

## Research Article

# Correlation of bronchial biopsy with bronchoalveolar lavage in lung malignancies

Nazia Bhat<sup>1\*</sup>, Mir Junaid Nazeer<sup>2</sup>, Humaira Bashir<sup>1</sup>, Nusrat Bashir<sup>3</sup>, Summiya Farooq<sup>1</sup>,  
Kaneez Fatima<sup>5</sup>, Khalil M. Baba<sup>4</sup>

<sup>1</sup>Department of Pathology, Government Medical College, Srinagar, India

<sup>2</sup>Department of Radiodiagnosis, SKIMS, Srinagar, India

<sup>3</sup>Department of Haematology, SKIMS, Soura, Srinagar, India

<sup>4</sup>Department of Pathology, SKIMS, Soura, Srinagar, India

<sup>5</sup>Department of Radiotherapy, SKIMS, Soura, Srinagar, India

**Received:** 05 December 2015

**Accepted:** 06 January 2016

### \*Correspondence:

Dr. Nazia Bhat,

E-mail: [bhatnazia2@gmail.com](mailto:bhatnazia2@gmail.com)

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## ABSTRACT

**Background:** Lung cancer is currently the most frequently diagnosed major cancer in the world and the most common cause of cancer mortality worldwide. It comprises about 17% of the total new cancer cases in males and 23% of the total cancer deaths. The objectives of this study were to compare bronchial biopsy and bronchoalveolar lavage (BAL) cytology and in the diagnosis of carcinoma lung. Study design was comparative study.

**Methods:** The study was done in the Department of Pathology, Sher-i-Kashmir Institute of Medical Science (SKIMS), Srinagar, Kashmir. All patients clinically/radiologically suspected of lung malignancies who presented between April 2004 and May 2012 and underwent both bronchial biopsy and BAL were included in the study.

**Results:** Out of a total of 902 clinically suspected cases of lung cancer tumor was found in 760 cases (84.25%) by biopsy and in 301 cases (33.37%) by BAL. The total number of false positive cases was 31 and false negative cases were 490. Sensitivity of BAL was found to be 35.5% and specificity 78.16%.

**Conclusions:** In the present study yield of diagnosis was highest with the bronchoscopic biopsies and in maximum number of cases, specific histologic diagnosis was made by biopsies only. Though BAL was inferior to bronchial biopsy in diagnosing lung malignancies but it was effective for peripheral lung malignancies and when the patient was at risk of hemorrhage.

**Keywords:** Bronchoalveolar lavage, Lung carcinoma, Biopsy, Sensitivity

## INTRODUCTION

Lung cancer is currently the most frequently diagnosed major cancer in the world and the most common cause of cancer mortality worldwide.<sup>1</sup> It comprises about 17% of the total new cancer cases in males and 23% of the total cancer deaths.<sup>2</sup> The incidence is increasing dramatically in women and lung cancer has surpassed breast cancer as a leading cause of cancer death in women. Cancer of the lung most often occurs between age 40 and 70 years

with a peak incidence in the fifties and sixties. Only two percent of all cases appear before the age of 40.<sup>2</sup> The global incidence of lung cancer is increasing at the rate of 0.5 % per year and is the leading cause of death in most countries. Smoking is considered to be the cause of 85 % of deaths due to lung cancer.<sup>3,4</sup>

To combat the disease successfully, lung cancer should be diagnosed at earliest possible stage preferably before the lesion has reached the stage of a visible and palpable

tumour. For earliest diagnosis different modalities are available which include radiology, bronchoscopy bronchial biopsy, exfoliative cytology brushing washing and FNAC.<sup>5,6</sup> The sampling techniques performed at flexible bronchoscopy examination for histopathological diagnosis of lung cancer include endobronchial forceps biopsy (EBB) and transbronchial forceps biopsy (TBB) for more peripheral tumours.<sup>7</sup> Bronchial washing (BW), bronchoalveolar lavage (BAL) and brushing specimens can also be obtained for cytopathological examination.<sup>8-11</sup> Historically, 4 specimens have been shown to be adequate for optimal diagnostic yield in central lesions.<sup>12</sup> The diagnostic sensitivity of bronchial biopsy in diagnosing lung malignancies ranges from 65-83%.<sup>13,14</sup> Though histopathological diagnosis of bronchial tissue biopsy is considered the gold standard for diagnosis of lung tumors, it has certain drawbacks. It is an invasive procedure and more expertise is required. The yield is higher in patients with endoscopically-visible tumours than in those with tumours not visible endoscopically.<sup>15</sup> Diagnostic ratio of bronchoscopies is lower for peripheral lesions.

It is, however, in the context of more peripheral lesions that cannot be visualised that cytology has historically played a more crucial role, with bronchial brushing and washing/BAL samples being obtained from the relevant lobar segments. The sensitivity of BAL varies between 14-76% in various studies reported<sup>8,16</sup>. BAL can provide diagnostic information in cases of primary and metastatic lung cancer<sup>17,18</sup>. It is a valuable diagnostic tool in detecting peripheral primary pulmonary malignant neoplasm.<sup>19</sup>

BAL is an easily performed and well tolerated procedure that is used in routine assessment of patients for carcinoma lung. It also helps tamponade any bleeding that may have occurred as a result of biopsy.<sup>20</sup>

The present study was therefore undertaken to ascertain the role and diagnostic utility of bronchoalveolar lavage and bronchial biopsy in diagnosing and subsequent management of patients with bronchogenic carcinoma.

## METHODS

The study was carried out in the department of pathology at Sheri-i-Kashmir Institute of Medical Sciences, Kashmir India. The study included 902 cases of clinically/ radiologically suspected lung cancer received over a period of 8 years from May 2004 to April 2012, with 6 years retrospective and 2 years prospective.

In the retrospective study all the cytology slides of BAL and histopathologic slides of bronchial biopsy of patients diagnosed as bronchial cancer were taken from records of the department and slides were reviewed in detail.

In the prospective study all histopathologic slides of bronchial biopsies and cytology slides of BAL received

in the department were followed. Bronchial biopsies and BAL done for diseases other than cancer were excluded from the study. The bronchial biopsies were examined, size and number of bits counted. The tissues were processed as per standard procedure. 4-5m thick sections were cut on microtome and stained by hematoxylin and eosin stain. The stained slides were studied in detail microscopically; special stains like Periodic acid Schiff's were used where needed. The diagnosis and typing of tumour was made according to World Health Organization's classification.<sup>1</sup>

Bronchoalveolar Lavage fluid was received within half an hour along with relevant clinical details. It was immediately centrifuged for 5 minutes at 1500 revolutions per minute. Three slides were prepared from the sediment. Two of the slides were fixed in absolute alcohol and one air dried. Two of the alcohol fixed slides were stained with Papanicolaou stain and third with May – Grunwald Giemsa stain. These slides were studied in detail microscopically and diagnosis confirmed. The smears were grouped into malignant, suspicious/atypical and negative for malignant cells. For both cytology and histology only specimen with unequivocal malignant features, were considered to be positive. The malignant cells were further typed as squamous cell carcinoma and small cell carcinoma, adenocarcinoma and undifferentiated. The sensitivity, specificity, accuracy, positive and negative predictive values were calculated.

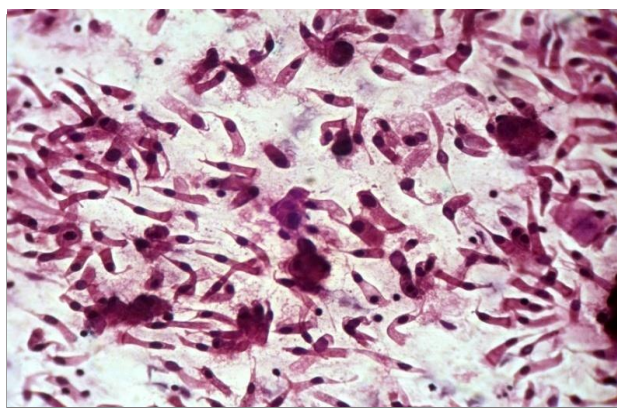
## RESULTS

This eight year study was conducted on 902 bronchial biopsies [including transbronchial biopsies (TBB)] and BAL. The study included only those cases where both BAL and bronchial biopsy was done simultaneously.

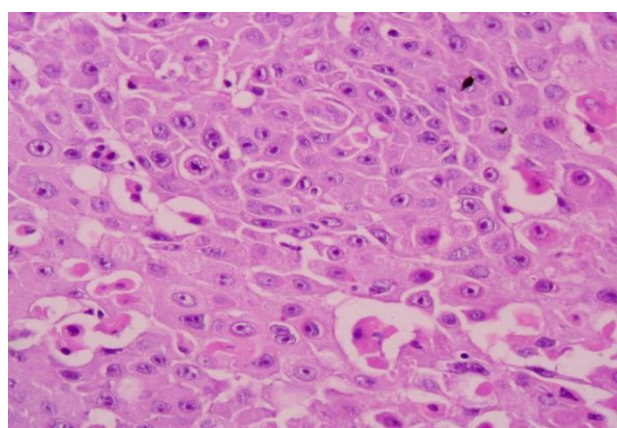
Male to female ratio was 6.3:1. The overall mean age of patients of primary lung cancer was 58.62 years with maximum number of cases seen between 61-70 years. The mean age for men was 59.16 years and for females, it was 55.44 years (Table 1).

**Table 1: Age and sex wise distribution of cases.**

Age group	Males	Females	Total	Percentage
10-20	4	0	4	0.44%
21-30	9	3	12	1.33%
31-40	38	14	52	5.76%
41-50	136	36	172	19.06%
51-60	224	36	260	28.82%
61-70	266	23	289	32.03%
71-80	84	10	94	10.42%
>81	17	2	19	2.10%
Total	778	124	902	100%



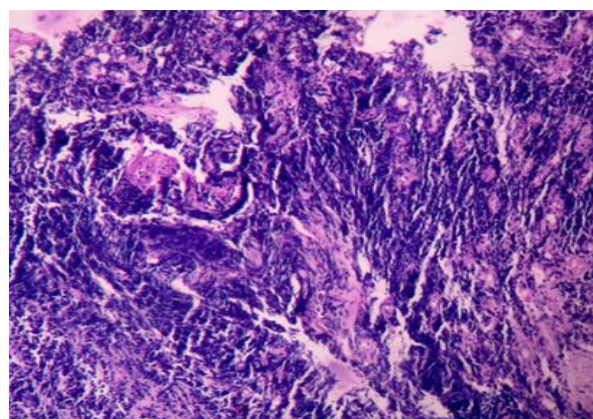
**Figure 1: Photomicrograph of BAL showing normal respiratory epithelial cells along with hemosiderin laden macrophages (Pap 400x).**



**Figure 2: Photomicrograph of Squamous cell carcinoma lung moderately differentiated (H&E 100x).**

The smoker to non-smoker ratio was 2.89:1 The most common type of lung cancer on biopsy was squamous

cell carcinoma 68.55% (521cases) (Figure 2) followed by small cell carcinoma 23.02% (175cases) (Figure 3) and adenocarcinoma 4.93% (37cases) (Figure 9) including bronchioalveolar carcinoma (7 cases). There was one case each of adenoid cystic carcinoma and leiomyosarcoma diagnosed on biopsy (Table 2). These were included in the miscellaneous group.



**Figure 3: Photomicrograph of small cell carcinoma (H & E 100x).**

In our study BAL was positive for malignancy in 33% (301) cases and negative in 67% cases (601). It was seen that for bronchoscopically visible lesions BAL was positive in 31.83% cases and for lesions not visible bronchoscopically BAL was positive in 41.91% cases. 61 cases (20.26%) were categorized by BAL as squamous cell carcinoma (Figure 10), 4 cases (1.33%) as small cell carcinoma 8 cases (2.65%) as adenocarcinoma (Figure 5, Figure 8), 2 cases (0.66%) as large cell carcinoma, 31 cases (10.29%) as suspicious / atypical and 195 cases (64.78%) as poorly differentiated carcinoma (Table 3).

**Table 2: Histopathological findings on bronchial biopsy.**

Histopath.	Males	%	Females	%	Total	%
Squamous cell carcinoma	462	71.40%	59	52.21%	521	68.55%
Small cell carcinoma	145	22.41%	30	26.54%	175	23.02%
Adeno carcinoma	21	3.24%	16	14.15%	37	4.93%
Large cell carcinoma	6	0.92%	1	0.88%	7	1.26%
Carcinoid	1	0.15%	3	2.65%	4	0.7%
Undifferentiated carcinoma	2	0.30%	2	1.76%	4	0.7%
Miscellaneous	2	0.30%	0	-	2	0.26%
Poorly differentiated carcinoma	8	1.23%	2	1.76%	10	1.31%
Total	647	100%	113	100%	760	100%

In the present study 270 were correctly diagnosed by BAL as malignant, 31 cases as suspicious/atypical which later on biopsy proved to be benign lesions and were included in the false positive cases (Table 4). Total

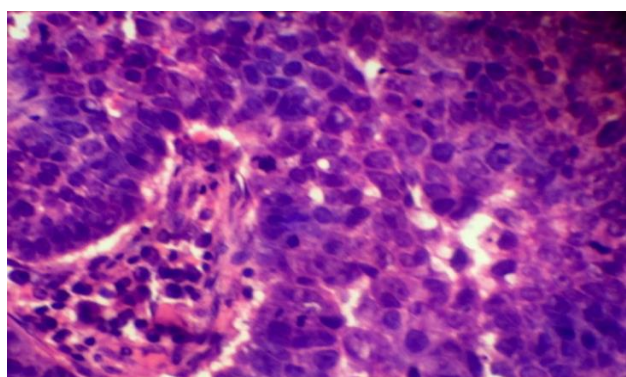
number of negative cases diagnosed on BAL was 601 out of which 111 were true negative and 490 were false negative. The reasons for large number of false negative cases in our study were due to superadded inflammation



in majority of the cases and non representative material in rest of the cases. In our study there were 31 false positive cases. On biopsy these were benign lesions which included 15 cases of inflammatory lesions (48%), 9 cases of tuberculosis (29%) and 7 (23%) were of no significant pathology.

**Table 3: Correlation of BAL with biopsy.**

Tumour Type	BAL		Biopsy	
	No.	Percentage	No.	Percentage
Squamous cell carcinoma	61	20.26%	521	68.55%
Small cell carcinoma	4	1.33%	175	23.02%
Adenocarcinoma	8	2.65%	37	4.93%
Large cell carcinoma	2	0.66%	7	1.26%
carcinoid	-		4	0.7%
undifferentiated	-		4	0.7%
Miscellaneous type	-		2	0.26%
Atypical/Suspicious	31	10.29%		
Poorly differentiated	195	64.78%	10	1.31%
Total	301		760	

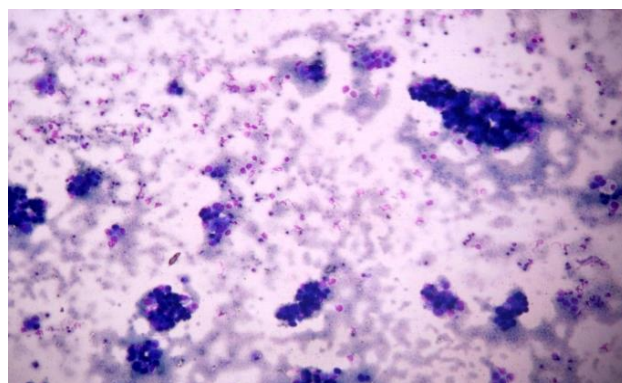


**Figure 4: Photomicrograph of large cell carcinoma lung (H&E100x).**

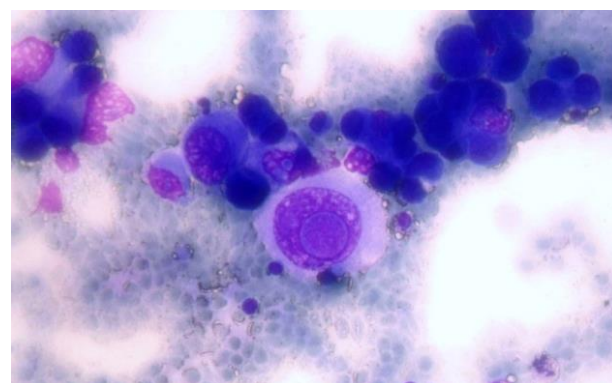
**Table 4: Cyto histological correlation of lung malignancies.**

Diagnostic accuracy of BAL (n=902)			
Gold standard histopathology			
BAL For malignancy	Disease Present	Disease not present	Total
Test positive	True positive 270	False positive 31	301
Test negative	False negative 490	True negative 111	601

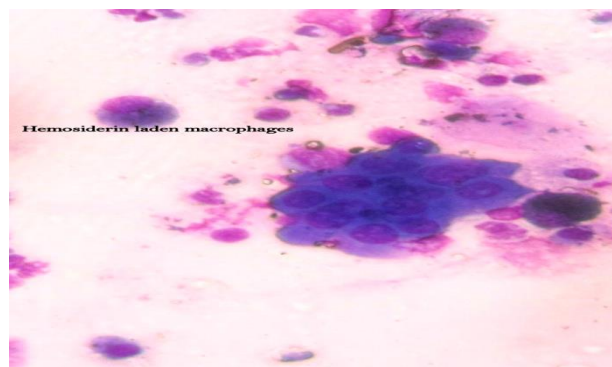
True positive (TP) = 270 (29.93%)  
 True Negative (TN) = 111 (12.30%)  
 False Positive (FP) = 31 (3.43%)  
 False Negative (FN) = 490 (54.32%)  
 Sensitivity = 35.5%  
 Specificity = 78.16%  
 Positive predictive value (PPV) = 89.70%  
 Negative Predictive value = 18.46%  
 Diagnostic accuracy = 42.23%  
 Sensitivity of BAL was found to be 35.5% and specificity 78.16%



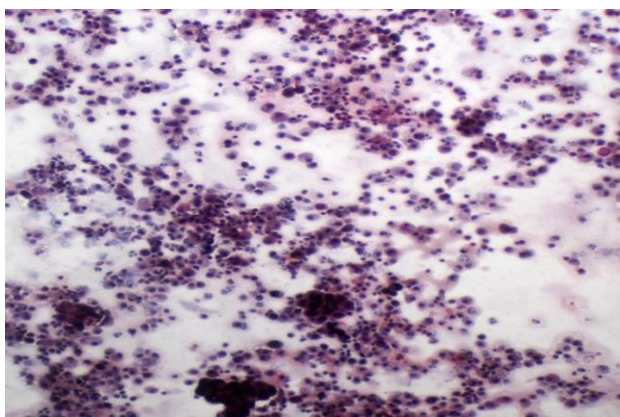
**Figure 5: Photomicrograph of BAL fluid with tumor cells forming papillae- Papillary Adenocarcinoma (MGG 100x).**



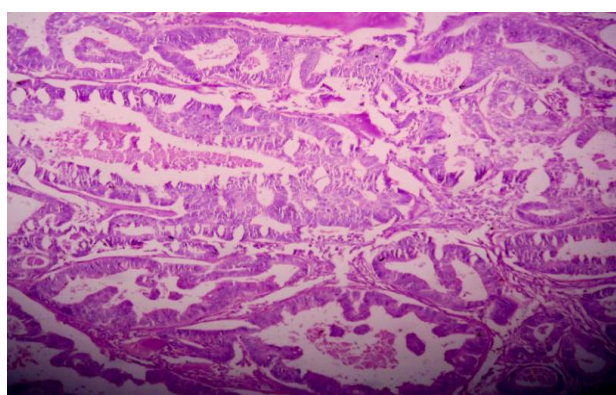
**Figure 6: Photomicrograph of bronchoalveolar carcinoma showing prominent intranuclear inclusion (MGG 400x).**



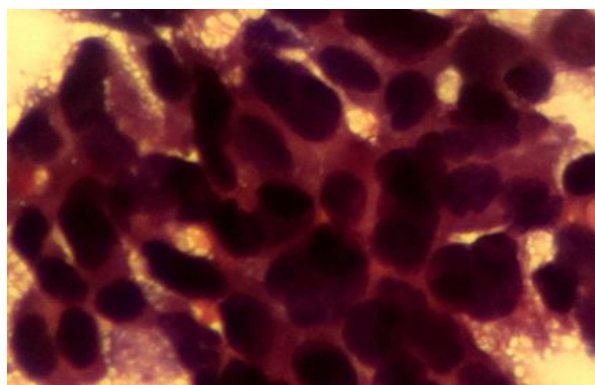
**Figure 7: BAL fluid positive for malignant cells (MGG100x).**



**Figure 8: Photomicrograph of BAL positive for malignant cells- adenocarcinoma in a background of hemosiderin laden macrophages (Pap100x).**



**Figure 9: Photomicrograph of adenocarcinoma Lung (H & E100x).**



**Figure 10: Photomicrograph of BAL fluid positive for malignancy (Squamous cell carcinoma) (Pap 100x).**

## DISCUSSION

Lung cancer is the leading cause of cancer related deaths in men and in women it has surpassed even breast cancer.<sup>2</sup> The increase number of the lung cancer deaths is mainly because it is detected at a late stage. Timely detection of disease plays an important role in the management and long term survival of patients<sup>21</sup>. Pulmonary cytology and histopathology are

valuable tools in the diagnosis of lung malignancies. Bronchoalveolar lavage is considered as an effective tool for diagnosis of this condition. It can provide diagnostic information in cases of primary and metastatic disease to the lung. Though BAL is inferior to bronchial biopsy in diagnosing lung malignancies, it has certain advantages. It is a relatively safe procedure requiring less expertise, can be undertaken in peripheral lesions and in patients at risk of bleeding.<sup>22</sup> Even those lung tumors which are not visible through bronchoscope might be picked up in bronchoalveolar lavage.

In all, 760 (84.25%) cases were diagnosed by bronchial biopsy to be suffering from lung cancer, of which 647 were males and 113 were females. The male to female ratio was 5.72:1. In a study Faludi et al in their study found male to female ratio 6.22:1, whereas study by Bodh et al 2013 showed male: female ratio of 3.35:1.<sup>10,22</sup> Tobacco smoking and environmental pollution have been found to be the main etiological factors for lung cancer. The smoker to non-smoker ratio was 2.91:1. Our smoker to non-smoker ratio correlates well with that of Rajasekaran et al and Gupta D et al the most common type of lung cancer on biopsy was squamous cell carcinoma 68.55% (521cases).<sup>23,24</sup> The second most common type of lung cancer was small cell carcinoma 23.02% (175 cases), followed by adenocarcinoma 4.93% (37cases) including bronchioalveolar carcinoma (7 cases). The relative frequency of small cell carcinoma and adenocarcinoma was higher in females as compared to males in our study. Similarly carcinoid tumours were found more in females than in males in our study. Khan et al in their study found squamous cell carcinoma as the most common type of lung tumor (77.3 percent) followed by small cell carcinoma (17.1 percent).<sup>21</sup> Adenocarcinoma was seen in 5.3 percent and large cell anaplastic carcinoma in 1 percent.

Bodh et al in their study found squamous cell carcinoma (38.70%) as the most common sub type followed by small cell carcinoma in (27.10%), and adenocarcinoma (23.87%).<sup>10</sup> From the above we conclude that squamous cell carcinoma is the most common histological type of lung cancer as is seen in our study also. Small cell carcinoma is the second commonest type of lung cancer in our study which is comparable with the studies done by Sheema et al and Bodh et al.<sup>10,25</sup> Our study is at variance to western literature where incidence of adenocarcinoma has surpassed squamous cell carcinoma.<sup>26,27</sup> In the present study BAL was positive in 301 cases. Of these 61 cases (20.26%) were categorized by BAL as squamous cell carcinoma, 4 cases (1.33%) as small cell carcinoma 8 cases (2.65%) as adenocarcinoma, 2 cases (0.66%) as large cell carcinoma, 31 cases (10.29%) as suspicious/atypical and 195 cases (64.78%) as poorly differentiated carcinoma.

Pirozynski found in their study on 145 patients with biopsy proven lung cancer, BAL was diagnostic in 64.8% revealing malignant cells.<sup>28</sup> In 35.9% of these patients,



the cytologic diagnosis correlated with final diagnosis of resected tumour. Wongsurkai et al found that the cell type diagnosed by BAL correlated with final diagnosis in 50% of patients.<sup>29</sup> Gaur D S et al in their study BAL was diagnostic in 17.9% cases (5/28) of squamous cell carcinoma, 7.1% cases (2/28) as small cell carcinoma and as many as 71.4% samples classified as poorly differentiated carcinoma.<sup>30</sup> Thus squamous cell

carcinoma is the most common type of lung cancer diagnosed by BAL in the above studies which is comparable to our study.

In our study tumor categorisation was done by BAL in approximately 25% cases which is comparable to the study done by Gaur et al but at variance to the study done by Pirozynski, Wongsurkai et al.<sup>28,29,30</sup> (Table 5).

**Table 5: Comparative statistical values on cyto-histological correlation.**

Author (year)	Place	No. of cases	Sensitivity (%age)	Specificity (%age)	Diagnostic accuracy (%age)
Pirozynski et al (1992) <sup>28</sup>	Poland	145	64.8%		
de Gracia (1993) <sup>31</sup>	Spain	67	33%	-	-
Wongsurkai et al (1998) <sup>29</sup>	Thailand	55	46.7%		
Gaur DSetal (2007) <sup>30</sup>	India	196	39.4%	89.6%	71.40%
Tuladhar et al (2011) <sup>32</sup>	Nepal	55	66.7%		
Present study (2015)	India	902	35.5%	78.16%	42.43%

In the present study, 270 were correctly diagnosed by BAL as malignant, 31 cases as suspicious/atypical which later on biopsy proved to benign lesions and were included in the false positive cases. Total number of negative cases diagnosed on BAL was 601 out of which true negative were 111 and false negative were 490. The reasons for large number of false negative cases in our study were due to superadded inflammation in majority of the cases and non representative material or hypocellular aspirates in rest of the cases.

Similar results were achieved by Wongsurakiat et al.<sup>29</sup> They had a lot of false negatives in their study. They reported that in five patients with metastatic lung cancer BAL gave negative results in all.

The study by Gaur et al showed 43 false negative of 196 cases.<sup>30</sup> Since cytological sampling by BAL technique relies mainly on cells 'exfoliated' from the malignant lesion in the bronchial epithelium, the adequacy of its samples depends on several vital factors, especially a) the degree of differentiation of malignant growth b) preservation of the morphology of cytological material obtained; and c) technical skill of the pulmonologist who is retrieving the lavage fluid from the bronchus. In general, less differentiated, anaplastic lesions have more loosely cohesive cells in comparison to well differentiated lesions.

Thus such lesions exfoliate larger number of cells into the bronchial cavity than the well differentiated lesions. Secondly, while these exfoliated cells are lying in the bronchus, they start developing degenerative changes, thus progressively losing their morphological details which are important in differentiating them from non-malignant cells shed off by the normal bronchial

epithelial lining. Usually around 20ml. saline is instilled through the bronchoscope for BAL samples. If the technique of the pulmonologist is not proper, the sample retrieved might be less in amount and thus may have lesser cytological material than expected, thus again increasing the chances of false negative results. All these factors present individually or together, affect the overall yield and diagnostic value of BAL specimens. Present study revealed 31 false positive cases (3.43%). In a study by Gaur et al there were 13 false positive of 196 cases (6.63%).<sup>30</sup>

This comparison suggests that less number of false positive is strength of BAL cytology.

False positive can be mainly due to misinterpretation of the smears by the cytologist due to cellular changes in chronic inflammatory disorders such as chronic pneumonia (atypical histiocytes), tuberculosis (epithelioid cells), pneumonitis (misinterpretation of cuboidal alveolar cells as small cell carcinoma), squamous metaplasia and alveolar cell polymorphism in lung fibrosis.

In the present study false positive cases were because of misinterpretation of squamous metaplasia as suspicious in 10 cases. In rest of the cases it was due to misinterpretation of cuboidal alveolar cells as suspicious/atypical. False positives have very unfortunate consequences for the individual patients, therefore it is better "under reporting" instead of "over reporting" in suspicious cases. If cytology is positive for malignancy or suspicious cells repeat biopsy, clinical correlation with radiological/bronchoscopic findings is necessary.

The values obtained in the present study are comparable to the study done by Gaur et al but a little less than the values obtained in most of the previous studies.<sup>30</sup> The reason for this variance is use of different techniques for retrieval and processing of cytological specimens, and different practices with regard to suspicious cytological appearance.

Studies have shown that increasing the number of attempts at obtaining BAL sampling can improve its sensitivity, specificity and accuracy.<sup>33,34</sup> The statistical values in our study were obtained with a single sample. This might have been another result as to why our values were less than the previous studied.

## CONCLUSIONS

Thus in the present study yield of diagnosis was highest with the bronchoscopic biopsies and in maximum number of cases, specific histologic diagnosis was made by biopsies only. Sensitivity of BAL in our study was 35.5% and specificity was 78.16%. Though BAL was inferior to bronchial biopsy in diagnosing lung malignancies but it was effective for peripheral lung malignancies and when the patient was at risk of hemorrhage.

## ACKNOWLEDGEMENTS

Authors acknowledge the immense help received from the scholars whose articles are cited and included in references of this manuscript. The authors are also grateful to authors / editors / publishers of all those articles, journals and books from where the literature for this article has been reviewed and discussed.

*Funding: No funding sources*

*Conflict of interest: None declared*

*Ethical approval: The study was approved by the Institutional Ethics Committee*

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**Cite this article as:** Bhat N, Nazeir MJ, Bashir H, Bashir N, Farooq S, Fatima K, Baba KM. Correlation of bronchial biopsy with bronchoalveolar lavage in lung malignancies. *Int J Res Med Sci* 2016;4:428-35.