it has several modifications over the years to have following advantages over conventional method. It can concentrate very minimal amount of cellular material in a smaller area which can be examined very easily as all the cells are laying in a small focal area of microscope. As it uses the histopathological techniques for fixation and staining it gives better cell morphological with better nuclear and cytoplasmic preservation, intact cell membrane and also proper nucleolar and chromosomal details. It also preserves different architectural patterns e.g. papillae, acini, rosettes and individual cell morphology representing its primary site of tumor and fragments of this tissue can easily interpreted as a biopsy sample[4, 5]. As the materials are prepared as biopsy specimen, the sections from the paraffin blocks of the materials can be obtained for special stains, immunohistochemistry and identifying the primary site of malignancy. It helps in further retrospective studies as the paraffin blocks can be stored which is not possible in cases with conventional smears[2, 3].

Due to these advantages of cell block techniques we prepared and analysed both conventional and cell block techniques from the same body fluid specimens by Agar gel method to study about the morphology, cellular architecture and primary site of malignancy.

Materials and Methods

A prospective study was conducted after obtaining the approval of human ethical committee in Meenakshi Medical College and Research Institute, Kanchipuram. The study includes fresh body fluids from 80 patients with all relevant clinical details and detailed history. Consent was taken from each patient regarding the test.

Technique of aspiration

After explaining complete procedure written consent was taken from patient in each and every case before performing any aspiration.

Under all aseptic conditions ascitic fluid, peritoneal fluid, pleural fluid, Bronchoalveolar lavage fluid, vulval cystic fluid, endometrial fluid, fine needle aspiration fluids and urine samples were collected from the outdoor patients and admitted patients in the hospital.

Fluids were brought to laboratory for further processing fluids for preparing conventional sediment cytology smears and cell block.

Processing of fluids

(a) Conventional smear

2ml of fluid from the container transferred into two 5ml glass test tubes and centrifuged at 2000rpm for 5minutes.

Supernatant was then discarded and the sediment was taken onto clear, grease free glass slides.

With the help of a clean glass rod two thick smears were prepared and fixed with 95% methanol and stained with hematoxylin and eosin stain.

(b) Cell block preparation

Fluid from the container was transferred into 5ml of glass test tubes and centrifuged at 3000rpm for 5minutes. The supernatant was discarded.

Meanwhile 0.1mg of dry agar-agar powder was added to 5ml of distilled water, mixed well and heated up to 45-50^{9} c. at this temperature agar-agar and distiller water solution becomes semisolid and it remains same after lowering temperature.

The semisolid agar gel then added to the sediment of 5ml glass test tubes and kept in normal room temperature for 30 minutes in a test tube stand.
After 30 minutes agar gel solidifies and a cell button was prepared and transferred into a 10% buffered formalin solution for overnight fixation. Fixed cell button then labelled properly and processed in a tissue processor followed by paraffin embedding. Cell block tissues then stained with hematoxylin & eosin and special stains.

Criteria for diagnosis

Every conventional and cell block slide was analysed for cellularity, background, cytoplasmic and nuclear details, tissue architecture. On the basis of following criteria the conventional smear and cell block slides were diagnosed:

(1) Negative for malignancy:
- Scanty cellularity
- Reactive effusion
- Lymphocyte rich effusion
- Benign cystic fluid
- Inflammatory lesion

(2) Suspicious for malignancy

(3) Positive for malignancy

Statistical methods

Descriptive statistics e.g. mean, standard deviation were obtained for both conventional and cell block methods. Sensitivity, specificity, positive predictive value, negative predictive value and accuracy were determined for these two methods. The statistical analysis was carried out using SPSS version 25.

Results

Study materials consisted of ascetic fluid, pleural fluid, peritoneal fluid and other body fluids which include bronchoalveolar lavage fluid, endometrial fluid and FNA fluids from breast, thyroid, parotid etc and urine. Total 80 fluid samples were collected.

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

AGE (Years)	Type of fluid								Total
	Ascitic fluid		Pleural fluid		Peritoneal fluid		Other body fluids and urine		
	Male	Female	Male	Female	Male	Female	Male	Female	
Less than 20	0	1	0	0	0	0	0	0	1 (1.3%)
21 TO 40	1	2	1	0	1	3	3	13	24 (30%)
41 TO 60	6	1	1	4	0	8	7	8	35 (43.8%)
More than 60	1	1	5	0	2	1	8	2	20 (25%)
Total	8	5	7	4	3	12	18	23	80

Table no. 1 shows only 1 (1.3%) of the patients belonged to age group less than 20 years of age, 24 (30%) patients belonged to age group 21 to 40 years, 35 (43.8%) patients belonged to age group 41 to 60 years and 20 (25%) patients belonged to age group more than 60 years of age.

Table 2: Gender percentage

	Frequency	Percent (%)
Male	36	45.0
Female	44	55.0
Total	80	100.0

Among 80 samples (table 2), 36 (45%) samples were from male and 44 (55%) samples from females.

Among 80 samples (table 3, figure 1), 13 (16.3%) were ascitic fluid, 11 (13.8) were pleural fluid, 15 (18.8%) were peritoneal fluid, 3 (3.8%) were fluids from vulval cyst, 16 (20%) were fine needle aspiration fluids, 10 (12.5%) were bronchoalveolar lavage fluids, 6 (7.5%) were urine samples, 4 (5%) were ovarian cystic fluids and 1 (1.3%) fluid from cerebrospinal fluid and endometrium. In this study the fluids were divided in four categories as ascitic fluid, pleural fluid, peritoneal fluids and other body fluids with urine.

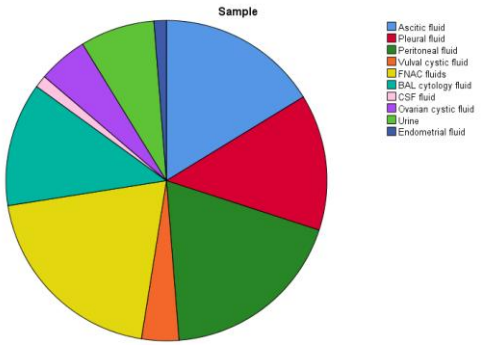


Figure 1: Pie chart of different fluid samples received

Table 3: Fluid samples and diagnosis on Conventional smear

	Conventional smear diagnosis							
	Scanty cellularity	Reactive effusion	Lymphocyte rich effusion	Benign cystic fluid	Inflammatory lesion	Suspicious for malignancy	Positive for malignancy	**Total**
Ascitic fluid	0	7	1	0	1	1	3	**13**
Pleural fluid	0	2	3	0	0	1	5	**11**
Peritoneal fluid	0	8	4	0	0	1	2	**15**
Vulval cystic fluid	0	2	1	0	0	0	0	**03**
FNAC fluids	0	3	1	9	2	1	0	**16**
BAL cytology fluid	0	3	1	0	2	2	2	**10**
CSF fluid	1	0	0	0	0	0	0	**01**
Ovarian cystic fluid	0	1	1	1	0	1	0	**04**
Urine	0	1	0	0	0	3	2	**06**
Endometrial fluid	0	0	0	0	0	1	0	**01**
Total	**01**	**27**	**12**	**10**	**05**	**11**	**14**	**80**

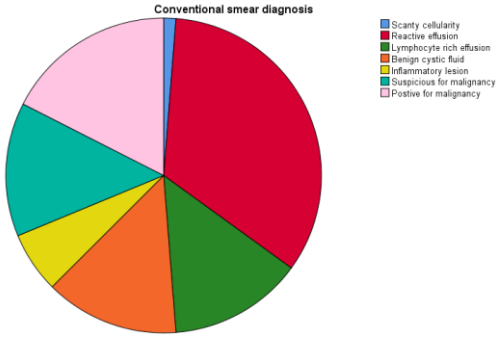


Figure 2: Pie chart of Conventional smear diagnosis

The diagnosis of the conventional smears (table no. 3, 4 and figure 2, 3) showed 56 (68.75%) cases were negative for malignancy which includes

Table 4: Diagnosis of Conventional smear

	Negative for malignancy	Suspicious for malignancy	Positive for malignancy
Ascitic fluid	9	1	3
Pleural fluid	5	1	5
Peritoneal fluid	11	1	2
Other body fluids and urine	30	8	4
Total (%)	55 (68.75%)	11 (13.75%)	14 (17.5%)

Diagnosis as scanty cellularity 1 (1.3%), reactive effusion 27 (33.8%), lymphocyte rich effusion 11 (13.8%), benign cystic fluid 11 (13.8%), inflammatory lesion 5 (6.3%). 11 (13.75%) cases were suspicious for malignancy and 14 (17.5%) cases were positive for malignancy.

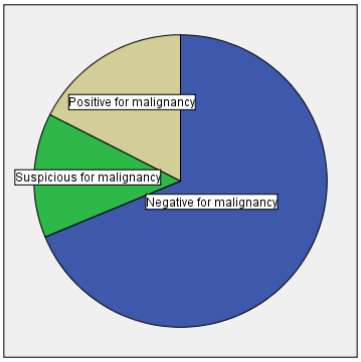


Figure 3: Pie Chart of Conventional smear diagnosis

The diagnosis of the cell block (table no. 5 and figure 4) showed 67 (83.75%) cases were negative for malignancy, 01 (1.25%) cases were

Table 5: Diagnosis of Cell block

	Negative for malignancy	Suspicious for malignancy	Positive for malignancy
Ascitic fluid	13	0	0
pleural fluid	7	0	4
Peritoneal fluid	12	1	1
Other body fluids and urine	35	0	7
Total (%)	67 (83.75%)	01 (1.25%)	12 (15%)

suspicious for malignancy and 12 (15%) cases were positive for malignancy.

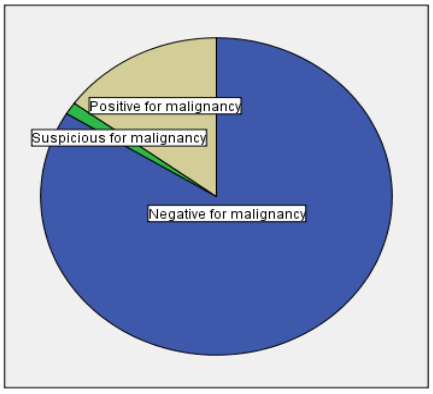


Figure 4: Pie Chart of Cell block diagnosis

Table 6: Descriptive statistics

		Diagnosis of Conventional smear	Diagnosis of Cell Block
N	Valid	80	80
	Missing	0	0
Mean		1.49	1.31
Std. Deviation		.779	.722

Mean of diagnosis of conventional smears and cell blocks are 1.49 and 1.31 respectively (table 6).
Standard deviation of conventional smears and cell blocks are .779 and .722 respectively (table 6).
The final diagnosis was achieved from follow-up biopsy specimens and tissue samples of all 80 cases. Both conventional smear and cell block diagnosis was compared with final diagnosis.

Table 7: Final biopsy diagnosis

	Frequency	Percent (%)
Positive	12	15.0
Negative	68	85.0
Total	80	100.0

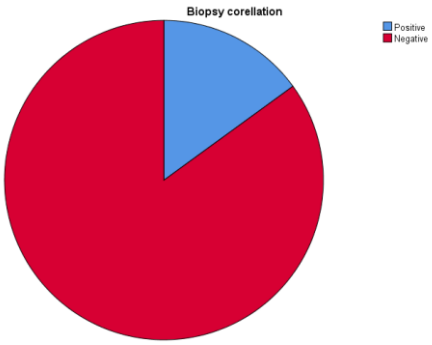


Figure 5: Final biopsy diagnosis

The final diagnosis from the biopsy specimens (table 7, figure 5) showed 12 (15%) cases were positive for malignancy and 68 (85%) cases were negative for malignancy.

Table 8: Contingency table showing the comparative diagnosis of conventional smear with final diagnosis

		Final diagnosis		
		Negative	Positive	Total
Conventional smear	Negative	54	01	55
	Positive	05	09	14
	Suspicious	09	02	11
	Total	68	12	80

Table 9: Contingency table showing the comparative diagnosis of cell block with final diagnosis

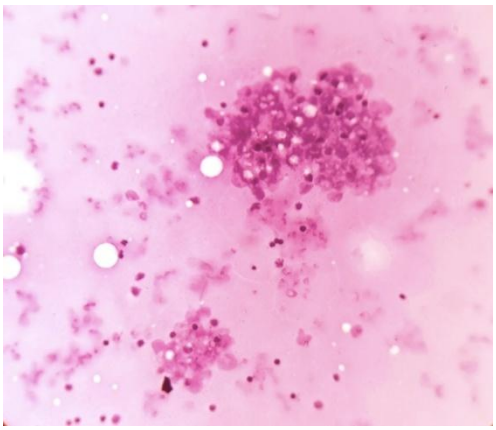
		Final diagnosis		
		Negative	Positive	Total
Cell block	Negative	67	00	67
	Positive	01	11	12
	Suspicious	00	01	01
	Total	68	12	80

To obtain sensitivity and specificity (table 10) of both the methods suspicious for malignancy cases were considered as negative (table 8, 9). The conventional smear showed a sensitivity of 75%, specificity of 92.64%, positive predictive value 64.28%, negative predictive value 95.45%, false positivity of 7.35% and false positivity of 25%.

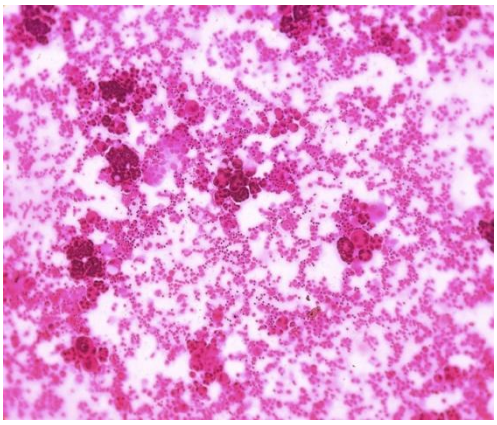
Table 10: Sensitivity, specificity and accuracy of conventional smears and cell block

Test	Conventional smear	Cell block
Sensitivity	75%	91.67%
Specificity	92.64%	98.53%
Positive predictive value	64.28%	91.67%
Negative predictive value	95.45%	98.53%
False positive	7.35%	1.47%
False negative	25%	8.3%

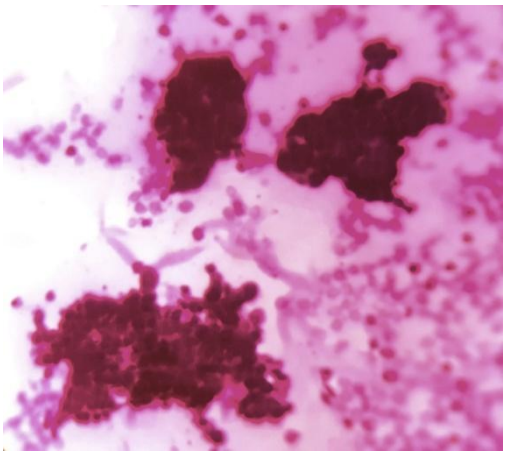
The cell block technique showed a sensitivity of 91.67%, specificity of 98.53%, positive predictive value of 91.67%, negative predictive value of 98.53%, false positivity of 1.47% and false negativity of 8.3%.



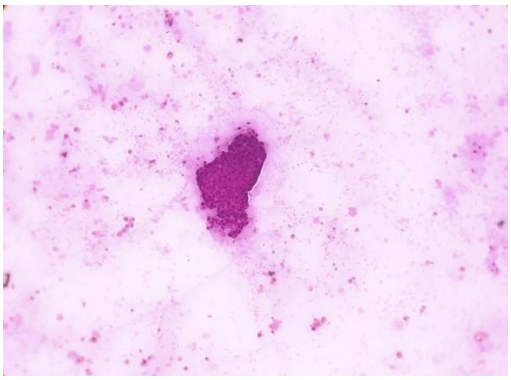
Photomicrograph showing Atypical mesothelial cells. Suspicious for malignancy on conventional smear of pleural fluid. (40X, H&E)



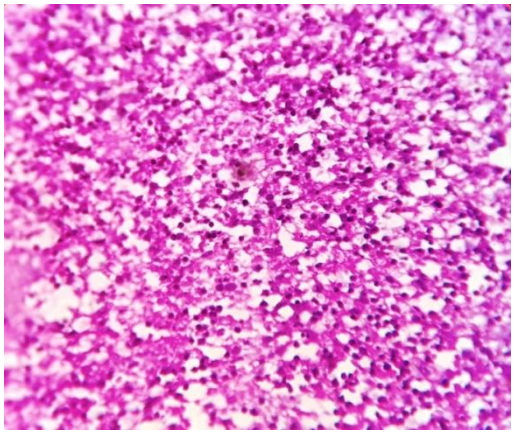
Photomicrograph showing Conventional smear of ascitic fluid - positive for malignancy (10X, H&E)



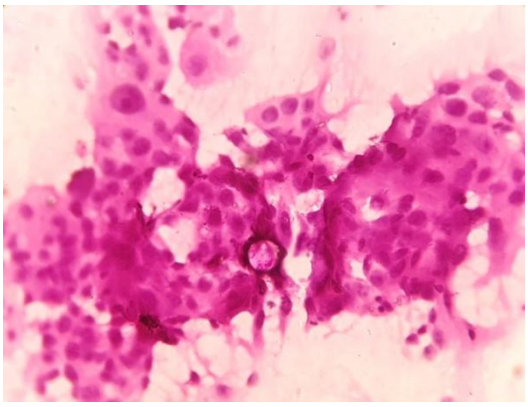
Photomicrograph showing Conventional smear of ovarian cystic fluid – positive for malignancy (40X, H&E)



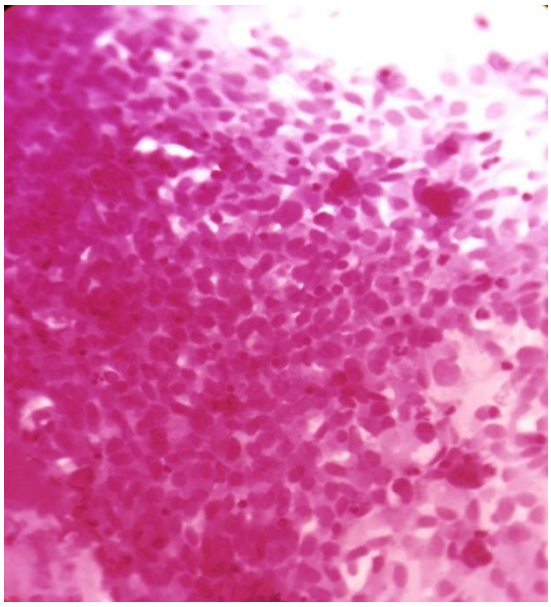
Photomicrograph showing Conventional smear of urine – suspicious for malignancy (10X, H&E)



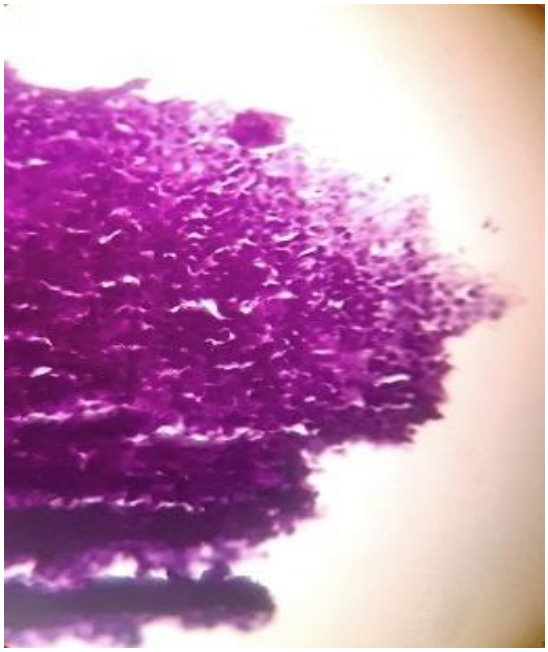
Photomicrograph showing Cell block of pleural fluid – lymphocyte rich effusion (40X, H&E)



Photomicrograph showing Cell block of pleural fluid – squamous cell carcinoma of lung (40X, H&E)



Photomicrograph showing Cell block of BAL fluid – positive for malignancy (40X, H&E)



Photomicrograph showing Cell block of urine – high grade urothelial carcinoma (40X, H&E)

Discussion

In this study diagnostic consideration was given on age, sex, type of fluids, type of cell block technique and also special stains whenever needed for a final diagnosis[1].

In this study cell block preparation was done by agar method. As this method uses only agar powder to produce agar gel, it is very cost effective. The advantages of this method are cost effective, easily reproducible, can yield more cell material which makes a better cell button for cell block. The disadvantages are background staining of agar gel in the cell block slides. 10% buffered formalin was used as fixative. Kulkarni MB et al[6] and Bansode et al[1] used plasma-thromboplastin method for cell block preparation. Bodele et al[4], Thaper et al[5], Shivakumarswami et al[7, 8] used sediment method with10% alcohol formalin as fixative, while Sujathan et el[9] used sediment method with ethanol-acetic acid and formalin as fixative. Both of these methods gave better cellularity when compared to conventional smears as formalin minimizes cell loss by forming protein cross links and gel formation. But disadvantages of these methods are this can be used only fluids with more sediments and showed less sensitivity for low volume of fluid or cell button with preservation of nuclear and cytoplasmic details from the cytological perspective is not satisfactory[10]. Nathan et al[11] prepared cell block by using improvised ethanol-alcohol as fixative.

In this study total 80 samples were collected and both conventional smear and cell block slides were made. Among these 13 were ascitic fluids, 11 were pleural fluid, 15 were peritoneal fluid, 3 were fluids from vulval cyst, 16 were fine needle aspiration fluids, 10 were bronchoalveolar lavage fluids, 6 were urine samples, 4 were ovarian cystic fluids and 1fluid from cerebrospinal fluid and endometrium. In Sujathan et al[9] out of 85 fluid samples 32 were pleural and 53 were peritoneal effusions. Where as in Shivkumarswami et al[7, 8] 60 pleural effusions were studied. In Sears et al study 61% were pleural and 39% samples were peritoneal effusions.

The age ranged from less than 20 years to more than 61 years with maximum number of cases seen in 41 years to 60 years of age for both males and females. In Shivkumarswami et al[7, 8] age ranged from 18 to 90 years. In case of gender, 36 (45%) cases were males and 44 (55%) cases were females.

The negative for malignancy category has scanty cellularity (01) were seen in cerebrospinal fluid showed few scattered neutrophils in both conventional smear (CS) and cell block (CB).

Reactive effusions were seen in 27 cases. Among these 15 cases showed reactive mesothelial cells

with few scattered inflammatory cells in background. They were diagnosed as decompensated liver disease and liver cirrhosis (7), congestive cardiac failure (03), and chronic kidney disease (5). Fluids from ovarian cyst (01), bronchoalveolar lavage (3), vulval cyst (2) showed few scattered epithelial cells in a chronic inflammatory cell background.

Lymphocyte rich effusions (12) seen in pleural (3) and BAL (1) are diagnosed as para-pneumonic effusions by clinical, biochemical and radiological investigations. In microbiological investigation these are mycobacterium tuberculosis culture positive which confirms the diagnosis of TB. Also fluids from peritoneum diagnosed as pancreatitis (4).

Benign cystic fluids (10) were diagnosed as colloid nodular goiter (07), cystic lesions of breast (2) and simple ovarian cyst (1) by clinic-radiological investigations.

Inflammatory lesions were exudative effusions and diagnosed as subacute bacterial peritonitis (1), abcess (2) and tuberculosis (1) by clinic-radiological, biochemical and radiological investigations.

In suspicious for malignancy category of CS, 11 cases were suspicious. Among these 8 cases was confirmed as malignancy by CB. They showed acinar, tubular and papillary architecture and diagnosed as adenocarcinoma on CB. 1 case was suspicious in CB too and application of IHC helped to rule out as a malignancy and diagnosed as peritonitis.

4 negative for malignancy cases on CS were diagnosed positive for malignancy and were diagnosed as serous adenocarcinoma of ovary (1), endometrial adenocarcinoma (1), ductal carcinoma of breast (1) and high grade urothelial carcinoma (1) on CB. As scanty cellularity and loss of cellular architecture on CS makes it difficult in diagnosis. On the other hand CB can concentrate more amount of cellular yield by making cell button and it also helps in maintain cellular and tissue architectural patterns.

In this study most of the cases were in negative for malignancy category with 68.75% on conventional smears and 83.75% on cell block. Similar findings were

seen in studies done by Takagi et al[12], Bodele et al[4], Khan et al[13], Shivkumarswami et al[7, 8]. Suspicious for Table 11:

Table no 11: Comparison of cytodiagnosis of serous effusions in present study with other studies

Sl. No	Study & Year	No. of cases	Negative for malignancy		Suspicious for malignancy		Positive for malignancy	
			CB	**CS**	**CB**	**CS**	**CB**	**CS**
1	Takagi et al (1954)[12]	184	145 (79%)	129 (70%)	6 (3%)	4 (2%)	33 (18%)	51 (28%)
2	Sujathan et al (2000)[9]	85	61 (72%)	63 (74%)	5 (6%)	1 (1%)	19 (22%)	21 (25%)
3	Bodele et al (2003)[4]	150	118 (79%)	111 (74%)	3 (2%)	0 0	29 (19%)	39 (26%)
4	Khan et al (2006)[13]	75	23 (31%)	14 (19%)	10 (13%)	7 (9%)	42 (56%)	54 (72%)
5	Shivkumar-swami et al[7, 8]	60	54 (90%)	50 (83%)	5 (8%)	0 0	1 (2%)	10 (17%)
6	Present study	80	67 (83.75%)	55 (68.75%)	1 (1.25%)	11 (13.75%)	12 (15%)	14 (17.5%)

malignancy were seen in 11 (13.75%) on conventional smear and 1 (1.25%) on cell block (table 11).

The final diagnosis was achieved from follow-up biopsy specimens and tissue samples of all 80 cases which showed 12 cases were positive for malignancy and 68 cases were negative for malignancy.

Positive for malignancy diagnosed on cell block is 12 (15%) and conventional smear is 14 (17.5%).

Similar findings were seen in studies of Takagi et al[12], Sujathan et al[9], Bodele et al[4], Khan et al[13], Shivkumarswami et al[7, 8].

Table no 12: Accuracy in different studies in Conventional smear and Cell block

Study	Conventional smear	Cell block
Thaper et al[5]	71.42%	85.72%
Zemansky et al[14]	-	90%
Ceelen[15]	71%	89%
Present study	75%	91.67%

In this study (table 12) accuracy of cell block was 91.67% then conventional smear 75%. Similar type of increased accuracy on cell block was seen in studies of Thaper et al[5] (85.72%), Zemansky et al[14] (90%) and Ceelen[15] (89%).

Conclusion

Cell block techniques are simple and reproducible and these use routine laboratory reagents and processing like normal histopathological specimens. But in cell block techniques more amount of sample is required for obtaining proper cell button. Cell block techniques offer concentrated all cellular materials and increase cellular yield. Though cell block techniques show preservation of cellular architectural pattern, but cellular morphology can be better appreciated on conventional smears. Use of cell blocks give more definitive diagnosis and show additional increase in diagnostic yield. In cell block technique multiple sections of the same material can be processed for immunohistochemistry that help to identify primary site of origin in malignant fluids. Combined approach of cell block and conventional smear can be used in suspicious for malignancy cases. Positive results, identification of primary site in malignant effusions help in patient's early management and overall good prognosis. Disadvantages of cell block technique are time consuming, need expert and well trained staffs to obtain good cell button and cause delay in issuing report. But having these disadvantages it has higher sensitivity and accuracy than conventional smears in diagnosis.

References

1. Evaluation of Cell Block Technique in cytodiagnosis of body fluids. Authors: Bansode Shubhada, Kumbalkar D, Nayak S, International Journal of Science and Research (IJSR), ISSN (online): 2319-7064, Index Copernicus Value (2013): 6.14 I Impact factor (2013): 4.438.
2. Koss LG et al: Effusions in the presence of cancer. In Koss' Diagnostic Cytology and its Histopathologic Bases, 5th edition. Edited by Koss LG, Melamed MR, Vol 2, Philadelphia: Lippincott, Williams & wilkins 2006, 950-951.
3. Dekker A, Bupp PA. Cytology of serous effusions. An investigation into the usefulness of cellblocks versus smears. Am J Clin Pathol 1978;70 (6):855-860.
4. Bodele AK, Parate SN, Wadadekar AA, Bobhate SK, Munshi MM. Diagnostic utility of cell block preparation in reporting of fluid cytology. Journal of Cytology 2003;20 (3):133-135.
5. Meenu Thapar, Rajiv K Mishra, Amit Sharma, Vikas Goyal, Vibhuti Goyal. Critical analysis of cell block versus smear examination in effusions Journal of Cytology.April 2009;26(2):60-64
6. Manisha B Kulkarni, Sangeeta B Desai, Dulhan Ajit, R.F .Chinoy. Utility of the thromboplastin-plasma cell block technique for fine needle aspiration and serous effusions.Diagnostic Cytopathology 2009 Feb;37(2):86-90.
7. Shivkumarswami U. Surekha U. Arakeril, Mahesh H. Karigowdar, B. R. Yelikar : The role of Cell block method in the diagnosis of malignant ascetic fluid effusion Journal of clinical and diagnostic research, 2012 ;September (suppl), vol-6(7):1280- 1283
8. Shivkumarswami U. surekha U. Arakeril, Mahesh H. Karigowdar, B. R. Yelikar: Diagnostic utility of the cell block method versus the conventional smear study in

pleural fluid cytology: j cytol 2012; 29 : 11-5.

9. Sujathan K, Pillai KR, Chandralekha B, Kannan S, Mathew A, Nair MK. Cytodiagnosis of serous effusions: A combined approach to morphological features in Papanicolaou and May-Grunwald Giemsa stained smears and modified cell block technique. Journal of Cytology 2000;17(2):89-95.
10. Shidham V, Atkinson B. Collection and processing of effusion fluids. In Cytopathologic diagnosis of serous fluids 1st edition Philadelphia: Saunders Elsevier, 2007; 207-33
11. Nathan NA, Narayan E. Smith MM, Horn MJ cell block cytology : improved predation and its efficacy in diagnostic cytology Am. J Clin pathol 2000; 114; 599- 606.
12. Takagi F. Studies on tumor cells in serious effusion, Am J Clin Pathol 1954; 24:663-675.
13. Khan N, Sherwani KR, Afroz N, Kapoor S. Usefulness of cellblocks versus smears in malignant effusion cases. Journal of Cytology 2006;23(3): 129-132.
14. Zemansky AP Jr. The examination of fluids for tumor cells: An analysis of m113 cases hecked against subsequent examination of tissue. Am J M Sci 1928;175:489-504.
15. Ceelen GH. The cytologic diagnosis of ascetic fluid. Acta cytol 1964; 8:175-183.