

Role of Bronchial Washing and Bronchial Brushing in the Cytological Diagnosis of Pulmonary Lesions

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Abstract

Introduction: The Cytology of pulmonary lesions provides relevant information on diagnosis by non/minimally invasive procedures. It may be a helpful investigation in situations where biopsy procedure cannot be attempted due to high risk of haemorrhage and associated other co-morbid conditions. Various cytological methods currently available for studying the lung pathology are exfoliative (induced sputum), abrasive cytology, bronchial brushing [BB], bronchial washing and percutaneous / endobronchial fine-needle aspiration cytology. Bronchial washings and Brushings can be used to establish the presence of primary or metastatic neoplasms, benign tumors, and certain infections like Herpesvirus, Cytomegalovirus, Aspergillus, Coccidial infection, Candida, Actinomyces, Cryptococcus, Histoplasma, Pneumocystis carinii, and Strongyloides and in the diagnosis of Interstitial Lung diseases. Early diagnosis and treatment play an

important role in the mortality rates associated with lung carcinomas, therefore for early diagnosis bronchial brushings and washings play an important role.

Materials And Methods: All samples of bronchial washings, bronchial brushings and bronchial biopsy whenever available received over a period of 36 months retrospective and 12 months prospective were studied at a tertiary medical care hospital. Total 153 cases were received. All cytology specimens were collected by clinicians of the Department of Respiratory Medicine and were received in the Department of Pathology. The samples were processed and 5-6 smears were made directly from the brush by rolling the brush in a circular motion in a region 1.5 – 2cms in diameter. Smears were fixed in 95% ethyl alcohol for Papanicolaou stain, and air-dried for May-Grunwald Giemsa stain. Bronchial biopsies were received in 10% formalin, paraffin blocks were cut and slides were made. The slides were stained by hematoxylin eosin stain.

Results: The present study assessed total of 153 cases. The most common age group studied was 51-60 years. M: F ratio = 1.18:1. Total number of cases studied on bronchial brushings and bronchial washings were 153. In both bronchial brushings and bronchial washings, Nonspecific inflammation was the most common non-neoplastic entity, while Squamous cell carcinoma was the most common neoplastic lesion. Out of the 153 cases studied, histopathological correlation was possible in 87 cases. Out of 87 cases, diagnosis of 80 cases was confirmed and hence correlated on histopathology, 7 cases showed discrepancies in the cytology and histopathology findings. Discrepancy of 7 cases was because of misinterpretation by the cyto-pathologist due to chronic inflammation, superadded inflammation or squamous metaplasia. The Sensitivity, Specificity, PPV, NPV, Accuracy of bronchial brushings were 95.4%, 98.4%, 95.4%, 98.4%, 97.7% respectively. The Sensitivity, Specificity, PPV, NPV, Accuracy of bronchial washings were 90.9%, 98.4%, 95.2%, 96.9%, 96.5% respectively.

Conclusion: The present study concluded that bronchial cytology was found to be accurate, safe, and minimally invasive method for initial diagnosis of non-neoplastic and neoplastic pulmonary lesions. Bronchial brushings were more sensitive and specific than bronchial washings. Accurate diagnosis can be achieved by correlation of cytological features with clinical, radiological & histopathological findings. It is very helpful and can be carried out in patients with evidence of obstruction or risk of haemorrhage. The time of diagnosis can be reduced by 3-7 days as cytology report can be given in 2 days, thus alleviating patient's anxiety. In rural areas where histopathology facilities are not available, cytology can be used as an effective tool.

Keywords: Bronchial brushings, bronchial washings, bronchial biopsy, Histopathological correlation.

Introduction

Exfoliative cytology, to study the cells of respiratory tract was first used in 1845. It was in 1919, the ability to diagnose pulmonary diseases was first appreciated. However, it was not until 1950's, that pulmonary cytology came into.¹ In 1960's when flexible fiber-optic bronchoscope was developed, it improved the technique for to give a better yield to diagnose pulmonary diseases.²

Various cytological methods currently available for studying the lung pathology are exfoliative (induced sputum), abrasive cytology, bronchial brushing [BB], bronchial washing and percutaneous / endobronchial fine-needle aspiration cytology).

Bronchoscopy is the examination of airways under direct visualization. It is used primarily as a valuable tool in the diagnosis of lung diseases. It is extremely helpful in diagnosing endobronchial or centrally located lesions such as squamous cell carcinoma and carcinoid tumour.

Bronchial washing was collected at bronchoscopy under general or local anaesthesia in the operation theatre by clinicians in the Department of Respiratory medicine under strict aseptic conditions. Bronchial washing was obtained by instilling sterile isotonic saline into the bronchi via the bronchoscope and washings from different bronchopulmonary segments were re-aspirated and then smears were prepared from centrifuged deposits.

Bronchial brushings were following bronchial washings. The flexible fiber optic bronchoscope (FFB) was inserted through the mouth or nostril and slowly guided down into the trachea and bronchi. In lesion which were not easily visualized or biopsied the nylon brushes were

extended beyond the tip of the bronchoscope and material was obtained.³

Bronchial washings and Brushings can be used to establish the presence of primary or metastatic neoplasms, benign tumors, and certain infections like Herpesvirus, Cytomegalovirus, Aspergillus, Coccidial infection, Candida, Actinomyces, Cryptococcus, Histoplasma, Pneumocystis carinii, and Strongyloides and in the diagnosis of Interstitial Lung diseases.

Early diagnosis and treatment plays an important role in the mortality rates associated with lung carcinomas, therefore for early diagnosis bronchial brushings and washings play an important role. However, for the diagnosis of malignancy disagreement persists regarding the reliability of Bronchial brushings and washings in comparison with histopathology. It is practically difficult to perform all the tests in each patient because of the advantages and disadvantages of each technique and patient's co-operativity(4.)

The introduction of fiber-optic bronchoscopy revolutionized the cytological sampling of lung lesions, which resulted in better assessment of pulmonary lesions that were either not detected by radiological examination or unsuspected.

This study aims:

1. To assess the utility of bronchial washing and bronchial brushing in all pulmonary lesions.
2. To differentiate neoplastic (Benign and Malignant) and Non- Neoplastic Lesions (infective and non-infective).
3. To correlate with histopathology findings whenever available.

Material and Methods

This study was conducted at tertiary care hospital, over a period of 36 months, 24 months retrospective and 12 months prospective.

Total 153 cases were received.

The initial work up was through performing a flexible fiber optic bronchoscopy.

Bronchial washings (BW) were collected after brushing samples and were obtained by lavage with 20-40 ml of normal saline, and subsequent aspiration into a trap connected to the suction tubing.

Bronchial brushing was performed using re-usable brush with nylon bristles, which was cleaned carefully between procedures to enhance collection of satisfactory material for cytology. The samples were processed and 5-6 smears were made directly from the brush by rolling the brush in a circular motion in a region 1.5 – 2cms in diameter. Smears were fixed in 95% ethyl alcohol for Papanicolaou stain, and air-dried for May-Grunwald Giemsa stain⁴.

The cytological smears were then grouped into satisfactory/unsatisfactory and adequate/inadequate for interpretation. Smears with bronchial epithelial cells/alveolar macrophage were considered adequate. The satisfactory and adequate smears were further categorized as normal pattern, inflammatory, suspicious/atypical favoring neoplasm, and positive for malignant cells. Histopathological diagnosis by bronchial biopsy was considered as “Gold Standard”.

Bronchial biopsies were received in 10% formalin, paraffin blocks were cut and slides were made. The slides were stained by hematoxylin eosin stain.

Inadequate samples, Samples containing mucopurulent exudates, Cells showing degenerative changes on smears were excluded from the study.

Results

The present study was conducted from July 2014 to July 2017 in the department of pathology at tertiary care center. A total of 153 cases were studied, most common age group was 51-60 years consisting of total 34(22.22%) cases with Male:Female ratio being– 1.18:1.

Table 1: Distribution of cases on bronchial brushings (BB) and bronchial washings (BW)

Type of cases	Number of cases on BB	Number of cases on BW
Non-neoplastic	121	127
Neoplastic	32	26
Total	153	153

In the present study, on both Bronchial Brushings and bronchial washings, Nonspecific Inflammation was most common in 51-60 years age group and Malignancy was most common in 61-70 years' age group.

Table 2: Types of lesions on Bronchial Brushings and Washings (n=153)

Type of Lesion	Number of cases
Non-neoplastic	121
Normal Cytological appearance (No significant pathology)	0
Nonspecific Inflammation	71
Granulomatous Inflammation	40
Fungal etiology	10
Neoplastic	32
Squamous cell carcinoma	14
Adenocarcinoma	08
Suspicious for malignancy	09

As per given in the table no 2, most common etiology in both BB and BW samples was non- specific inflammation in non-neoplastic category and Squamous cell carcinoma under neoplastic category.

In BB, 45% cases i.e. 70 cases out of 121 were diagnosed as non-specific inflammation, whereas in BW, 78 cases out of 127 i.e. 56% accounted the same.

In neoplastic category, Squamous cell carcinoma was diagnosed in 14 cases out of total 30 BB samples, whereas in BW Squamous cell carcinoma was seen in 10 out of 26 cases.

Correlation Between Cytology And Histopathology

Out of these 153 cases, histopathological correlation was possible in 87 (56.86%) cases.

In remaining patients, correlation was not possible, because many patients opted for surgical procedure, few were transferred to other hospital and few were lost to follow up.

Out of 87 cases, in both Bronchial Brushings and Bronchial Washings diagnosis of 80 cases were confirmed on biopsy and 7 cases showed discrepancy.

The sensitivity, specificity, accuracy, positive and negative predictive value were calculated.

Cases which on cytology as well as on histology were malignant were considered as True positives (a). All the suspicious cases which later on histopathology were proven as malignant were also considered as True positives. True negatives (b) were those cases which were benign/non-neoplastic on both cytology as well as histology.

Case with a suspicion for malignancy which on histopathology turned out to be benign/ negative for malignancy was labelled as false positive (c). False negative were those cases diagnosed benign on cytology but later on histopathology turned out to be malignant.

Sensitivity was defined as the percentage of cases which were histologically proven were correctly diagnosed by the cytology.

Specificity was the percentage of cases in which

histologically proven benign / (negative) cases were correctly diagnosed negative on cytology.

Accuracy was defined as the fraction of patients whose conditions were correctly diagnosed by cytology.

Table 3: Comparison of cytological and biopsy results (n=87)

Type of lesion	No of cases on cytology	No of cases on Histopathology
Non-neoplastic	65	66
Nonspecific Inflammation	48	49
Granulomatous Inflammation	12	12
Fungal etiology	05	05
Neoplastic	22	21
Squamous cell carcinoma	11	14
Adenocarcinoma	06	07
Suspicious for malignancy	05	00

Correlation of cytology and histopathology on bronchial brushings and washings

Table 4: Agreement of bronchial brushing and washing findings with biopsy (n=80)

Cytology findings	Histopathology findings	Bronchial Brushings		Bronchial Washings	
		No of cases	Percentage	No of cases	Percentage
NSI	Inflammatory/empyema/lung Abscess	43	53.01%	44	51.89%
Granulomatous Inflammation	Tuberculosis	11	13.25%	12	15.18%
Fungal Filaments Seen	Aspergillosis	05	4.81%	04	5.32%
Squamous cell Carcinoma	Squamous cell carcinoma	11	13.25%	07	8.86%
Adenocarcinoma	Adenocarcinoma	06	8.43%	04	5.32%
Suspicious for Malignancy	Squamous cell carcinoma	02	2.40%	06	7.59%
Suspicious for Malignancy	Adenocarcinoma	02	4.81%	03	4.00%

Out of 87 cases where correlation was possible, diagnosis of 80 cases was confirmed on biopsy.

In Bronchial brushing, Amongst the 80 cases, 59 cases were non neoplastic and 21 cases were neoplastic, as described in table no 5.

Amongst the 80 cases in Bronchial washings, 60 cases were non-neoplastic and 20 cases were neoplastic, as mentioned in above table.

Table 4: Discrepancies on bronchial brushings (n=7)

Case	Cytology diagnosis	Histopathology diagnosis
1	Nonspecific inflammation	Squamous cell carcinoma
2	Nonspecific inflammation	Pulmonary Alveolar Proteinosis
3	Nonspecific inflammation	Tuberculosis
4	Nonspecific inflammation	Tuberculosis

5	Nonspecific inflammation	Tuberculosis
6	Nonspecific inflammation	Bronchogenic Cyst
7	Suspicious of malignancy	Squamous metaplasia

Out of the 87 cases studied, 6 cases of nonspecific inflammation turned out to be pulmonary alveolar proteinosis, Tuberculosis & Bronchogenic cyst and 1 case turned out to be squamous cell carcinoma.

One case where malignancy was suspected showed features of squamous metaplasia with dysplasia on histopathology. this case was considered as false positive.

Table 5: Discrepancies on bronchial Washings (n=7)

Case	Cytology diagnosis	Histopathology diagnosis
1	Nonspecific inflammation	Tuberculosis
2	Nonspecific inflammation	Tuberculosis
3	Nonspecific inflammation	Bronchogenic cyst
4	Nonspecific inflammation	Pulmonary Alveolar Proteinosis
5	Non Specific inflammation with Squamous metaplasia	Squamous cell carcinoma
6	Suspicious for malignancy	Squamous metaplasia
7	Suspicious for malignancy	Squamous metaplasia

Out of the 87 cases studied, 4 cases of nonspecific inflammation turned out to be 2 Tuberculosis, 1 case pulmonary alveolar proteinosis & 1 case bronchogenic cyst.

1 case of nonspecific inflammation showed occasional focus of squamous metaplasia which turned out as

Squamous cell carcinoma on histopathology. This case was considered as false negative.

2 cases where malignancy was suspected showed features of squamous metaplasia on histopathology. So these cases were considered as false positive.

Table 6: Comparison between Bronchial brushings & Biopsy

Bronchial brushing/biopsy	Biopsy positive	Biopsy negative	Total
Brushing positive	21	01	22
Brushing negative	01	64	65
Total	22	65	87

Table 7: Comparison between Bronchial washings & Biopsy

Bronchial washing/biopsy	Biopsy Positive	Biopsy Negative	Total
Washing positive	20	01	21
Washing negative	02	64	66
Total	22	65	87

Table 8: Tests results in tabulated form

	TP(a)	TN(b)	FP(c)	FN(d)	Total
Bronchial Brushings	21	64	01	01	87
Bronchial Washings	20	64	02	01	87

(TP = True positive, TN= True negative, FP= False Positive, FN= False negative)

Table 9: Comparison of indices of Bronchial Brushings & Bronchial Washings & overall cytology

Indices	Brushings	Washings
Sensitivity	95.4%	90.9%
Specificity	98.4%	98.4%
PPV	95.4%	95.2%
NPV	98.4%	96.9%
Accuracy	97.7%	96.5%

*PPV- positive predictive value, NPV- Negative predictive value

Figure 1

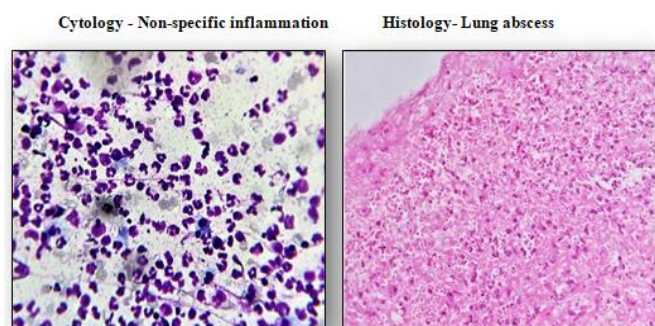


Figure 2

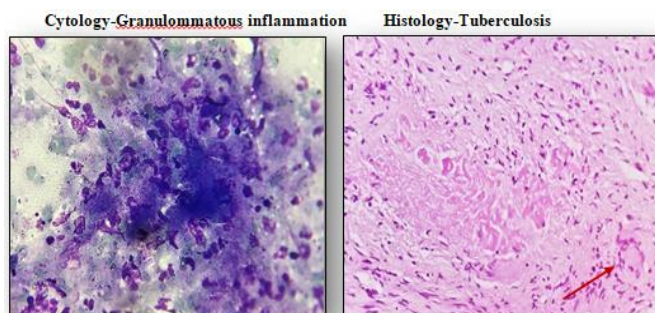


Figure 3

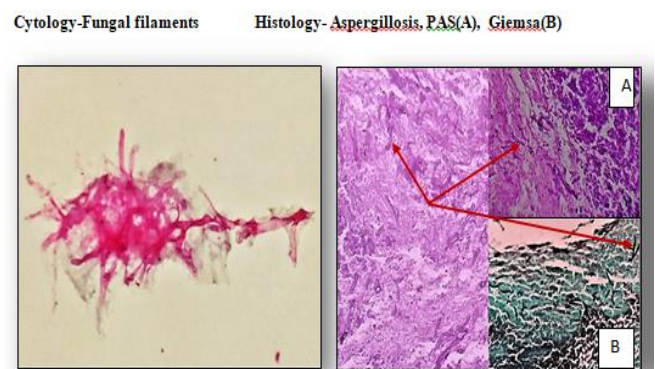


Figure 4

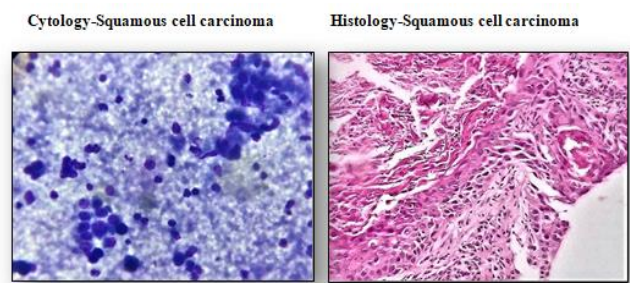


Figure 5

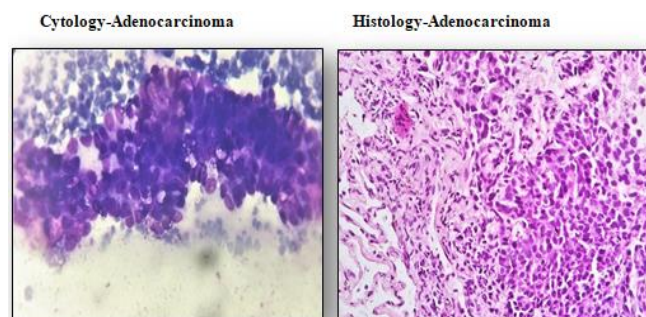


Figure 6: Cytology-Suspicious for malignancy
Histology- Squamous cell carcinoma

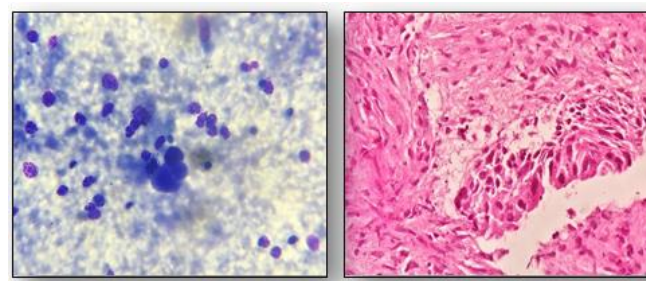


Figure 7

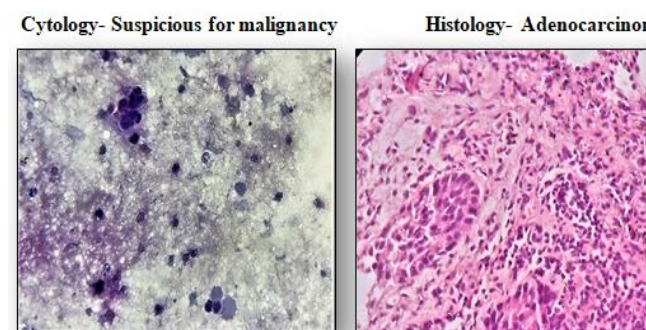


Figure 8: Cytology- Non-specific inflammation
Histology-Pulmonary Alveolar proteinosis

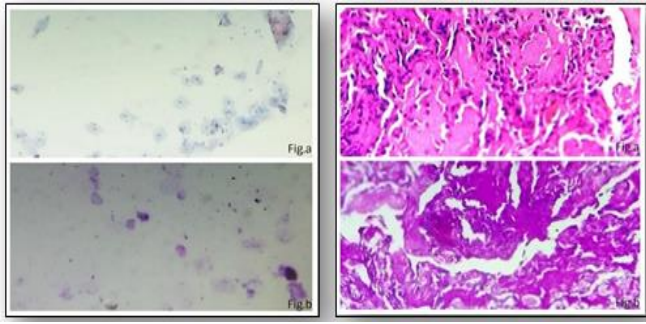


Figure 9: Cytology-Non-specific inflammation
Histology- Bronchogenic cyst

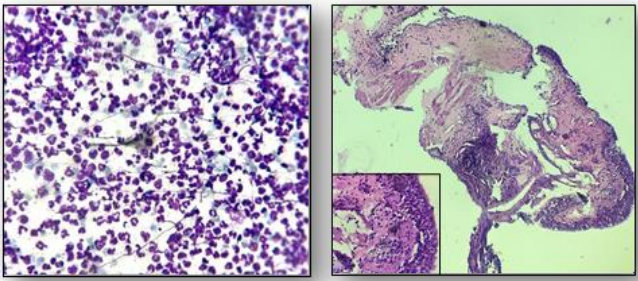


Figure 10: Cytology- Non-specific inflammation
Histology- Squamous metaplasia

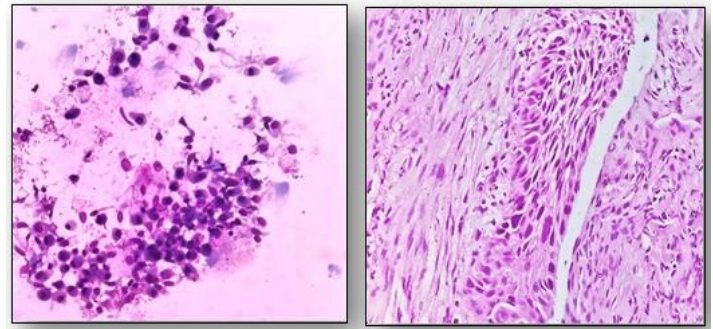
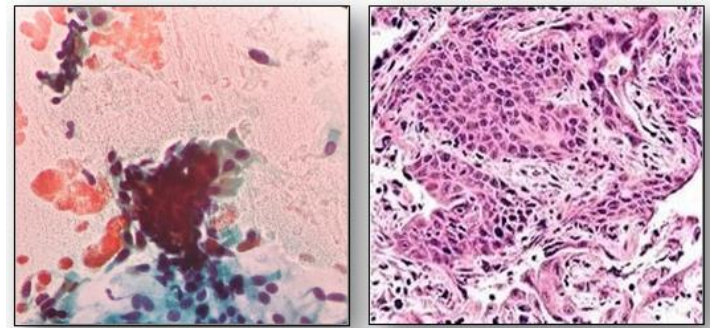


Figure 11: Cytology-Non-specific inflammation with
Squamous Metaplasia Histology- Squamous cell
carcinoma



Discussion

Table 10: Comparative incidence of pulmonary lesions of BB and BW

Studies	Neoplastic	Non Neoplastic
Choudhury et al (5) (n=35)	21 (60%)	14 (40%)
Rahul Giti et al (6) (n=100)	31	69
Raiza et al (7) (n=38)*	17(44.73%)	19(55.27%)
Bhavna Garg et al (8) (n=250)**	64(26%)	178(59%)
M. Pavani et al (9) (n=60)	20 (33.3%)	40 (66.67%)
Current study 40 (n=153)	32(20.91%)	121(79.09%)

In the present study, total of 153 cases of all pulmonary lesions were studied, 18 were normal, 103 were non neoplastic and 32 cases were malignant. The study done by Bhavna Garg et al reported 26% neoplastic cases which is comparable to our study. The percentage of non neoplastic cases in our study is greater than other studies because of high incidence of nonspecific inflammation followed by increasing incidence of tuberculosis.

In the present study, the age range was 15-85 years, which is similar to the study done by Choudhury et al (18-88) and M. Pavani et al (18-85).

The sex ratio in the present study was 1.83:1, which is comparable to the study conducted by Rahul Giti et al (1.5:1).

Table 11: Comparison of various pulmonary lesions on bronchial brushings

Type of Cases	Current study (n=153)	Shroff CP (10) (n=200)*	Manalo - Estrella P et al(11) (n=205)**
Normal cytological appearance (No significant pathology)	02(1.30%)	60(30%)	0
Nonspecific inflammation	70 (45.75%)	41(20.5%)	80(39.02%)
Granulomatous inflammation	39(25.49%)	10(5%)	-
Fungal etiology	10(6.53%)	0	-
Others	-	48(24%)	34(16.58%)
Malignancy	32(20.91%)	41 (20.5%)	

In the present study, the percentage of malignant cases was comparable with the study conducted by Shroff CP(10).

The no of cases showing nonspecific inflammation accounted for 70(45.75%) in the current study, was comparable with the study done by Manalo - Estrella P et al (11).

Table 12: Comparison of various pulmonary lesions on bronchial washings

Type of Cases	Current study (n=153)	Khandelwal R et al (12) (n=163)	Ahmed M et al (13) (n=73)
Non-neoplastic/benign lesions/Negative for malignancy	127(83%)	53(32.5%)	37(50.68%)
Suspicious for malignancy	12(7.84%)	58(35.6%)	09(12.32%)
Malignancy	14 (9.15%)	40 (24.5%)	25(34.24%)

In the current study the number of non-neoplastic/benign cases were 127(83%) which was greater than the study done by Ahmad M et al(13) 50.68% (37 cases) as the total number of cases studied by Ahmad M et al(13) (n=73) were less than the current study.

Table 13: Comparison of Indices with other studies in bronchial brushings

Study	Sensitivity	Specificity
Chaudhury BA et al(14)	47.6%	71.4%
DS Gaur et al(15)	7.3%	97.6%
Bhavna Garg et al(8) (n=250)**	88.2%	98.9%
Truong et al(16)	80%	-
Current study	95.4%	98.4%

The sensitivity of bronchial brushings in the current study was (95.4%) which was greater than the study done by DS Gaur et al (15) and the specificity was 98.4% which was similar to the study done by DS Gaur et al(15) (98.8%) respectively.

Table 14: Comparison of Indices with other studies in bronchial washings

Study	Sensitivity	Specificity
Chaudhury BA et al(14)	80.9%	85.7%
Rao S et al(17)	52.63%	80%
Bhavna Garg et al(8)	34.9%	98.6%
Sarma A et al(19)	80.5%	96.6%
Truong et al(16)	60%	-
Current study	90.9%	98.4%

The sensitivity of bronchial washings in the current study was 90.9% which was comparable to the study done by Chaudhury BA et al¹⁴ and Sarma A et al¹⁹. specificity in our study was 98.4% which was similar to the study done by Bhavna Garg et al.⁸

Conclusion

The present study concluded that bronchial cytology was found to be accurate, safe, and minimally invasive method for initial diagnosis of non-neoplastic and neoplastic pulmonary lesions. Bronchial brushings were more sensitive and specific than bronchial washings. Accurate diagnosis can be achieved by correlation of cytological features with clinical, radiological & histopathological findings. It is very helpful and can be carried out in patients with evidence of obstruction or risk of haemorrhage. The time of diagnosis can be reduced by 3-7 days as cytology report can be given in 2 days, thus alleviating patient's anxiety. In rural areas where histopathology facilities are not available, cytology can be used as an effective tool.

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