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A Descriptive Study of Diagnostic Accuracy of Small Biopsy Imprint Cytology in Neoplastic Lung Lesions in a Tertiary Care Hospital

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Abstract: ***Introduction:*** *Lung cancer is currently the most frequently diagnosed cancer in the world and is a major healthcare problem in India. It should be diagnosed at the earliest possible stage. The prognosis of lung cancer is related to factors like stage, size, presence or absence of visceral pleura and lymph node invasion.*
Materials and Methods: *A descriptive study was done on first 54 small biopsy specimens of suspected neoplastic lung lesions and imprint smears prepared from same biopsy received in the department of pathology, Kurnool Medical College, Kurnool, during the study period of 12 months (June 2017- May 2018).*
Results: *Diagnostic test evaluation of imprint cytology smears was done on 54 cases of lung lesions received in Department of pathology, Kurnool Medical College, Kurnool during study period of 12 months (June 2017-June 2018). The mean age distribution of the present study population is 62.11 years, minimum age is 28 years and maximum age is 78 years and majority belong to age group 60-69 years (43%). 88% of cases were male patients. 65% of cases had lesion in Right lung and 35% had lesion in left lung. 24 % (n=13) of patients had lesion in the Right lower lobe of lung followed by Left upper lobe (12 cases, 22%). 77.8 % of the cases had a history of smoking.*
Conclusion:
Diagnostic accuracy of imprint cytology in case of NSLC and SCLC were 79% and 94.4% respectively. The diagnostic accuracy of imprint cytology for the squamous cell carcinoma was 70.3% and that for adenocarcinoma was 90% Metastasis and carcinoid showed 100% cyto-histology concordance. So, performing imprint smear cytology as an adjunct diagnostic tool to biopsy in cases of neoplastic lung lesions may be recommended.
Key Words: *Neoplastic Lung Lesions, Imprint Cytology, Small Biopsy, Diagnostic Accuracy*

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I. Introduction

Lung cancer is currently the most frequently diagnosed cancer in the world and is a major healthcare problem in India.1 It should be diagnosed at the earliest possible stage. The prognosis of lung cancer is related to factors like stage, size, presence or absence of visceral pleura and lymph node invasion.[1,2,3]

Histologic diagnosis and staging is essential for selecting the mode of treatment. Stages I and II are amenable to surgery, advanced stages need combination of surgery, chemotherapy and radiotherapy Currently with better understanding of cellular mechanisms operating in carcinogenesis, has led to discovery of activating mutations in the epidermal growth factor receptor (EGFR) and rearrangements of the anaplastic large-cell lymphoma kinase (ALK) gene.[4]

Clinical guidelines have now incorporated molecular testing and the use of drugs targeting these genes.[5] For early diagnosis different diagnostic modalities are available which include; radiology, bronchoscopy, bronchial biopsy, exfoliative cytology, imprint smears, brushing, and washing. Cytologic examination of imprint cytology of small biopsy, needle aspirate, sputum and bronchial secretions helps to determine the presence and classify the tumour as accurately as possible according to predominant histologic type. This task is of considerable importance, since it may influence the mode of treatment in individual cases. Cytologic procedures serve to render the diagnosis of lung cancer with precision, speed, and accuracy equal to or even superior to other techniques. The benefits of cytologic methods are substantial.

II. Materials and Methods

Study Design: Descriptive study.
Study Setting: Department of Pathology, Kurnool Medical College, Kurnool.
Study Period: June 2017- May 2018.

Study Population

First 54 small biopsy specimens taken from lung lesions in suspected cases of carcinoma lung and their imprint smear sample taken from same biopsy received in Department of Pathology, Kurnool Medical College, Kurnool.

Inclusion Criteria

First 54 small biopsy specimen of lung lesions in suspected cases of carcinoma lung and imprint smear sample taken from same biopsy.

Exclusion Criteria

Inadequate biopsy samples and imprint smears with low cellularity.

Study Tools

1. Instruments for tissue processing.
2. Reagents for tissue processing.
3. Glass slides and cover slips for mounting.
4. Microscope
5. Reagents for Haematoxylin & eosin staining and special stain if needed
6. Reagents for PAP and Giemsa staining for cytology
7. Proforma to record serial number, Biopsy number, Name, age, sex, gross, histopathology and cytology features.

Study Procedure

Samples of small tissue biopsy specimen and their imprint smears taken from lung lesion of suspected cases of carcinoma lung received in our department were selected. Both cytology and histopathology of samples were studied, and their results were compared. IHC study and special stains were also done in needed cases for subtyping. All slides were viewed under low power and high power.

Imprint Smear Cytology Examination

Imprint smear taken from small biopsy received in the cytology section will be fixed in 85% isopropyl alcohol and stain with Papanicolaou stain and Giemsa stain.

Histopathological Examination

Small biopsy will be entirely submitted for tissue processing after 24-hour 10% formalin fixation. 4 microns thick sections will be made from paraffin embedded formalin fixed tissue. All sections will be stained with H&E and special stains like mucicarmine and IHC will be done in needed cases. Imprint smear from same small biopsy received in the cytology section will be fixed in 85% isopropyl alcohol and stain with Papanicolaou stain and Giemsa stain. All cytology smears and histopathology slides are viewed under low power and high power.

Immunohistochemical Staining

Immunohistochemistry was performed on 3-micron thick sections cut from formalin fixed paraffin embedded tissue. p63 and TTF1 were done for subtyping of non-small cell carcinoma which shows nuclear positivity in neoplastic cells. Chromogranin A and synaptophysin were done for confirming small cell carcinoma. Synaptophysin shows nuclear positivity and chromogranin A shows cytoplasmic positivity in neoplastic cells.

III. Results

Diagnostic test evaluation of imprint cytology smears was done on 54 cases of lung lesions received in Department of pathology, Kurnool Medical College, Kurnool during study period of 12 months (June 2017-June 2018). The mean age distribution of the present study population is 62.11 years, minimum age is 28 years and maximum age is 78 years and majority belong to age group 60-69 years (43%). 88% of cases were male patients. 65% of cases had lesion in Right lung and 35% had lesion in left lung. 24 % (n=13) of patients had lesion in the Right lower lobe of lung followed by Left upper lobe (12 cases, 22%). 77.8 % of the cases had a history of smoking.

Among the 54 cases of histopathologically proven lung tumours, 42 were diagnosed (78%) in imprint cytology also, a few atypical cells observed in 13% and rest 5% were negative for malignancy. Among the 42 cases diagnosed by imprint cytology, NSCC predominate (83%, 35 cases), but the subtyping of NSCC was possible in 51% of cases. Among the lung tumours diagnosed by histopathology, NSCC predominate (44 cases, 81%) but subtyping was possible in 70% of cases. Among the 54 cases studied, 8 cases were diagnosed as Small cell carcinoma by histopathology, which showed 63% concordance with cytology. Of the 44 cases of non-small cell carcinoma diagnosed by histopathology, 77% cases were concordant with cytology. Metastasis and carcinoid showed 100% cytology- histopathology concordance. Among the 44 cases of NSCC studied, 22 cases were diagnosed as Squamous cell carcinoma by histopathology, which showed 55% concordance with cytology.

Nine cases of Adenocarcinoma were diagnosed by histopathology, 67% of which showed concordance with cytology. 17 cases of poorly differentiated carcinoma were diagnosed by histopathology which showed 76% concordance with cytology. Sensitivity of imprint smear cytology in diagnosing carcinoma lung was found to be 77%. Sensitivity of imprint cytology in the diagnosis of small cell carcinoma was found to be 62% and specificity 100%. Sensitivity of imprint cytology in the diagnosis of Non- small cell carcinoma was found to be 77% and specificity 90%. Sensitivity of imprint cytology in the diagnosis of Squamous cell carcinoma is only 40% and specificity is 90%. Sensitivity of imprint cytology in the diagnosis of Adenocarcinoma is 55% and specificity is 97%. Sensitivity of imprint cytology in the diagnosis of poorly differentiated carcinoma is 61% and specificity is 78%.

In the present study, IHC marker study was done on 16 selected cases of bronchial biopsy, which included 9 cases with cyto-histo correlation and 7 cases without cyto-histo correlation. Among the nine cases, which had cyto-histo correlation, the IHC was consistent with cyto-histological diagnosis in five cases. Rest of the four had morphological diagnosis of poorly differentiated carcinoma, which were sub typed by IHC as adenocarcinoma in two cases & squamous cell carcinoma in two cases. Among the seven cases, which had no cyto-histo correlation, the IHC was found to be consistent with cytological diagnosis in two cases while consistent with histopathological diagnosis in 3 cases. Rest two cases in which cytology smears were negative for malignant cells but diagnosed as poorly differentiated carcinoma in histopathology, the IHC helped to identify one case as squamous cell carcinoma and other as adenocarcinoma.

S.No	Cytology Diagnosis	Number (%)
1	**SCLC**	**5 (12%)**
2	**NSCC-SQCC**	**12 (29%)**
3	**NSCC-ADC**	**6 (14%)**
4	**NSCC-PDC**	**17 (41%)**
5	**METS**	**1 (2%)**
6	**NET-carcinoid**	**1 (2%)**

Table 1: Specific Distribution of Lung Tumours According to Imprint Cytology Diagnosis

S.No	Cytology Diagnosis	Number (%)
1	**SCLC**	**8 (14.8%)**
2	**NSCC-SQCC**	**22 (40.7%)**
3	**NSCC-ADC**	**9 (16.7%)**
4	**NSCC-PDC**	**13 (24.1%)**
5	**METS**	**1 (1.9%)**
6	**NET-carcinoid**	**1 (1.9%)**

Table 2. Specific Distribution of Lung Tumors According to Histopathological Diagnosis

S.No	Histo-pathological Diagnosis	No of cases	Cyto- Histological Concordance	Cyto- Histological Discordance
1	**SCLC**	**8**	**5 (63%)**	**3 (37%)**
2	**NSCC**	**44**	**34 (77%)**	**10 (23%)**
3	**METS**	**1**	**1 (100%)**	**0**
4	**NET-CND**	**1**	**1 (100%)**	**0**

Table 3: correlation between cytology and histopathology of lung carcinoma

Cytology	Histopathology		Total
	Positive	**Negative**	
Positive	**42**	**0**	**42**
Negative	**12**	**0**	**12**

Table 4: Imprint Smear Cytology v/s Histopathology Cross Table to Assess Sensitivity

A Case with Cytology- Histopathology Concordance

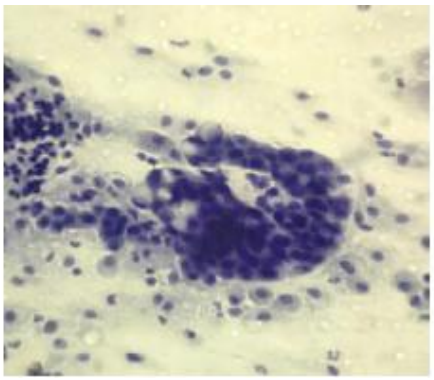

Figure 1: Imprint Cytology-Adenocarcinoma

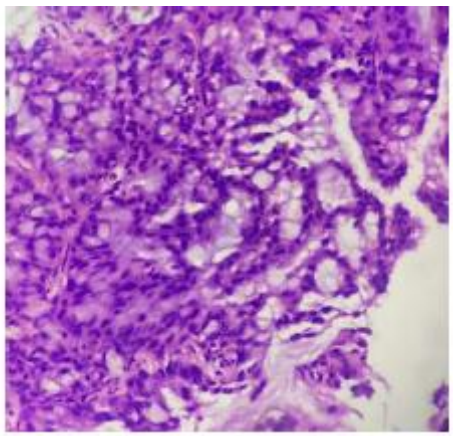

Figure 2: *Adenocarcinoma Lung (H &E Stain 10X)*

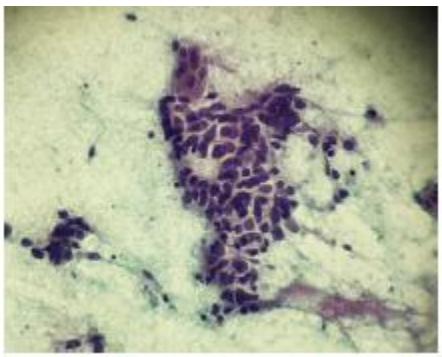

Figure 3: Imprint Cytology-Small Cell Carcinoma

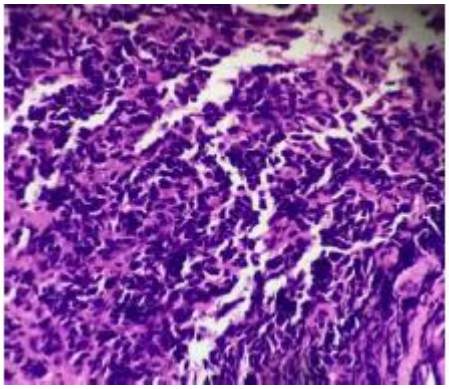

Figure 4: *Small Cell Carcinoma Lung (H &E stain 40X)*

A Case with Cytology- Histopathology Discordance

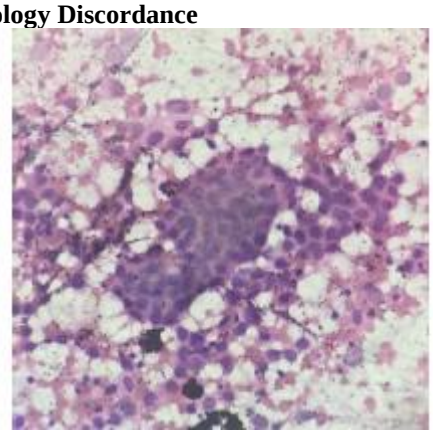

Figure 5: Imprint Cytology Diagnosed as Adenocarcinoma

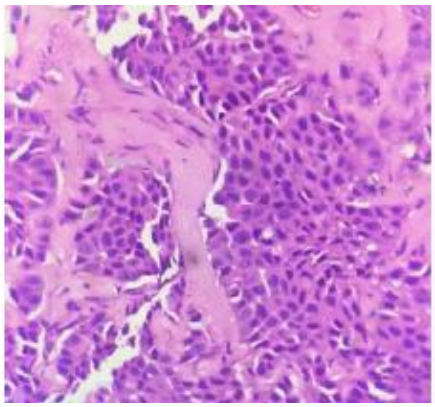

Figure 6: Histopathology Diagnosed as Squamous

IHC of the Histopathology Section

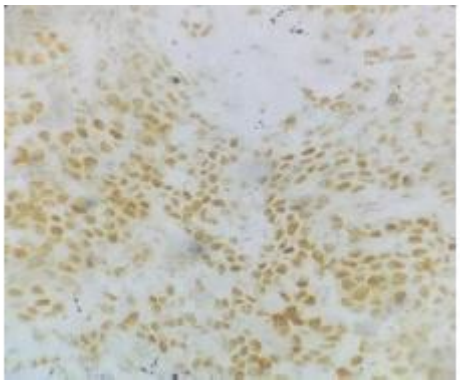

Figure 7: *TTF1 Positivity in Nucleus (40X)*

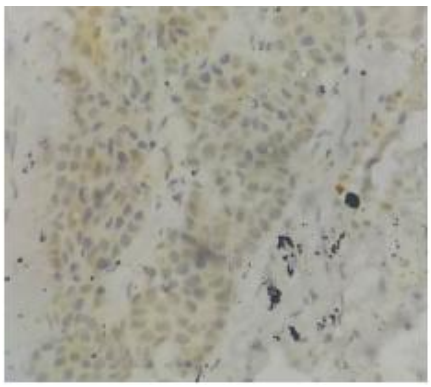

Figure 8: p63 Negative Nuclei (40X)

IV. Discussion

The present study was conducted with the objective of assessing the diagnostic accuracy of small biopsy imprint cytology compared to gold standard histopathology. In the present study, cases showed a wide range of distribution from 28-78 years, with a peak in the age group of 60-69 years, and mean age of 62.11 years.[6] This finding was correlating with the study of Anita Bodh et al.[7] 65% of cases in the study group had lesion in Right lung, which is comparable with study by Sumita Das et al.[8] In the present study, by imprint smear cytology, Non- small cell lung carcinoma predominate, but subtyping was possible in 51% of cases. Squamous cell carcinoma (29 %) was found to be commonest, followed by adenocarcinoma (14 %) and small cell carcinoma (12%), the findings are comparable with study by Anita Bodh et al.[9] By histopathological examination, Non-small cell lung carcinoma predominates, but subtyping was possible in 70% of cases. Squamous cell carcinoma (40%) was found to be commonest, followed by adenocarcinoma (16%) and small cell carcinoma (14%), the findings are comparable with study by Anita Bodh et al. The sensitivity of imprint cytology in diagnosing carcinoma lung is 77% in this study, which is comparable with study by Anita Bodh et al.[10]

V. Conclusion

Imprint cytology can be used as a diagnostic tool as it is a quick, simple, cost effective, reliable and accurate technique, which enhances the known benefits of small biopsy. It does not alter the quality of biopsy specimen. There is no requirement of any specialised instruments for taking imprint smears. Imprint cytology helps to guarantee that the specimens obtained adequately represent the lesion. It reduces the waiting time for diagnosis and increases diagnostic performance over aspiration cytology. This helps in patient management through the earlier availability of the diagnosis and fewer outpatient appointments. In the present study, Small cell carcinoma showed 63% cyto-histological concordance whereas, non-small cell carcinoma has 77% cyto-histological concordance. Metastasis and carcinoid showed 100% cyto-histology concordance. Diagnostic accuracy of imprint cytology in case of NSLC and SCLC were 79% and 94.4% respectively. The diagnostic accuracy of imprint cytology for the squamous cell carcinoma was 70.3% and that for adenocarcinoma was 90%. Metastasis and carcinoid showed 100% cyto-histology concordance. So, performing imprint smear cytology as a diagnostic tool in cases of neoplastic lung lesions can be recommended.

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