# Cytological Diagnosis of Body Fluids and Fine Needle Aspirates-Correlation between Cytospin Method and Cell Block Preparation, at a Tertiary Centre

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Abstract: Diagnostic cytology is a science based on three basic sampling techniques:- Collection of exfoliated cells; Collection of cells removed by brushing or similar abrasive technique; Aspiration biopsy or removal of cells from non - surface bearing tissues by means of needle with or without a syringe. <u>Method</u>: Patients were diagnosed by clinical history, lab tests, & cytological examination by cytocentrifuge and cell block technique as and when required. Fixative was usually not added because ethanol the usual cytological fixative causes heavy precipitation of proteins which interferes with concentration of cells. <u>Conclusion</u>: It is thus concluded from the present study that it is advisable to study by both cytospin and cell block method before discarding specimens that are negative for malignant cells by smear examination.

Keywords: Cytospin smear, Cell block preparation, Body fluids, Fine needle aspirates

## 1. Introduction

History of diagnostic cytopathology began in 1<sup>st</sup> era- 19<sup>th</sup> century. The second and most important era began in 1941 with the publication by Dr. George N. Papanicolaou (father of cytology) of an article in the American journal of obstetrics and Gynecology: The diagnostic value of vaginal smears in carcinoma of the uterus.

Diagnostic cytology is a science based on three basic sampling techniques: Collection of exfoliated cells; Collection of cells removed by brushing or similar abrasive technique; Aspiration biopsy or removal of cells from non surface bearing tissues by means of needle with or without a syringe.

Exfoliative cytology: Exfoliative cytology is based on spontaneous shedding of cells derived from the lining of an organ into a cavity, where they can be removed by nonabrasive means. Typical examples being vaginal smear prepared from cells removed from posterior fornix of the vagina and sputum. The principles of exfoliative cytology also apply to the body cavity effusions where collection of fluid is made by pleural, peritoneal or pericardial tapping. The principle lesions that can be identified are metastatic cancer and primary mesotheliomas. Apart from the routine cytomorphological study, tumour marker. Immunocytochemistry scanning electron microscopy etc. can be used to aid in diagnosis.<sup>1</sup>

<u>Aspiration cytology</u> Fine needle aspiration cytology (FNAC) was introduced with a view to reduce the number of excisional biopsies of lymph nodes. Needle aspiration of lymph nodes was used at the beginning of the 20<sup>th</sup> century to identify parasites in sleeping sickness, secondary syphilis, filariasis and bubonic plague. FNAC is a simple, minimally invasive and rapid diagnostic technique. It is safe, reliable,

cost effective outpatient department procedure which avoids morbidity and psychological trauma. Various studies supported the use of FNA as a primary method of diagnosis in reactive, infective and metastatic lymphadenopathy and also to know the primary site of malignancy. Definitive diagnosis can be made in most cases, thus sparing more expensive radiological investigations and/or surgical intervention.

## 2. Materials and Method

#### Materials needed for processing:

Disposable needled of 22-24 G with length of 2.5-3.8cm; Disposable plastic syringes of 10-20cc; Glass slides-clean and grease free; Methanol as fixative; Reagents for H&E stain, PAP stain; Sterile gloves, gauze piece and spirit swab.

#### Method for processing:

An effusion of serous cavity is diagnosed by correlating clinical & laboratory methods to differentiate Benign & Malignant effusions. The present study is from indoor patients of Government Hospital. Patients were diagnosed by clinical history, lab tests, & cytological examination by cytocentrifuge and cell block technique as and when required. Fixative was usually not added because ethanol the usual cytological fixative causes heavy precipitation of proteins which interferes with concentration of cells. Freshly tapped specimens are preferred for cytology, if facilities for immediate processing are available; if such facilities are not available, it can be preserved in refrigerator for 24-48 hours.

#### Cytological examination

Routine smear processing. Fluid received was stirred briskly to concentrate the suspended cells. The representative volume of fluid is centrifuged at 2500 rpm for 5 minutes. Smears were prepared from sediment on to glass slide by a thin cotton swab applicator stick & immediately fixed in 95% ethanol.

- a) Cell block preparation: Plasma Thromboplastin Cell Block technique was used.
- b) Cytocentrifugation: Cytospin method was used with routine smear.

In present study, slides were stained by H&E stain. Various Cytomorphological criteria were used for diagnosis & typing of neoplasm & for determining primary site of the tumors.

## 3. Discussion

In the present study, routine and cytological examination of ascitic and pleural fluid and various aspirates was carried out to differentiate between malignant and non malignant effusions. Total 80 cases of ascitic and pleural fluid and aspirates were studied by routine examination and cytological examination. The results obtained were compared with other authors, who have worked previously in this field on routine and cytological examination of fluids and the significant difference and similarities in results are discussed follows.Cordozo<sup>2</sup> (1966) used an anticoagulant in his study and added either heparin or EDTA to every fresh fluid specimen. Malvi and Anthony<sup>3</sup>(2000) studied the various methods of processing hemorrhagic serous fluid. In the present study, freshly tapped specimens without addition of anticoagulant were used for preparing smears and in cases where delay was inevitable, specimens were refrigerated. Spieler and Friedrich<sup>4</sup> (1985), Di Bonito et al<sup>5</sup> (1992), Sears and Hadju<sup>6</sup> (1987) had used alcohol fixed smears in their study while Cordozo<sup>2</sup> (1966) and Johnson<sup>7</sup> (1966) has used air dried smears in their study. In the present study ether alcohol (v/v) fixed smears were used. Konikov et  $al^8$  (1966) and Broghamer et al<sup>9</sup> (1984) had used Hand E stained cell

block technique.	block	technique.
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Table 1				
SAMPLE	Pleural fluid	Peritoneal fluid	Aspirates	Hydrocolpos
No of cases	56	07	19	01

Table 2				
Sr. No.	Gender of the patient	No. of cases of Pleural fluid	No. of cases of Ascitic fluid	
1.	Male	35	3	
2.	Female	21	4	
	Total	56	7	

Table 3				
Exfoliative Cytology	Inflammatory	Reactive Mesothelial Cells	Malignant Effusion	No Cellular Details
No. of cases	49	06	10	01

Table 4				
	EXFOLIATIVE			
AGE	Inflammatory	Reactive Mesothelial	No Cellular	
(in years)		Cells	Details	
1-10	-	-	-	
11-20	03	01	-	
21-30	03	-	-	
31-40	05	02	-	

41-50	17	-	01
51-60	13	03	-
>60	05	-	-

Та	ble 5
	Exfoliative
AGE ( in years )	Malignant
1-10	-
11-20	-
21-30	-
31-40	01
41-50	-
51-60	07
>60	02

Table 6					
	A	Aspiration	Cytology		
Age (in years)	Cystic	Infllam- matory	Inflammatory Cystic	Abscess	Hydroclpos
1-10	-	-	-	-	-
11-20	-	-	-	01	01
21-30	01	-	-	-	-
31-40	-	-	-	-	-
41-50	01	01	01	01	-
51-60	-	-	02	-	-
>60	-	-	01	-	-

Table 7

	Aspiration Cytology		
Age (in years)	Malignant		
1-10	-		
11-20	-		
21-30	-		
31-40	-		
41-50	-		
51-60	03		
>60	03		

## Comparative Study of Cytospin and Cell Block Technique

Only few studies have been reported regarding utility of cell block and cytospin methods in a case of diagnostic dilemma in a cytological smear.

Liu  $K^{10}(1998)$  compared the results of smears to those of cytospin and cell block preparations to determine which should be routinely obtained. Cytospin smear alone diagnostic in 100% of cases, and cell blocks alone diagnostic in 100% of cases. Cell blocks contributed no additional information beyond that obtained from cytospin smears.

In present study, out of 10 cases of malignancy, all 10 cases were detected by both cytospin smears and cell block methods.

## 4. Conclusion

Routine examination and cytological examination of effusions has an important diagnostic value to find out the cause of the effusion. There are many non invasive tests available which are relatively very simple, easy to perform, less time consuming and repeatable. Routine examination can help in differentiation of transudative and exudative

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fluids and cytological examination with smear, cytospin, cell block technique of fluid sediment and some special histochemical stains are helpful in the diagnosis and typing of malignancy in effusion. It is recommended that cytospin, cell blocks and routine smears should be used in evaluating all fluids submitted to the cytology laboratory. Knowledge of cytology of body fluids is of great help to the patients having inoperable malignancies where palliative treatment is at need. It also obviates the need of unnecessary surgery.

Cell blocks have helped in increasing the diagnostic yield can be used for the recognition of histological patterns of the disease, and for various ancillary studies that would not be possible with routine cytological examination and cytospin technique.

The other proved advantages of Cell Block method are:

- Preservation of tissue architecture and obtaining multiple sections for special stains.
- Recognition of Histological patterns of diseases that sometimes may not be identified in conventional smears.
- Less cellular dispersal, this permits easy microscopic observation.
- Storage of slides for retrospective studies.
- Use of blocks for advanced investigations like Immunostaining & Immunohistochemistry.

It is thus concluded from the present study that it is advisable to study by both cytospin and cell block method before discarding specimens that are negative for malignant cells by smear examination.

Following are the figures for comparison between cytospin smears and cell block sections:-

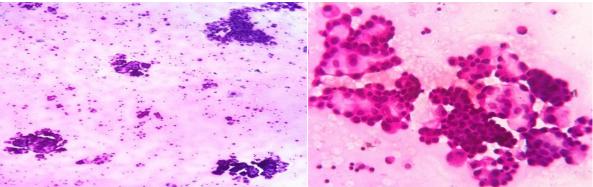


Figure 1: Reactive Mesothelial cells, 10X view and 40X view, H&E stain.

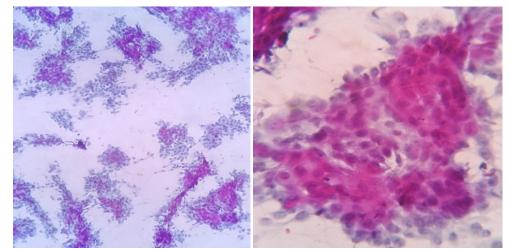


Figure 2: Metastatic Squamous Cell Carcinoma- 10X view, and 40X view, respectively, PAP stain

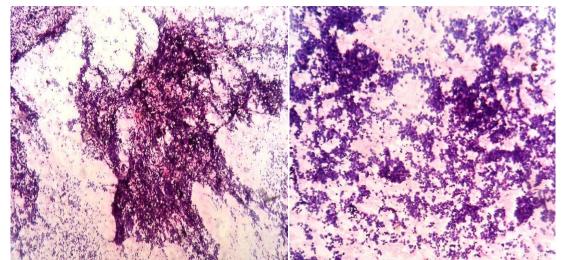


Figure 3: Metastatic Small Cell Carcinoma- 10X view, H&E Stain.

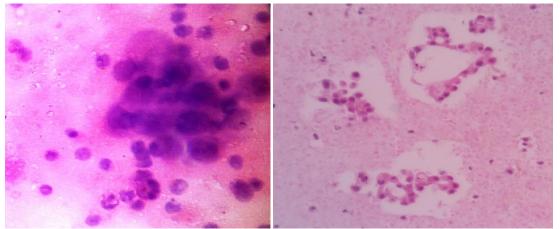


Figure 4: Metastatic Adenocarcinoma- 40X view, H&E Stain

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