

Cytopathological spectrum of pulmonary lesions and their correlation with histopathological findings

Jenice Chhatkuli¹, Ananda Neupane², Dilasma Gharti Magar³, Sushma Thapa³, Sudeep Regmi³

¹ Department of Pathology, Rapti Academy of Health Sciences, Ghorahi, Dang Nepal

² Department of General Surgery, Rapti Academy of Health Sciences, Ghorahi, Dang Nepal

³ Department of Pathology, Manipal College of Medical Sciences, Pokhara, Kaski, Nepal

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Corresponding Author:

Jenice Chhatkuli
Lecturer
Department of Pathology,
Rapti Academy of Health Sciences,
Ghorahi, Dang, Nepal
Email: drjenice07@gmail.com

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Abstract

Introduction: Neoplastic and non-neoplastic pulmonary lesions account for majority of mortality and morbidity worldwide. Respiratory tract cytology is a globally recognized diagnostic technique employed to assess individuals with presumed lung abnormalities. The objective of this study was to analyze the cytopathological features of pulmonary lesions and correlate with respective histopathological findings.

Methods: A hospital based longitudinal study was conducted in Department of Pathology, Manipal Teaching Hospital from October 2021 to November 2022 including 155 cytology samples. Ethical clearance was obtained from the institutional review committee and consent was taken from the participants. The microscopic findings in the cytology samples were analyzed and the diagnosis was compared with the histopathological diagnosis.

Results: The mean age of patients was 60.9 ± 1.32 SD years and male: female ratio was 1.31:1. Cough was the most common chief complaint. Cytological diagnosis was classified into five categories as positive for malignancy (22.6%), suspicious for malignancy (5.8%), benign infectious lesion (8.4%), negative for malignancy (60.6%) and inadequate for opinion (2.6%). Adenocarcinoma was the most common (42.8%) malignancy followed by squamous cell carcinoma (34.2%). Abscess was the predominant lesion (46.1%) followed by tuberculosis (30.7%) in benign infectious category. Biopsy samples for histopathological correlation were available for 33 cases. The sensitivity, specificity, positive predictive value and negative predictive was 88% (95% CI 68.8-97.5), 66.67% (95% CI 9.4-99.2), 95.6% (95% CI 78.1-99.9) and 40% (95% CI 5.3-85.3) respectively. The concordance rate between cytological and histopathological diagnosis was 85.6%.

Conclusion: The findings from this study demonstrate that respiratory cytology is a valuable initial diagnostic modality with high sensitivity and substantial concordance.

Keywords: Cytology, histopathology, malignancy

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Introduction

Neoplastic and non-neoplastic pulmonary lesions account for majority of mortality and morbidity worldwide. Currently, lung cancer is the most frequently diagnosed cause for cancer related deaths. The increasing incidence could be due to increase in smoking, change in life style,

increased environmental pollution and also the availability of different modern diagnostic modalities to detect lung cancer.¹

Cytology of pulmonary lesions provide valuable diagnostic information by non-invasive or minimally invasive procedures.² Cytology is of great significance in situations

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where biopsy procedure is difficult or impossible to perform due to high risk of hemorrhage.³ Various sampling techniques currently available include exfoliative cytology (induced sputum, pleural fluid and thoracentesis), abrasive cytology (bronchioalveolar lavage (BAL), bronchial biopsy (BB) and bronchial washing); and endobronchial, transesophageal ultrasound-guided (USG) and computed tomography (CT) - guided percutaneous / transthoracic fine needle aspiration cytology (FNAC).² It is not possible to perform all techniques in each patient because each has its own specific advantages and disadvantages. However, their combined use yields the best results.⁴ Although some studies regarding the efficacy of these cytological techniques in diagnosing lung diseases have been conducted in Nepal, enough literature is lacking regarding the same based on western region of Nepal.

This study aimed to assess the cytopathological features of pulmonary lesions prevalent in a tertiary care center in the Western region of Nepal, through the use of different cytological techniques and evaluate their diagnostic accuracy by comparing with the histopathological findings.

Methods

A hospital based longitudinal study was conducted in Department of Pathology, Manipal Teaching Hospital as this is a tertiary health care center serving patients from wide geographical region.

Sample size determination was done by using formula:

$$N = (z^2 \times p \times q) / d^2 = (1.96)^2 \times 0.122 \times 0.878 / (0.10)^2 = 41.14$$

Where: N = required sample size, z = reliability coefficient (1.96 for 95% reliability), p = estimated prevalence (p= 12.2% WHO, Globocan 2020)⁵, q = 1-p, d = margin of error at 10%.

With this formula the total sample size was minimum of 42. However, all consecutive cytopathology samples i.e. pleural fluid, FNAC, BAL and sputum received during the study period (October 2021 to November 2022) were included along with corresponding biopsy samples whenever available using simple convenience sampling technique. Written informed consent from the participants were obtained after ethical clearance was taken from the institutional review committee (IRC) of Manipal College of Medical Sciences, Pokhara, Nepal (IRC clearance dated 15th August 2021). The samples were available from the department of pathology which was received from different clinical departments of hospital. All cytology samples (including sputum, BAL, BB, Pleural fluid, FNAC) and corresponding biopsy specimen from patients presenting with pulmonary lesions were included in the study. Inadequate cytology and biopsy specimen, poorly fixed and unlabeled specimens were excluded from the study.

For FNAC, the procedure was carried out under radiology (USG/CT) image guidance and aspiration was done by 10 ml syringe. BAL and pleural fluid sample were centrifuged and the sediment was used for preparing smears. For each sample, 2 smears were stained with Papanicolaou stain and at least 2 air dried smears were stained with Leishman stain. Special stains such as Ziehl Neelsen and Periodic-Acid Schiff stain (PAS) were used when required. Biopsy samples were processed and stained with Hematoxylin and Eosin (H&E) stain and special stain (PAS) when needed. The slides were observed under microscope and findings were noted.

Statistical analysis: The data were entered in Microsoft Excel and the statistical calculations were done using Statistical Package for Social Science (SPSS) version 25. Results were expressed as frequencies, percentage and mean with standard deviation (SD) in bar diagram, pie chart and tables Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and diagnostic accuracy were calculated at 95% confidence interval for the different cytological techniques.

Results

A total of 155 cases were included in the study with a male: female ratio of 1.3:1. The mean age of the patients included in the study was 60.9 ± 1.32 SD years with the age ranging from 3 years to 98 years.

Although a wide range of age group was included in the study, higher number of samples were received from the elderly population. The age group of 71-80 years constituted the most cases (43 cases).

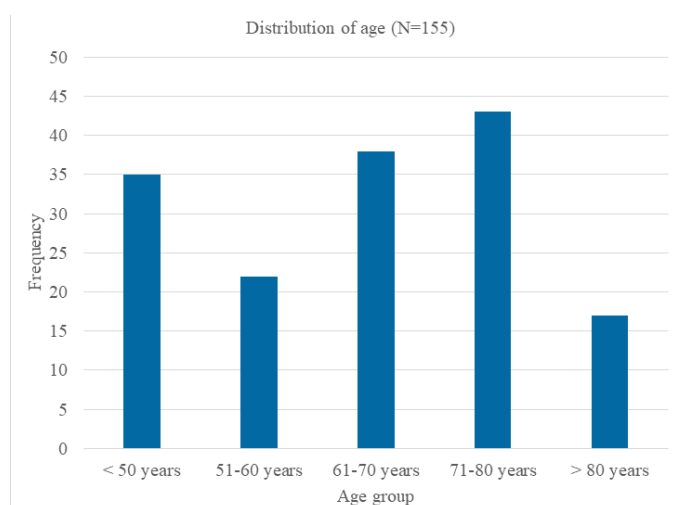


Figure 1: Bar diagram showing distribution of age among sample population (N=155).

Cytology specimen received for the study were pleural fluid, FNAC, BAL and sputum. Pleural fluid was the most frequent sample sent for analysis (58%) whereas sputum was the least received (4%).

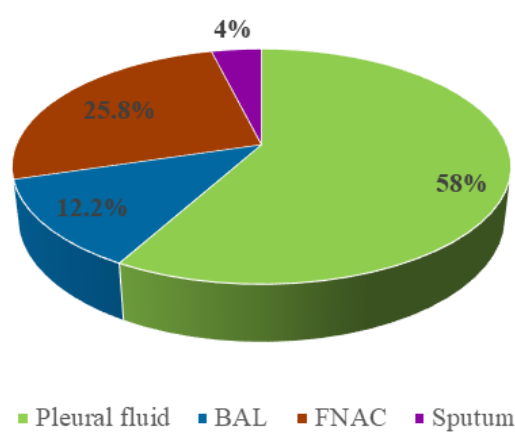


Figure 2: Pie chart showing frequency distribution of different cytology specimen (N=155).

The commonest chief complaint in present study was cough 25 (16.1%), followed by fever and chest pain 20 (12.9%) each, shortness of breath 11 (7.1%), weight loss 6 (3.9%), and hemoptysis 4 (2.6%). However, 69 (44.5%) cases came with multiple complaints at presentation. In our study, the most common malignancy seen among smokers and alcoholic was adenocarcinoma (7 cases, 30.4%) followed by squamous cell carcinoma (SCC) (6 cases, 26.2%) and small cell lung carcinoma (SCLC) (3 cases, 13%).

FNAC was the most common diagnostic modality for detecting malignancies with 24 (15.5%) cases followed by BAL 8 (5.1%) cases and pleural fluid 3 (1.9%) cases out of total cases. No malignancies were detected on sputum examination. The rate of diagnosis of “negative for malignancy” was highest in pleural fluid sample 81 (52.2%) cases followed by BAL and sputum 5 (3.3%) cases each.

Table 1: Distribution of cytological diagnosis in different specimen (N=155).

Cytological Diagnosis	FNAC (%)	BAL (%)	Pleural fluid (%)	Sputum (%)	Total (%)
Positive for Malignancy	24 (68.5)	7 (20)	4 (11.5)	00	35 (22.6)
Suspicious for malignancy	2 (22.2)	6 (66.7)	1 (11.1)	00	9 (5.8)
Benign infectious lesion	7 (53.8)	00	5 (38.5)	1 (7.7)	13 (8.5)
Negative for malignancy	3 (3.2)	5 (5.3)	81 (86.2)	5 (5.3)	94 (60.6)
Inadequate for opinion	4 (100)	00	00	00	4 (2.5)
Total (%)	40 (25.8)	18 (11.6)	91 (58.8)	6 (3.8)	155 (100)

Biopsies for histopathological correlation was available in total 33 (21%) cases. Malignancy was the most common diagnosis on biopsy, 23 (69.7%) cases comprising of 10 (30.3%) each case of adenocarcinoma and SCC followed by 3 (9.1%) cases of SCLC. Low-grade dysplasia, solitary fibrous tumor (SFT) and mucormycosis was 1 (3%) case each. 4 (12.2%) cases were inadequate for providing a definite opinion.

Table 2: Correlation between cytological and histopathological diagnosis in all cases (N=155).

Cytological Diagnosis	Histopathological diagnosis									Total (%)
	Adenocarcinoma (%)	SCC (%)	SCLC (%)	Low-grade squamous dysplasia (%)	Benign infectious lesion (%)	Solitary fibrous tumor (%)	Normal histology (%)	Inadequate for opinion (%)	NA (%)	
Adenocarcinoma	7 (4.5)	-	-	-	-	-	-	-	8 (5.16)	15 (9.6)
SCC	-	9 (5.8)	-	-	-	-	-	1 (0.6)	2 (1.2)	12 (7.7)
SCLC	-	-	2 (1.2)	-	-	-	-	-	1 (0.6)	3 (2.0)
NSCLC	-	-	-	-	-	-	-	-	1 (0.6)	1 (0.6)
Metastatic malignancy	-	-	-	-	-	-	-	-	4 (2.6)	4 (2.6)
Suspicious for malignancy	1 (0.6)	1 (0.6)	1 (0.6)	-	-	-	1 (0.6)	1 (0.6)	4 (2.6)	9 (5.9)
Benign infectious lesion	-	-	-	-	1 (0.6)	-	-	-	12 (7.8)	13 (8.4)
Negative for malignancy	2 (1.2)	-	-	1 (0.6)	-	-	2 (1.2)	1 (0.6)	88 (56.7)	94 (60.6)
Inadequate for opinion	-	-	-	-	-	1 (0.6)	-	1 (0.6)	2 (1.2)	4 (2.6)
Total	10 (6.5)	10 (6.5)	3 (2)	1 (0.6)	1 (0.6)	1 (0.6)	3 (2)	4 (2.5)	122 (78.7)	155 (100)

SCC: Squamous cell carcinoma, SCLC: Small cell lung carcinoma, NSCLC: Non-small cell lung carcinoma, NA: Not available

A total of 85.71% cases on histopathological evaluation corroborated with their cytological diagnosis. 18 (11.7%) cases of carcinoma diagnosed on cytology had same diagnosis on histopathology. However, out of 3 (1.9%) cases diagnosed as benign on cytology, 2 (1.2%) cases were adenocarcinoma and 1 (0.6%) case was low-grade dysplasia on histopathology. The highest yield of a true positive diagnosis was given by FNAC followed by BAL. One case was false positive in BAL. FNAC, BAL and pleural fluid yielded in 1 each false negative diagnosis. The cytology smears in all three cases comprised of benign endobronchial cells and squamous cells admixed with mixed inflammatory cells and carbon laden macrophages.

The overall diagnostic accuracy of cytology excluding the inadequate samples was 85.6%. This study showed that the sensitivity of the cytological investigations was 88% while the specificity was 66.7%. The PPV of cases from cytological studies was 95.6% while the NPV was 40%.

Discussion

Lung cancer is the leading cause of cancer-related fatalities. Timely detection and treatment are crucial to reducing mortality.¹ The male to female ratio of this study was 1.3:1 while the age range was 3 to 98 years with the

mean age was 60.9 ± 1.32 SD years which is comparable to the study done by Shrestha et al.⁶ and Pavani et al.⁷

In the present study, the most common primary malignancy seen among the active smokers was SCLC whereas in past smokers it was SCC. In patients who consumed alcohol only, adenocarcinoma was the most common malignancy. Cases diagnosed as metastatic malignancy and suspicious for malignancy were either past smokers or alcoholic or both.

Dosemeci et al. on their study on effect of tobaccos and alcohol use in development of lung cancer noted that smokers displayed notably elevated risks across all cell types. The highest risk was witnessed in cases of SCLC. Additionally, SCLC exhibited the most pronounced trend of increased risk in correlation with quantitative gauges of tobacco consumption.⁸

Pleural fluid was the most received sample in our study as was the finding in a study by Ghildiyal et al.⁹

We distributed the cytological diagnosis into five categories: positive for malignancy 35 (22.6%) cases, suspicious for malignancy 9 (5.8%) cases, benign infectious lesion 13 (8.4%) cases, negative for malignancy 94 (60.6%) cases and inadequate for opinion 4 (2.6%) cases.

Table 3: Comparison of cytological diagnosis amid different studies.

Diagnostic categories	Present study (%)	Shrestha et al. ⁶ (%)	Chandra et al. ¹⁰ (%)	Ghildiyal et al. ⁹ (%)	Dzambas et al. ¹¹ (%)
Positive for malignancy	22.6	81	52	16.2	98.2
Suspicious for malignancy	5.8	-	10.2	3.8	1.8
Benign infectious lesion	8.4	5.1	6.5	5.2	-
Negative for malignancy	60.6	9.3	22.7	59.7	-
Inadequate for opinion	2.6	4.6	8.3	14.9	-

Out of the total 35 cases diagnosed as positive for malignancy in the present study, the most common primary malignancy was found to be adenocarcinoma comprising of 15 (42.8%) followed by SCC 12 (34.2%), SCLC 3 (8.2%) and non-small cell lung carcinoma (NSCLC) 1 (3%) case. Four (11.5%) cases were of metastatic malignancy. Studies by Shrestha et al.⁶ and Pavani et al.⁷ indicates that adenocarcinoma is the most common primary carcinoma of lung which is consistent with the findings of present study. Conversely, SCC was more common than adenocarcinoma in studies by Raiza et al.¹ and Chandra et al.¹⁰ Previously, SCC was considered as the most common primary lung malignancy followed by adenocarcinoma. However, with reference to the present study and various other studies, a changing trend in incidence of subtype of lung cancer is noted worldwide and adenocarcinoma has replaced squamous cell carcinoma as the commonest malignancy. With the advent of targeted therapy, the need for subtyping

non-small cell carcinomas into adenocarcinoma, SCC or other type is crucial in current scenario. Our study showed that the most common malignancy among all study population was adenocarcinoma. In a study by Piplani et al.¹² adenocarcinoma was the most common primary lung lesion in females whereas SCC was slightly more common than adenocarcinoma in males. Sex-based studies done by Sagerup et al.¹³ Stabellini et al.¹⁴ and Rodriguez et al.¹⁵ suggests adenocarcinoma as the most common malignancy in both sexes. This finding was contradictory from our study where females mostly had SCC.

These studies emphasized that there is a biological difference between the same lung carcinomas among two different genders. Females were usually diagnosed at an earlier stage as compared to men, and have a less advanced disease on presentation. There are differences in the carcinogenic effects of tobacco among men and women, and men are more susceptible to develop lung

malignancies. Thus, treatment protocols should stratify gender-specific clinical trials due to the differences in prognostic and predictive factors.¹²⁻¹⁵

In the present study, 13 cases were categorized as infectious lesions. Abscess was the most predominant lesion 6 (46.1%), which was also seen in the studies by Tuladhar et al.¹⁶ and Hanson et al.¹⁷ Various studies show that tuberculosis and fungal infections encompass major bulk of infectious lesions of lung which is similar to the findings of present study with tuberculosis comprising 4 (30.7%) cases and 2 cases of fungal infections comprising of 1 (7.7%) case of aspergilloma and mucormycosis each.^{1,10,18}

Smears from four (2.6%) cases of FNAC were inadequate to

confer a definite diagnosis in cytology. The smears in these cases were of scant cellularity predominantly comprising of neutrophils and some benign fibrofatty fragments against a hemorrhagic background. The inadequacy rate is remarkably low in our study in comparison to study by Ghildiyal et al.⁹ (14.9%), Chandra et al.¹⁰ (8.3%), Raiza et al.¹ (5.3%) and Piplani et al.¹² (4%). The reason for inadequacy could be attributed to the factors such as difficulty in localization of lesions, high vascularity of lesions or faulty techniques. The rate of inadequacy can be minimized by application of correct techniques, training of manpower and application of rapid on-site evaluation (ROSE), which is a simple yet cost-effective technique for on-site evaluation of adequacy of sample.

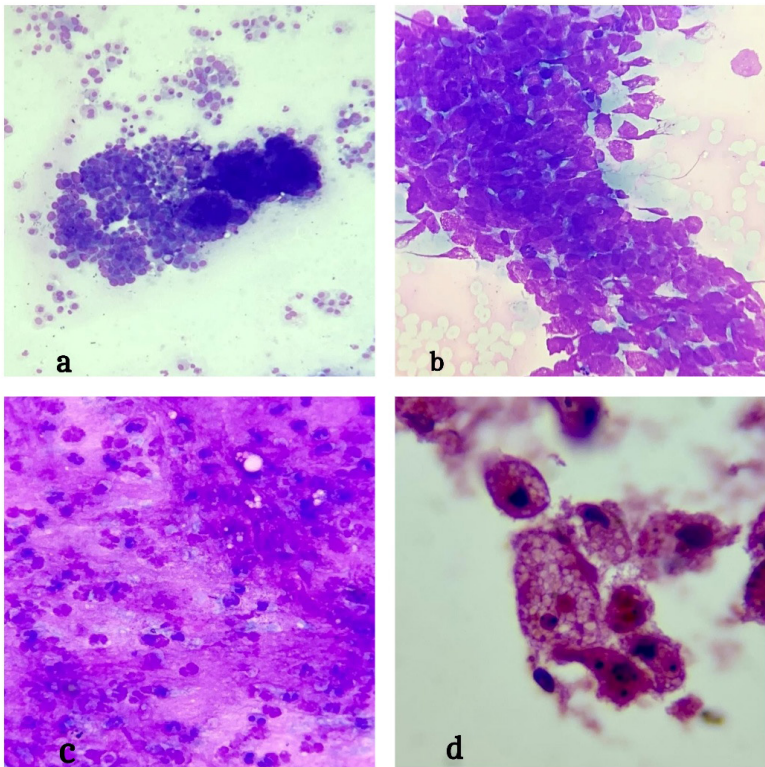


Figure 3: Photomicrograph of different lung lesions. a. FNAC smear from adenocarcinoma showing tumor cells arranged in clusters and papillaroid pattern with high N:C ratio and scant cytoplasm (Leishman, 10X), b. FNAC smear from small cell lung carcinoma showing tumor cells with scant cytoplasm, stippled chromatin, nuclear molding and streaking artifact (Leishman, 10X), c. FNAC smear from acute suppurative inflammation showing sheets of neutrophils and degenerated cells (Leishman, 40X), d. Histopathological section from mucormycosis showing macrophages engulfing fungus (PAS, 100X)

Table 4: Statistical analysis of cytopathology in different studies.

Parameters	Present study	Dzambas et al. ¹¹	Ghildiyal et al. ⁹	Jayaraj et al. ¹⁹	Sarma et al. ²⁰	Pavani et al. ⁷	Ahmad et al. ³
Sensitivity (%)	88	94.9	81.4	93.9	89.9	93.7	80.5
Specificity (%)	66.6	98.6	93.3	57.1	90.9	83.3	96.6
PPV (%)	95.6	95.7	93.6	91.1	96.3	95.2	97
NPV (%)	40	98.3	80.7	66.6	78.4	-	-
Concordance (%)	85.6	61.4	86.8	87.5	90	-	-

Among all the cytology specimen evaluated in the current study, only 33 cases had biopsy samples available for histopathological correlation. Overall, a concordant diagnosis among cytology and histopathology was obtained in 24 samples whereas discordant diagnosis was seen in 4 cases.

The sensitivity of FNAC was 92.8% which was higher than BAL (90%). The PPV was also higher for FNAC (100%) than BAL (90%). The specificity and NPV of BAL were both 66.6%.

We observed variation in the accuracy of our specimen as compared to the other studies, the major factor for which could be the low number of biopsy samples available for histopathological correlation. The present study had a sensitivity of 88% (95% CI 68.8-97.5) which is comparable to the study by Dzambas et al.¹¹ Jayaraj et al.¹⁹ Sarma et al.²⁰ and Pavani et al.⁷ The sensitivity rate was higher in present study as compared to other studies by Ghildiyal et al.⁹ and Ahmad et al.³ The specificity was 66.6% (95% CI 9.4-99.2) in the present study which was lesser than the Dzambas et al.¹¹ Ghildiyal et al.⁹ Sarma et al.²⁰ Pavani et al.⁷ and Ahmad et al.³ However, it was higher than Jararaj et al.¹⁹ The PPV in our study was fairly high i.e. 95.6% (95% CI 78.1-99.9) and comparable to all other studies. The NPV value was however, low in the present study, 40% (95% CI 5.3-85.3) as compared to other studies.^{3,7,8,11,19,20} The overall concordance between cytology and histology was 85.6% in the present study which is comparable to the other studies.^{9,19,20} This was remarkably higher than the rate of concordance seen in Dzambas et al.¹¹

Limitations

Confidence intervals in this study are wide due to the small amount of histopathology sample available, resulting in imprecise specificity and negative predictive value estimates. Similar study can be carried out in future including larger sample size. Use of immunohistochemistry and molecular cytogenetic evaluation can further solidify the cytological and histopathological diagnosis and aid in targeted therapy for malignant lung lesions.

Conclusion

The findings from the present study show that FNAC technique has higher sensitivity and PPV as compared to BAL. Adenocarcinoma is more common than other histological subtypes. Infectious lesions such as lung abscess, tuberculosis and fungal infections of lung can also be diagnosed via the help of cytology. Cytological techniques are cheap, rapid and effective which can be helpful for clinicians in the decision-making process, especially in developing countries like Nepal. Despite the constraints inherent in cytological procedures, they

have the potential to detect non-neoplastic lesions and lung malignancies in their earliest phases, prior to the emergence of visible or palpable tumors.

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