

Role of FNAC in Diagnosis of Malignant Lymphadenopathy: A Five-year Cross-sectional Study at a Tertiary Care Centre, Mumbai, India

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ABSTRACT

Introduction: Fine Needle Aspiration Cytology (FNAC) of the lymph node is a simple tool in the diagnosis of suspected and unsuspected primary and secondary lymph node malignancy. It is a useful first-line investigation to distinguish between infective and malignant lymphadenopathy, especially in a low-resource setting like our country.

Aim: To study the utility of FNAC in the diagnosis of lymph node malignancy and to evaluate the efficacy of cytology in diagnosing the primary site of malignancy in case of metastasis.

Materials and Methods: A cross-sectional retrospective and prospective study was conducted for a five-year period from July 2015 to June 2020, in the pathology department of Dr. RN Cooper Municipal General Hospital, Mumbai, Maharashtra, India. A total of 112 patients diagnosed with malignancy on FNAC of lymph nodes were studied. The FNAC procedure was performed by cytopathologists using a 23/24G needle attached to a 10 ml syringe. The alcohol-fixed smears were stained with Haematoxylin & Eosin (H&E) stain and Papanicolaou (PAP) stains, while the air-dried smears were stained with Giemsa stain. Diagnosis was based on cytomorphological features and clinical presentation.

Results: A total of 112 patients were diagnosed with malignancy on FNAC, of which 90 were males and 22 were females, with a male-to-female ratio of 4:1. Cytological analysis of these nodes revealed metastasis in 98 cases (87.50%) and lymphoma in 14 cases (12.50%). The maximum number of patients were in the 51-60 years age group, with 38 cases (33.92%). The most common site of involvement was cervical lymph nodes, with 75 cases (66.96%). The most common cytological diagnosis was metastatic squamous cell carcinoma in 65 cases (58.02%), followed by metastatic un-differentiated carcinoma in 26 cases (23.20%).

Conclusion: Fine Needle Aspiration (FNA) is a simple, safe, accurate, cost-effective, and valuable tool in the evaluation of malignant lymphadenopathy. Malignant diseases were easily diagnosed by this simple diagnostic procedure. In the case of a diagnosis of lymphoma, surgery can be completely avoided. It helps in planning further surgical management for metastatic disease, where definitive operative intervention can be performed in one session. In the case of an un-detected primary tumour, FNAC directs further investigations towards the possible primary site.

Keywords: Fine needle aspiration cytology, Lymphoma, Malignancy

INTRODUCTION

The FNAC is a rapid, inexpensive, and safe procedure that can be done at the time of the patient's first presentation and immediately after regional physical examination. FNAC has been extensively utilised as a primary diagnostic tool to examine enlarged lymph nodes and to exclude involvement of alternative organs, such as the salivary gland, head, neck, or other subcutaneous masses. It is a minimally invasive approach that allows fast diagnosis and treatment [1]. DeMay RM has summarised the advantages of FNAC with the acronym "SAFE," which means Simple, Accurate, Fast, And Economical [2]. Enlarged lymph nodes are easily accessible for FNAC, and hence it forms an important diagnostic tool in the diagnosis of lymph node lesions. FNAC not only confirms the presence of metastatic disease but also gives a clue regarding the nature and origin of the primary tumour [3]. It also gives an idea about the prognosis and management of patients for staging purposes. FNAC avoids the physical and psychological trauma occasionally encountered after an open surgical biopsy, is convenient for the patient and physician alike, and is a useful outpatient procedure [4]. FNAC is also useful in the detection of recurrent malignancy.

This study was conducted to determine the utility of FNAC in diagnosing malignant lymphadenopathy and also to differentiate between lymphoma and metastasis. In cases with a known primary, cytological examination confirms the presence of metastasis, which

helps in planning further surgery. In cases with an unknown primary, correlation of FNAC findings with clinical and radiological details in these cases can help to detect its site. FNAC along with ancillary techniques like immunocytochemistry and cell block preparations can help in identifying the primary metastasis. [5]. Though previous similar studies have been done in the past [3,5], the present study included a large sample size and reflected the common primaries in the Mumbai region. Moreover, cases of both lymphoma and metastasis were included.

Hence, the study was conducted to understand relative frequency and different cytomorphological features of malignancy in lymph node FNAC and to assess the cytomorphological features in metastasis to diagnose the primary site of malignancy.

MATERIALS AND METHODS

A five year retrospective and prospective cross-sectional study was conducted on malignant lymph node aspirates in the Department of Pathology of Dr. RN Cooper Municipal General Hospital from July 2015 to June 2020. The patients were referred from various clinical departments of Dr. RN Cooper Municipal General Hospital and other attached peripheral hospitals.

The study was approved by the Institutional Ethical Committee (IEC) with ethical clearance NHBTMC/IEC/03 dated 14/12/2019. A total of 5919 FNACs done in the five-year period, with 2452 (41.42%)

performed on lymph nodes. A total of 112 patients, 63 retrospective and 49 prospective, who presented with lymph node enlargement and were diagnosed with malignancy on FNAC, were studied. Patients with lymphadenopathy due to causes except malignancy were excluded from the study.

Procedure

In every case, demographic data, brief history, physical examination, along with evaluation of relevant investigations, if available, was carried out. In retrospective cases, cytology slides were retrieved from archival collections and reviewed, while clinical data was retrieved from the medical record department. For prospective cases, after obtaining informed consent, cytopathologists performed the FNAC procedure using a 23/24G needle attached to a 10 ml syringe, taking aseptic precautions. Multiple sites were aspirated, and the material was smeared onto slides. The smears were stained with H&E, PAP, and Giemsa stains and evaluated by a cytopathologist. Diagnosis was based on cytomorphological features and clinic pathological concordance, with noted concordance between cytological and histopathological diagnosis where possible.

STATISTICAL ANALYSIS

Statistical analysis was conducted using Statistical Package for Social Sciences (SPSS) IBM version 21.0, with data entry checks was done at regular intervals to ensure valid entries. Results were expressed as frequency and percentage, thus not requiring further statistical analysis.

RESULTS

The results showed that out of 112 (4.56%) lymph node aspirates that were malignant on cytology, 14 cases (12.50%) were lymphoma and 98 cases (87.50%) were metastasis. [Table/Fig-1] demonstrates the age and gender distribution of the cases studied. Patient ages ranging from 8 to 87 years. The maximum number of cases (33.93%) was in the 51-60 years age group, followed by the 41-50 years. The male population was more commonly affected, with 90 cases (80.36%) compared to 22 female cases (19.64%), with the male-to-female ratio of 4:1.

Age (years)	Number of patients (%)	
	Male	Female
0-10 years	01 (00.90%)	01 (00.90%)
11-20 years	02 (1.80%)	01 (0.90%)
21-30 years	05 (4.46%)	01(0.90%)
31-40 years	08 (7.14%)	02(1.80%)
41-50 years	22 (19.64%)	03 (2.68%)
51-60 years	30 (26.79%)	08 (7.14%)
61 years and above	22 (19.64%)	06 (5.36%)
Total	90 (80.36%)	22 (19.64%)

[Table/Fig-1]: Age and sex distribution of patients with malignant lymphadenopathy.

Lymphoma cases were most prevalent in the third decade (21.43%), while metastatic lymphadenopathy was commonly in the sixth decade (36.73%). There were 14 lymphoma cases, with 9 males (64.29%) and 5 females (35.71%), and 98 patients with metastatic lymphadenopathy, of which 81 (82.65%) were males and 17 (17.35%) were females.

The most common symptom was weight loss, observed in 79 cases (70.53%), followed by generalised weakness in 36 cases (32.14%). Other symptoms included fever in 29 cases (25.89%), dysphagia in 27 cases (24.10%), palpable liver in 25 cases (22.32%), splenomegaly in 5 cases (4.46%), skin rash in 7 cases (6.25%), and night sweats in 10 cases (8.92%).

[Table/Fig-2] displays the various sites of lymph nodes aspirated. The most commonly involved lymph nodes were in the cervical group, with 75 cases (66.96%). The majority of lymphoma lymph

Site of FNAC	Number of patients (N)		N (%)
	Lymphoma N	Metastasis N	
Cervical group lymph nodes	07	68	75 (66.96%)
Supraclavicular lymph node	01	18	19 (16.96%)
Axillary lymph nodes	05	08	13 (11.60%)
Inguinal lymph nodes	00	02	02 (01.78%)
Parotid lymph nodes	00	02	02 (01.78%)
Mediastinal lymph nodes	01	00	01 (00.92%)
Total	14	98	112 (100%)

[Table/Fig-2]: Different sites of primary and secondary lymph node malignancy.

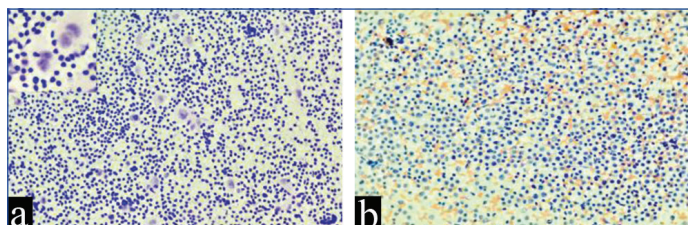
nodes with 75(66.96%) cases. Maximum cases of Lymphoma showed involvement of cervical lymph nodes, with seven cases (50%), followed by axillary nodes with five cases (35.71%). Out of the 98 cases of metastatic lymphadenopathy, 68 cases (69.39%) were detected in cervical lymph nodes, followed by 18 cases (18.39%) in supraclavicular lymph nodes.

[Table/Fig-3] presents the cytological diagnosis of all cases. There were 10 cases (8.93%) of Hodgkin's lymphoma and four cases (3.57%) of Non-Hodgkin's lymphoma. In the remaining, 98 cases (87.5%) of metastatic lymphadenopathy, the most common finding was metastasis of squamous cell carcinoma (65 cases), followed by metastasis of undifferentiated carcinoma (26 cases).

Cytological diagnosis	N (%)
Primary (N=14)	
1) Hodgkin's lymphoma	10 (08.93%)
2) Non-Hodgkin's lymphoma	4 (03.57%)
Secondary (N=98)	
1) Squamous cell carcinoma	65 (58.02%)
2) Undifferentiated carcinoma	26 (23.20%)
3) Melanoma	02 (01.78%)
4) Small cell carcinoma lung	01 (00.90%)
5) Small round cell tumour	01 (00.90%)
6) Carcinoma breast	01 (00.90%)
7) Papillary carcinoma thyroid	01 (00.90%)
8) High-grade papillary carcinoma	01 (00.90%)
Total	112 (100%)

[Table/Fig-3]: Cytological diagnosis in malignant lymphadenopathy.

In the 10 cases of Hodgkin lymphoma, classical Reed-Sternberg cells were present in four cases, while mononuclear Hodgkin's cells were seen in eight cases, and one case additionally showed ill-formed epithelioid cell granulomas [Table/ Fig-4a]. In the four cases of Non-Hodgkin's lymphoma, a monotonous population of small-to-medium-sized lymphoid cells was seen [Table/Fig-4b].



[Table/Fig-4]: (a) Hodgkin's Lymphoma: Classical Reed-Sternberg cell with moderate amount of cytoplasm and nucleus showing large prominent nucleoli along with many mononuclear Hodgkin cells with mixed population of reactive lymphoid cells. Inset shows RS cell with prominent nucleoli (H&E, 40X); (b) Non-Hodgkin's Lymphoma: Discrete small and large lymphoid cells showing scant cytoplasm, condensed nuclear chromatin and inconspicuous nucleoli (PAP, 40X).

Out of the total 112 cases, biopsies were performed in all 14 cases of lymphoma, with 100% concordance between cytology and histopathology. In 98 out of 112 cases of metastatic diseases, biopsies were performed at the primary site in 54 cases, with cytohistopathological concordance found in 45 out of 54 cases

(83.33%). In nine discordant cases of undifferentiated carcinoma, further typing could not be done due to high-grade malignancy and lack of IHC on both cytology and histopathology. In 44 out of 98 cases, due to multiple metastases and advanced disease on radiology, the primary site could not be identified, and biopsies were not performed in those cases.

[Table/Fig-5] shows the primary site of malignancy in the metastatic nodes. In the category of metastatic squamous cell carcinoma with 65 cases, the primary site was the oral cavity in 18 cases (27.70%), followed by the larynx in 11 cases (16.92%), oesophagus in 8 cases (12.30%), and oropharynx in 2 cases (3.08%). In 26 cases, the diagnosis of metastatic squamous cell carcinoma was primarily made through FNAC, as patients presented with lymph node enlargement without clinical suspicion of malignancy.

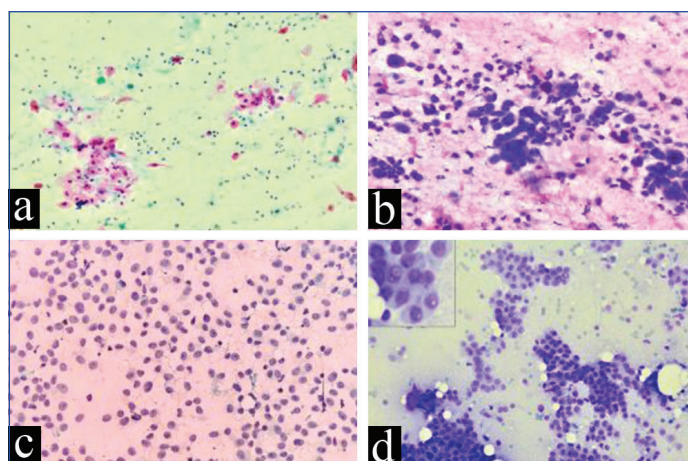
Cytological diagnosis	Lymph node aspirated	Primary site
Lymphoma (14 cases)	Cervical LN (07) Axillary LN (05) Supraclavicular LN (01) Mediastinal LN (01)	Lymph node (14)
Metastasis of squamous cell carcinoma (65 cases)	Cervical LN (49) Supraclavicular LN (08) Axillary LN (08)	Oral cavity (18) Larynx (11) Oesophagus (8) Oropharynx (2) Unknown (26)
Metastasis of undifferentiated carcinoma (26 cases)	Cervical LN (17) Supraclavicular LN (07) Parotid LN (02)	Nasopharynx (06) Oral cavity (02) Lungs (1) Unknown (17)
Metastasis of melanoma (2 cases)	Inguinal LN (02)	Skin (02)
Metastasis of small cell lung carcinoma (1 case)	Supraclavicular LN (01)	Lung (01)
Metastasis of small round cell tumour (1 case)	Cervical LN (01)	Nasopharynx (01)
Metastasis of carcinoma breast (1 case)	Supraclavicular LN (01)	Breast (01)
Metastasis of papillary thyroid carcinoma (1 case)	Cervical LN (01)	Thyroid (01)
Metastasis of high-grade papillary carcinoma (1 case)	Supraclavicular LN (01)	Unknown (01)

[Table/Fig-5]: Cytological diagnosis in malignant lymphadenopathy.

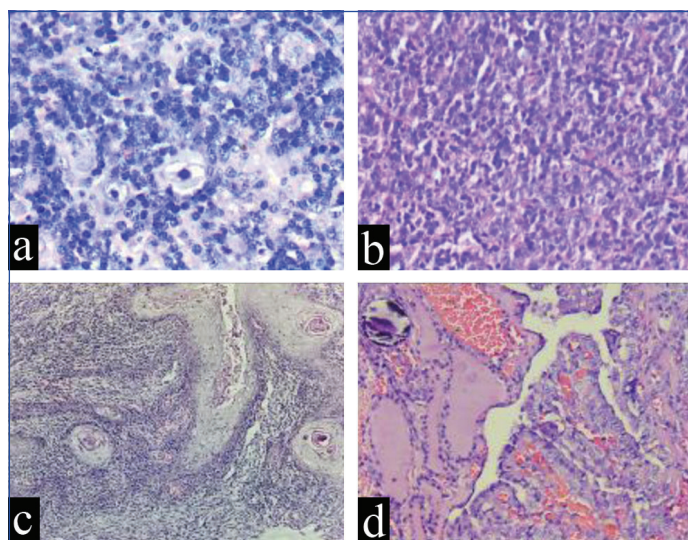
All cases of SCC showed malignant squamous cells with varied morphology, and 48 cases showed background necrosis and nuclear debris [Table/Fig-6a]. In cases of metastatic undifferentiated carcinoma, the primary site was the nasopharynx in six cases (23.08%), oral cavity in two cases (7.69%), and lung in one case (3.85%) [Table/Fig-6b]. The primary site remained unknown in 17 cases (65.35%). Metastatic melanoma was seen in two cases of inguinal lymph nodes, with the primary site being the toe in one case and the anal canal in another [Table/Fig-6c]. [Table/Fig-6d] shows one case of metastasis of papillary thyroid carcinoma with intranuclear pseudo inclusion. [Table/Fig-7] displays histopathological images of lymphoma and metastatic SCC and papillary thyroid carcinoma.

DISCUSSION

Enlarged lymph nodes are accessible for FNAC and are important in diagnosing the underlying disease. FNAC is a diagnostic tool that is cost-effective, simple, and minimally invasive. It can provide clues in the presence of an occult primary and sometimes surprise clinicians who do not suspect malignancy [6]. FNAC has become a well-established method for diagnosing metastatic malignancies in lymph nodes [7]. The diagnosis based on cytological material is accepted and sometimes there is no further correlation with histopathology is not always necessary, especially in cases of advanced malignancies [3]. [Table/Fig-8] compares the present study to other studies [3,5-12].



[Table/Fig-6]: a) Metastasis of squamous cell carcinoma: many discrete and clusters of malignant squamous cells along with necrosis and polymorphs (PAP, 40X); b) Metastasis of undifferentiated carcinoma: cellular smear showing highly pleomorphic cells in syncytial clusters with singly scattered cells in background. The nuclear membrane is irregular with coarse nuclear chromatin (H&E, 40X); c) Metastasis of melanoma: cells with markedly pleomorphic nuclei having prominent eosinophilic macronucleoli (PAP, 40X); d) Metastasis of papillary thyroid carcinoma: papillary clusters and dissociated cells showing pale nuclear chromatin. Inset shows prominent intranuclear pseudo inclusion. (Giemsa, 40X).



[Table/Fig-7]: Shows histopathological images for: a) Hodgkin lymphoma with presence of inflammatory background with Hodgkin's cells; b) Non-Hodgkin's lymphoma shows monomorphic population of lymphoma cells; c) Metastasis of squamous cell carcinoma shows island of atypical squamous cells with keratin pearl formation; and d) Metastasis papillary thyroid carcinoma shows presence of true papilla with ground glass appearance of nuclei.

Malignancies in lymph nodes are predominantly metastatic, with an incidence varying from 65.7% to 80.4%, while lymphomas range from 2% to 15.3% of lymph nodes aspirated from all sites [3]. In this study, lymph node involvement by metastasis (87.50%) was more common compared to primary lymphoid neoplasms (12.50%). This aligns with findings from other studies as shown in [Table/Fig-8] [3,5,7]. In the present study, 4.50% yielded malignant diagnosis, which is significantly lower than in other studies where the incidence ranged from 4.8% [8] to 69.74% [7]. This difference may be because our institute being a general hospital serving a diverse population where infectious aetiology is predominant.

In the present study, males were predominantly affected with 90 cases (80.36%), compared to females with 22 cases (19.64%), resulting in a male-to-female ratio of 4:1. A male preponderance was also noted by Rathod G and Singla D (3.2:1), Arora S et al., (3.5:1), and Mehdi G et al., (3.1:1) [6,9,10]. The most common age group in the present study was 51-60 years, similar to findings by Chakravarty-Vartak US et al., Mehdi G et al., and Meena P and Mishra RT [8,10,11].

The most common symptom in patients with malignant lymphadenopathy was weight loss, seen in 79 cases (70.53%).

Author	Frequency of malignant lymph nodes	Male to female ratio	Age predominance (years)	Number of primary and secondary malignancy	Common site of lymphadenopathy	Common site of primary malignancy	Common type of malignancy
Wilkinson AR et al., [3] 2012	15.4%	1.5:1	-	Metastatic- 90% lymphomas-10%	Cervical (62 %)	Oral cavity	SCC (46%)
Gupta C et al., [5] 2023	-	1.94:1	41-60	Metastatic- (80.4%) lymphomas- (19.6%)	Cervical (62.8%) followed by supraclavicular (13.4%)	Oral cavity	SSC (43.3%) followed by Adenocarcinoma (14.4%)
Rathod G and Singla D [6] 2015	20.46 %	3.2:1	-	-	Cervical (77.5%) followed by supraclavicular (12.5%)	Oral cavity	SSC (72.5%)
Arundhati JP [7] 2017	69.74 %	1:1.2	40-49	Metastatic- (95.89%) lymphomas- (04.10%)	Cervical triangles followed by supraclavicular	primary carcinomas in breast, lung, stomach,	Adenocarcinoma (42.05%) followed by SCC (27.69%)
Chakravarty-Vartak US et al., [8] 2016	4.8	1.8:1	45-54	-	Submandibular lymph nodes (33.6%)	Oral cavity followed by larynx, hypopharynx	SCC (66.3%),
Arora S et al., [9] 2018	18.37%	3.5:1	-	Metastatic- (88.46%) lymphomas- (11.54%)	Cervical	-	SSC (53.85%) followed by poorly differentiated epithelial carcinoma in (23.08%) cases.
Mehdi G et al., [10] 2015	-	3.1:1	51-60	-	Cervical (69.2%) followed by axillary (13.1%)	Tongue and tonsil (27 and 15 cases respectively), followed by larynx (15 cases).	SCC (26.68), followed by adenocarcinoma (9.82),
Meena P and Mishra RT [11] 2017	24.25	2.6:1	51-60	-	Cervical (75%) followed by (15%) supraclavicular	Aero digestive tract (60%)	SSC (76%)
Bosch X et al., [13] 2013	27.4	2.4:1	41-60	-	Cervical (55.56%)	Oral cavity, pharynx and larynx (27.77%)	SSC (43%).
Present study	04.50%	4:1.	51-60	Metastatic- (87.50%) lymphomas- (12.50%)	Cervical (66.96%) followed by Supraclavicular (16.96%)	Oral cavity (18 cases), followed by larynx (11 cases),	SSC (58.02%) followed by metastasis of Undifferentiated carcinoma (23.20%).

[Table/Fig-8]: Different studies and their comparative findings with the present study [3,5-11,13].

Bosch X et al., noted that the most common clinical symptom was a palpable liver in 88 (24%) cases, followed by weight loss in 86 (23%) patients in their study [13]. In the present study, hepatomegaly was seen in only 25 cases (22.3%). This may be because Bosch X et al., had more cases of lymphoma, hence hepatomegaly is a common finding in their study [13]. The cervical group was the most common group of lymph nodes involved, followed by supraclavicular nodes. This was similar to the findings of Gupta C et al., Rathod G and Singla D; Arundhati JP, and Meena P and Mishra RT [5-7,11]. The oral cavity was the most common primary site of malignancy in the present study, and similar findings were noted by the other authors [3,5,6,8,10-12].

The cervical group of lymph nodes was also the most common site of FNAC in lymphomas with 7/14 cases (50%), followed by the axillary group of lymph nodes, i.e., 5/14 cases (35.71%). This may be explained by the large number of cases with metastatic head and neck malignancies and the easy accessibility of cervical nodes for examination and evaluation. The most common cytological diagnosis in the present study was metastasis of squamous cell carcinoma, reported in 65 cases (58.02%), which was similar to findings in other studies [3,5,6,8-11,12]. The second most common diagnosis was metastasis of undifferentiated carcinoma in 26 cases (23.20%), and similar findings were noted by Arora S et al., [9]. Lymphomas were found in 14 cases (12.5%), with 10 cases (8.93%) of Hodgkin's lymphoma and four cases (3.57%) of Non-Hodgkin's lymphoma, which was similar to the findings noted by Martins MR and Santos GD, who reported 14.2% lymphomas, of which 8.6% were Hodgkin's lymphoma and 3.7% were Non-Hodgkin's lymphoma [14].

Other studies by Wilkinson AR et al., and Arora S et al., have reported a higher proportion of Non-Hodgkin's Lymphoma in their studies [3,9]. The most common primary site for squamous cell carcinoma and undifferentiated carcinoma was the upper aerodigestive tract in 48/112 cases (42.86%) with involvement of the oral cavity in (17.85%), larynx in (9.8%), and pharynx in (8.1%) patients. Similar findings were noted by Wilkinson AR et al., Gupta C et al., Rathod G and Singla D; Meena P and Mishra

RT; Sheikh S and Parmar JK; [3,5,6,11,12]. In 44 cases (39.29%), patients presented primarily with lymphadenopathy and were diagnosed to have metastatic disease from an unknown primary on FNAC. Out of these, there were 26 cases (23.2%) of metastatic squamous cell carcinoma, 17 cases (15.1%) of undifferentiated carcinoma, and a single case of metastatic high-grade papillary carcinoma. Similar findings were noted by Mehdi G et al., who reported 45.1% of cases with an unknown primary [10]. Even after thorough history taking and examination, the primary site could not be found. Most of these patients were referred to higher oncology centers and were lost to follow-up. In these cases, the cervical lymph node was most commonly affected, and the most common cytological diagnosis in this category was SCC and poorly differentiated carcinoma, which was similar to findings by Mehdi G et al., [10].

In the present study, two cases were labeled as metastasis of melanoma to the inguinal lymph nodes; the primary site was found to be in the foot and anal canal, respectively. Kaur A et al., studied 39 cases of malignant melanoma with 32 cases of metastatic melanoma [15]. Hence, metastatic melanoma is detected easily on FNAC, especially due to the presence of pigment. In the present study, although metastasis could be detected in 98 cases, the primary site could only be found in 54 out of 98 cases, while 44 cases presented as an unknown primary. The use of immunocytochemistry can further help in the detection of the primary in these cases.

Limitation(s)

The diagnosis of metastasis or lymphoma was given on FNAC; however, immunohistochemistry was not done at our institute, and cases were referred to higher centers for further work-up.

CONCLUSION(S)

The FNAC is a simple tool to distinguish between lymphoma and metastasis. The present study included a large sample size and reflected the common types of primary malignancies in the Mumbai region. The cytomorphological features can help identify the primary site in metastasis and detect the presence of lymphomas. FNAC

of malignant lymphadenopathy is an important diagnostic tool in a poor resource setting, where most patients present in the advanced stage and cannot afford expensive investigations.

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