

# Clinicopathological Analysis of Mediastinal Masses: A Mixed Bag of Non-Neoplastic and Neoplastic Etiologies

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## ABSTRACT

**Objective:** The mediastinum is the central portion of the thoracic cavity, housing numerous organs and harbouring a mixed bag of non-neoplastic and neoplastic lesions. Accurate diagnosis is essential owing to the widely variable therapeutic and prognostic implications.

**Material and Method:** Cases of mediastinal masses were retrospectively reviewed from January 2011 till January 2016. Clinico-radiological records of these cases were retrieved. Fine needle aspiration cytology (FNAC) was performed wherever feasible. Histopathological and immunohistochemical evaluation of the excised specimens was undertaken.

**Results:** Of the 60 cases included in our study, 22 were anterior, 20 were middle and 18 posterior mediastinal masses. The majority of the patients were symptomatic (96.8%). The most common pathology was thymoma (12 cases) followed by ten cases of lymphoma, eight cases each of tubercular lymphadenopathy and schwannoma, six cases of neurofibroma, four cases of extragonadal germ cell tumours, two cases each of thymic cyst, bronchogenic cyst, retrosternal goitre, ganglioneuroma and neuroblastoma, and one case each of lipoma and thymolipoma. FNAC was done in 54 cases of which 7 cases yielded inadequate material. Immunohistochemistry was required for classification of lymphoma cases and confirmation of a mixed component in germ cell tumours.

**Conclusion:** Mediastinal masses create significant diagnostic dilemma for the clinicians, radiologists and histopathologists. While imaging studies help in narrowing the differential diagnosis, accurate categorisation is not always possible. FNAC is a useful and cost effective tool. However, sampling error and complexities in performing the technique are major hurdles in the usefulness of this diagnostic modality.

**Key Words:** Mediastinum, Thymoma, Lymphoma, Seminoma, Neurogenic tumours

## INTRODUCTION

The mediastinum extends antero-posteriorly from the sternum to the spine and from the thoracic inlet down to the diaphragm. The so-called Pandora's Box has been divided into four compartments, i.e. superior, anterior, middle and posterior. While imaging studies suggest the location of the lesion and help in narrowing the differential diagnosis, morphological assessment by fine needle aspiration cytology (FNAC) or histopathological examination is imperative prior to therapeutic intervention. Although FNAC is a cost effective tool for the establishment of pre-operative diagnosis of mediastinal masses, there is a complexity and technical difficulty associated with this technique owing to the narrow anatomic space of mediastinum. Also, there are well known diagnostic pitfalls of this technique that limit its use (1).

Despite being a well-delineated narrow space, the diversity of lesions arising in the mediastinum pose an interesting diagnostic challenge for the clinicians, radiologists and

pathologists (2). There is limited data regarding the clinical and pathological features of these enigmatic mediastinal masses.

Our study highlights the clinico-pathological features in patients presenting with mediastinal masses with special emphasis on the utility of pre-operative diagnostic modalities like imaging and FNAC.

## MATERIAL and METHODS

The archives of the Department of Histopathology were retrospectively reviewed over a period of five years from January 2011 to January 2016. Of the 65,089 cases received in our department, patients who underwent surgical excision for mediastinal masses were included in this study. Patients with bronchogenic, esophageal and metastatic tumours were excluded. Clinical details including detailed history, physical examination and results of routine blood investigations were obtained. Pre-operative chest radiographs were obtained in all patients

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with suspected mediastinal lesions, following which computed tomography (CT) was performed for further characterisation of the lesion. CT-guided fine needle aspiration (FNAC) was performed wherever possible. Aspirates were smeared on clean glass slides, air dried and stained by May-Grünwald-Giemsa (MGG) stain. There were no cell block preparations studied. Following surgical excision, the excised specimen was fixed in 10% neutral buffered formalin and sent for histopathological evaluation. Diagnosis was confirmed on hematoxylin and eosin (H&E)-stained formalin-fixed paraffin-embedded sections. For cases in which final diagnosis could not be made on routine examination, a detailed immunohistochemical panel comprising of cytokeratin (CK), cluster of differentiation 45 (CD 45), CD 20, CD3, CD15, CD 30 and TdT was applied. Further, OCT-3/4, alpha feto protein (AFP) and beta-human chorionic gonadotropin ( $\beta$ -HCG) were applied to rule out a mixed component, if any, in a germ cell tumour. In brief, sections measuring 3-4  $\mu$ m thick were cut, deparaffinized with xylene and brought to water through graded levels of alcohol. Endogenous peroxidase activity was blocked by treating the slides with hydrogen peroxide for 30 min at room temperature. Antigen retrieval was done by immersing the slides in citrate buffer using the pressure cooker method. The slides were then incubated overnight with the primary antibody (rabbit polyclonal) at 4 °C in a humidified chamber. The following day, secondary antibody was added. The sections were then incubated with di amino benzidine (DAB) for visualization of the peroxidase reaction. After being washed in water, the sections were counter stained with haematoxylin,

dehydrated in alcohol, cleared in xylene and mounted. Immunohistochemistry (IHC) was interpreted in a binary fashion as positive or negative.

The FNAC smears, H&E stained sections and immunohistochemistry slides were reviewed by two pathologists and correlated with the clinical findings including radiology.

## RESULTS

Of the 65,089 cases received in our department, 60 patients with mediastinal masses were included in the study. Among these, the majority of the patients presented with a mass in the anterior mediastinum (36.7%), followed by the middle mediastinum (33.3%) and posterior mediastinum (30%). The compartment-wise histopathological diagnosis of the mediastinal masses is described in Table I. The mean patient age at the time of presentation was 37.5 years (range 2-61 years). There were 43 males and 21 females with a male: female ratio of 2:1.

Retrospective analysis of the clinical and radiological findings of these patients was done. The most common presenting complaint was cough, seen in 54 patients (87.0%), followed by chest pain (50; 80.6%), weight loss (36; 58.0%), dyspnoea (26; 41.9%) and hoarseness (14; 22.5%). Myasthenic symptoms were seen in four patients with thymoma (Table II). Two patients each of thymic cyst and mediastinal lipoma were diagnosed incidentally. Routine blood investigations including complete hemogram, liver function tests and kidney function tests were normal in all patients except four cases of tubercular lymphadenitis

**Table I:** Histopathological diagnosis of the mediastinal masses

	Anterior mediastinum	Middle mediastinum	Posterior mediastinum
Thymoma	12 (20%)		
Lymphoma		10 (16.6%)	
Tubercular lymphadenitis		8 (13.3%)	
Extragonadal germ cell tumour	4 (6.6%)		
Schwannoma			8 (13.3%)
Neurofibroma			6 (10%)
Ganglioneuroma			2 (3.3%)
Neuroblastoma			2 (3.3%)
Bronchogenic cyst		2 (3.3%)	
Thymic cyst	2 (3.3%)		
Retrosternal goitre	2 (3.3%)		
Lipoma	1 (1.6%)		
Thymolipoma	1 (1.6%)		
<b>Total</b>	<b>22</b>	<b>20</b>	<b>18</b>

**Table II:** Clinical features of patients with mediastinal tumours

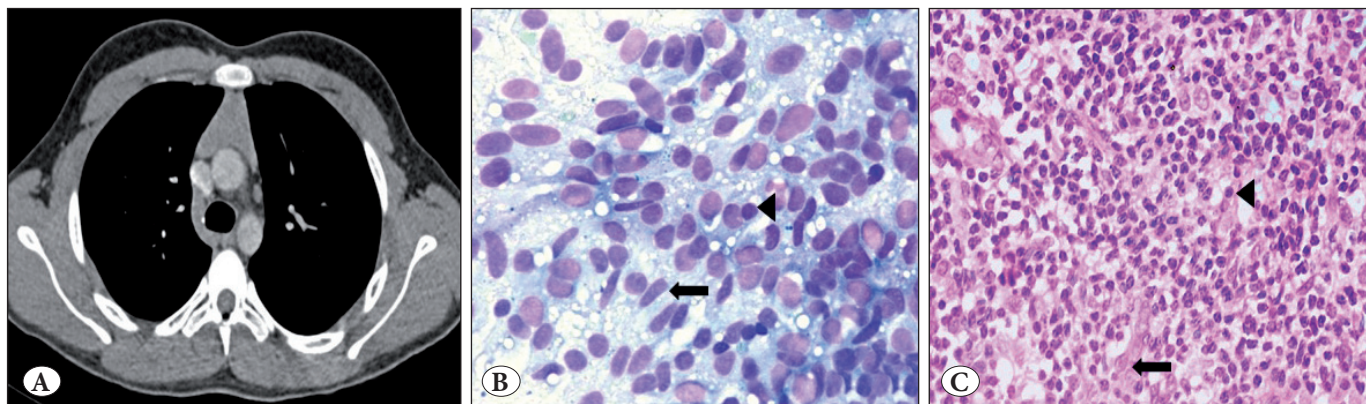
Symptom	Cases (Percentage %)
Cough	54 (87.0%)
Chest pain	50 (80.6%)
Weight loss	36 (58.0%)
Dyspnoea	26 (41.9%)
Hoarseness of voice	14 (22.5%)
Myasthenia gravis	4 (3.2%)

with raised erythrocyte sedimentation rate (ESR). Chest X-ray showed mediastinal widening in 40 patients. CT scan revealed the presence of mediastinal masses in their respective compartments in all cases. Mediastinal lymphadenopathy was noted in 18 cases on CT scan. CT guided FNAC was performed in 54 patients.

Histopathological examination revealed 73.33% benign and 26.67% malignant lesions. Among the twelve cases of thymoma, the most common variant was the AB subtype (9 cases) followed by two cases of B1 subtype and one case of thymic carcinoma. Thymic tumours were classified as per the World Health Organisation (WHO) classification. Six patients with anterior mediastinal masses showed a biphasic population of spindle-shaped epithelial cells interspersed with lymphoid cells on FNA smears. A provisional diagnosis of thymic hyperplasia versus thymoma was considered. The excision biopsy specimen showed features suggestive of thymoma, AB subtype. On the other hand, FNA smears from three patients predominantly showed presence of haemorrhage along with few scattered lymphoid cells and were inconclusive. Subsequently they were diagnosed as thymoma, AB subtype on histopathological examination (Figure 1A-C). Two female patients suffering with

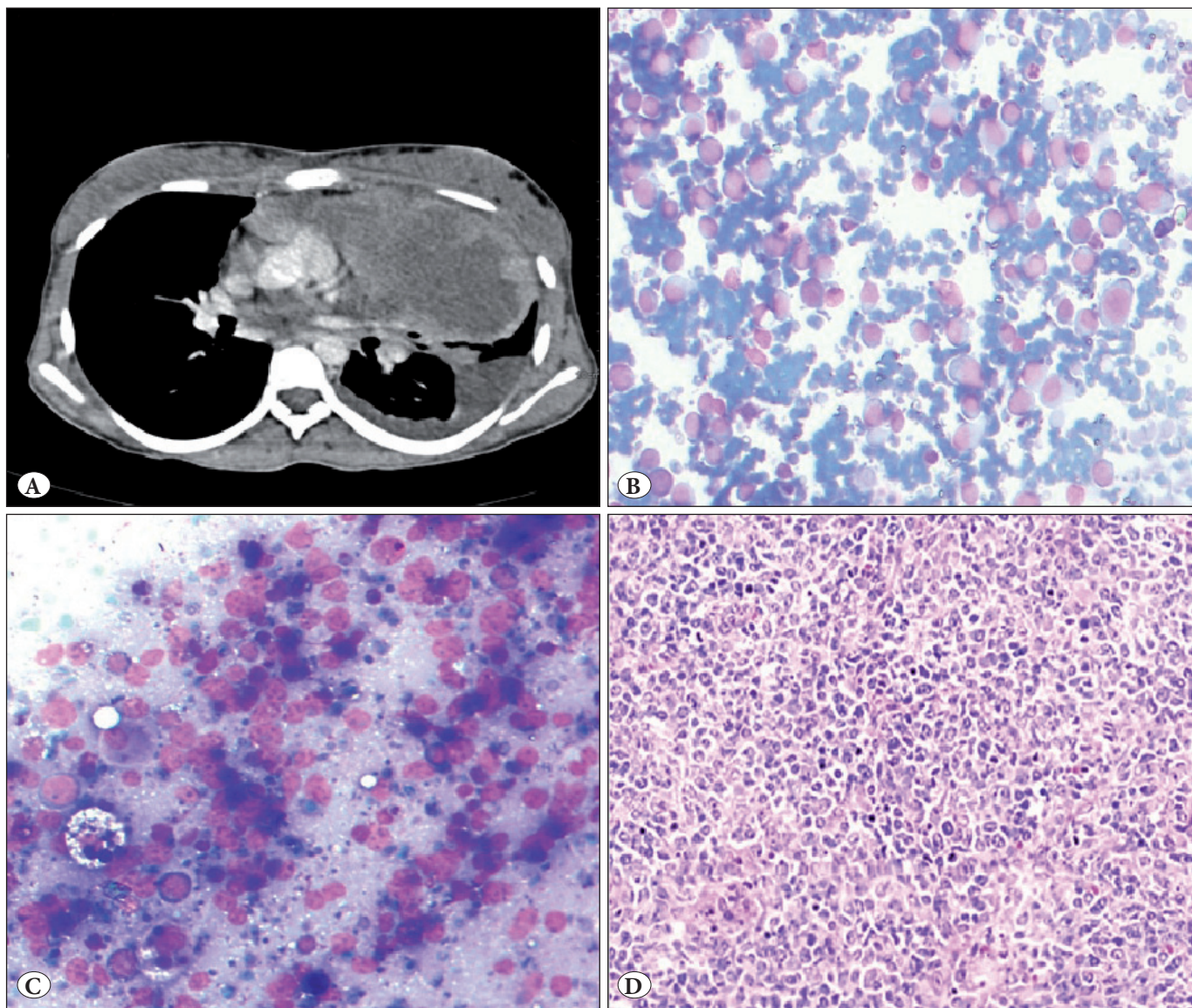
myasthenia gravis, aged 42 years and 46 years respectively, showed cellular FNA smears consisting of monomorphic lymphoid cells. A few scattered spindle-shaped epithelial cells were also seen along with occasional atypical mitosis. Differential diagnosis of lymphoma versus thymoma was considered. Histopathology revealed diffuse effacement of thymic architecture by a monomorphic population of lymphoid cells intermixed with epithelial cells. On IHC, the lymphoid cells were positive for TdT and CD3 while the epithelial cells were positive for CK, thereby excluding lymphoma. A final diagnosis of thymoma, B1 subtype was made in both the cases. On the other hand, an elderly female presented with a rapidly enlarging mediastinal mass associated with vague chest pain and dyspnoea. Histopathological examination revealed an un-encapsulated, poorly circumscribed tumour composed of large polygonal cells arranged in nests and diffuse sheets. The individual tumour cells were polygonal with moderate amount of dense eosinophilic cytoplasm and central vesicular nucleus with single nucleolus. Numerous atypical mitoses and tumour necrosis were seen. Foci of keratinisation were evident. Features were suggestive of keratinizing squamous cell carcinoma in concordance with the FNAC findings. A thorough clinico-radiological work up was done to rule out the possibility of metastasis, and a final diagnosis of primary thymic carcinoma was rendered.

Mediastinal lymphadenopathy was seen in 18 cases. Among 10 cases of lymphoma, there were 6 cases of Non- Hodgkin Lymphoma (NHL) and four cases of classical Hodgkin lymphoma (HL). There was no difficulty in FNAC diagnosis of these lesions except for three cases predominantly showing haemorrhage with few scattered lymphoid cells, inconclusive for an opinion. Histologically, most common type of NHL was diffuse large B cell lymphoma, seen in



**Figure 1:** A) CT scan of a case of thymoma showing symmetrical diffuse enlargement of thymus. B) FNA smear showing biphasic population of spindle-shaped epithelial cells (arrow) interspersed with lymphoid cells (arrow head) (May Grünwald Giemsa; x200). C) Presence of lymphocytes (arrow head) admixed with thymocytes (arrow) favouring thymoma- AB sybtype (H&E; x200).



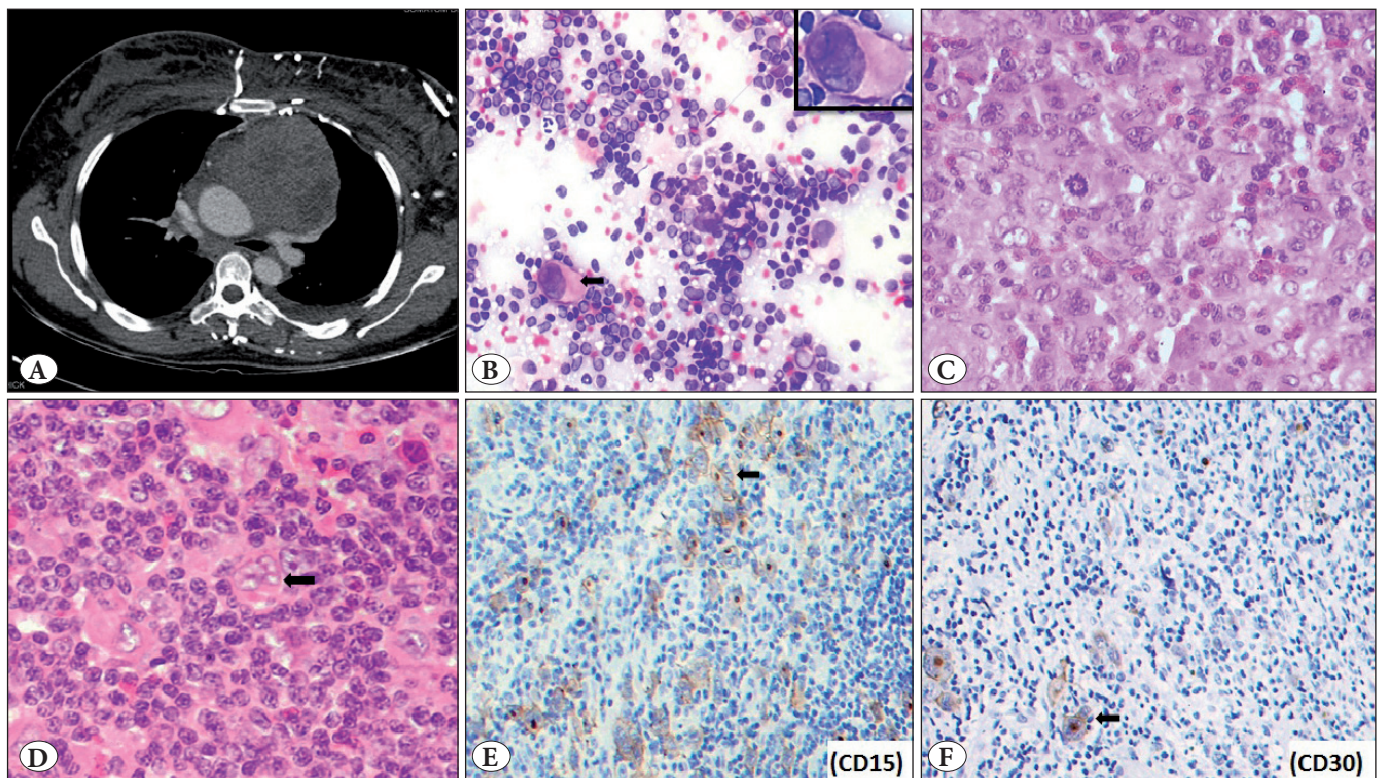


**Figure 2:** A) CT scan shows a large anterior mediastinal mass measuring 16x11 cm with central necrotic area invading into the adjacent myocardium and involving the pulmonary artery, ascending aorta & left lung upper lobe, suggestive of lymphoma. B-C) Diffuse arrangement of monomorphic population of immature lymphoid cells in FNA smears (May Grünwald Giemsa; x200). D) Diffuse effacement of lymph node architecture by large atypical lymphoid cells suggestive of NHL (H&E; x200).

5 cases (Figure 2A-D). On IHC the cells were positive for CD45, CD19, CD20 and negative for CD15, CD30 and EMA. Nodular sclerosis subtype was reported in all 4 cases of Hodgkin's lymphoma. IHC showed that the Reed-Sternberg (RS) cells were positive for CD45, CD 15 and CD 30 (Figure 3A-F). In another case, a 45-year-old man presented with dyspnoea and excision biopsy of the mediastinal lymph node revealed diffuse effacement of architecture by clusters of epithelioid histiocytes. Interspersed in between were small lymphoid cells with scant cytoplasm and round to oval hyperchromatic nuclei

showing mild atypia. Few binucleate RS like cells were also seen. On IHC, the lymphoid cells were positive for CD45 and CD3. The RS like cells were negative for CD15 and CD30, thereby excluding Hodgkin's lymphoma (Figure 4A-F). Thus a final immunomorphological diagnosis of Lennert's lymphoma was rendered. On the other hand, necrotizing granulomatous lymphadenitis was seen on histopathology of the remaining 8 patients with mediastinal lymphadenopathy. Stain for acid fast bacilli (AFB) was positive in these cases and a diagnosis of tubercular lymphadenitis was given (Figure 5A,B). Of these, seven





**Figure 3:** A) CT scan shows a homogeneous soft tissue mass in superior and anterior mediastinum encasing the great vessels without narrowing them. B) FNA smear showing classical RS cells seen in a case of HL (May Grünwald Giemsa; x200). C) Polymorphous background comprising of lymphocytes, eosinophils and macrophages (H&E; x200). D) Classical RS cell against a polymorphous background (H&E; x200). E) RS cells are positive for CD15 (CD15; x200). F) RS cells are positive for CD30 (CD30; x200).

patients had disseminated tuberculosis while one patient was diagnosed with isolated tubercular lymphadenopathy involving the mediastinal lymph nodes only.

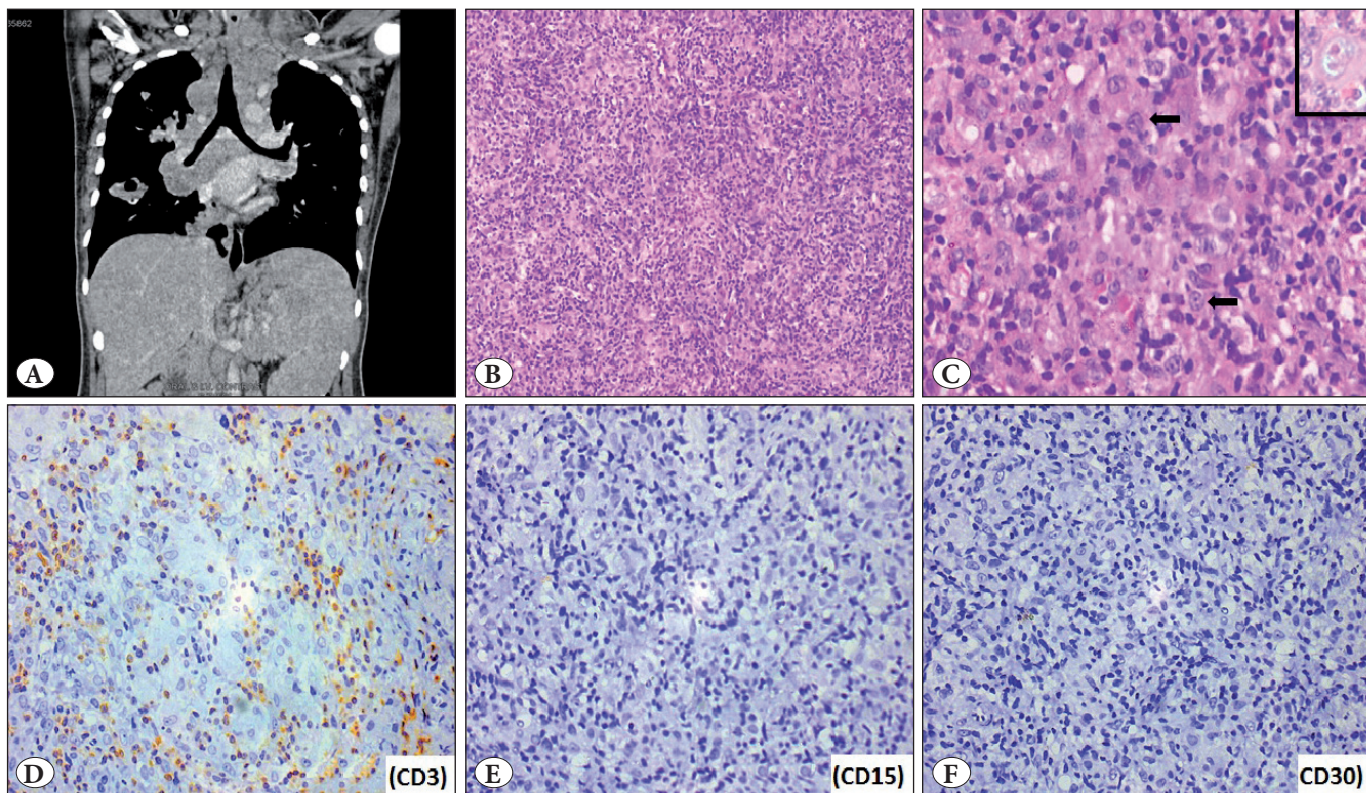
Germ cell tumours were seen in four cases located in the anterior mediastinum. Two cases showed moderately cellular FNA smears against a tigroid background with presence of scattered lymphocytes. The individual tumour cells were large with moderate amount of vacuolated cytoplasm and a large vesicular nucleus with one or two prominent nucleoli. Provisional diagnosis of germ cell tumour, possibly seminoma, versus lymphoma was considered. Histopathological examination and immunohistochemical positivity for OCT3/4 confirmed the diagnosis of classical seminoma in both these cases (Figure 6A-C). A single case of mature teratoma (Figure 6D-F) was diagnosed in a 22-year-old male patient. CT scan revealed a large mediastinal mass involving the anterior mediastinum with foci of calcification. FNA smears showed presence of nucleated and anucleated squamous cells only. Another case was diagnosed as mixed germ cell tumour showing histomorphological features of seminoma and yolk sac tumour confirmed by OCT3/4 and AFP IHC respectively.

While there was no difficulty in the FNA diagnosis of seminoma, the yolk sac component could not be identified cytologically.

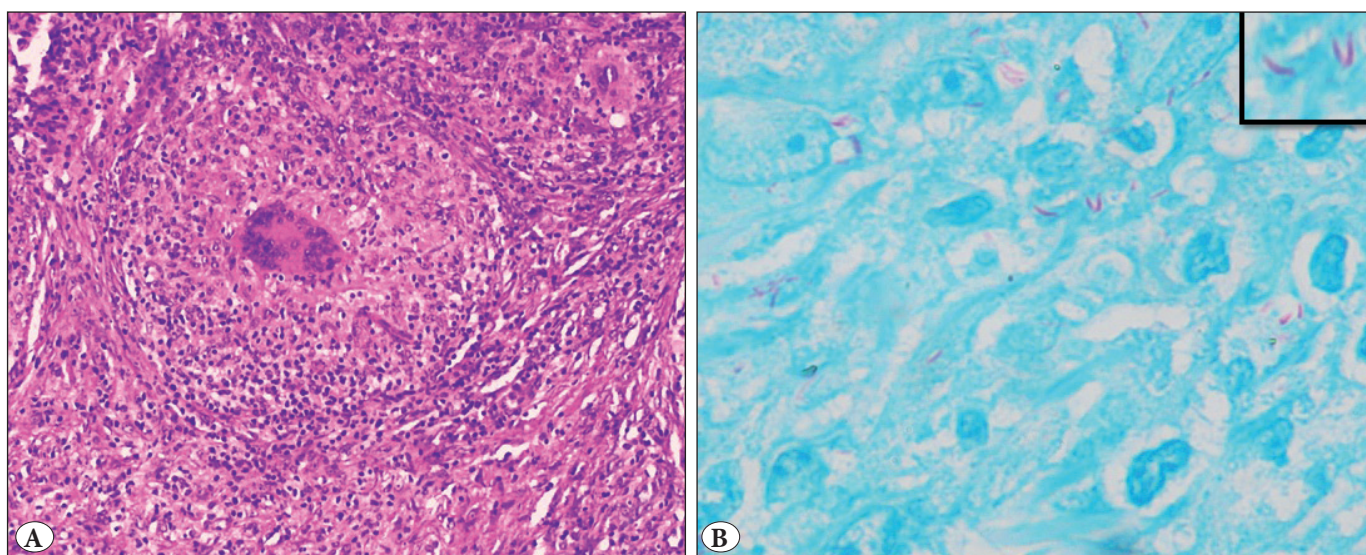
Eighteen cases of neurogenic tumours were identified in the posterior mediastinum. Of these, schwannoma was the most common and seen in 8 patients (Figure 7A), followed by six cases of neurofibroma (Figure 7B) and two cases of ganglioneuroma. FNA smears in the majority of these cases were moderately cellular with presence of spindle-shaped cells either in clusters or singly scattered. The individual cells were spindle-shaped with scant to moderate cytoplasm and hyperchromatic spindle-shaped nucleus. No Verocay bodies, nuclear atypia or stromal fragments were seen. A provisional FNA diagnosis of spindle cell tumour was given in ten of these cases, while the remaining cases showed paucicellular smears and were labelled inconclusive for opinion. Two cases of neuroblastoma (Figure 8A,B) were seen in a 2- and 4-year-old male child respectively, FNA smears of which were inadequate for diagnosis.

Cystic lesions of the mediastinum consisted of two cases each of bronchogenic cyst, thymic cyst and retrosternal goitre with cystic change respectively, diagnosed on



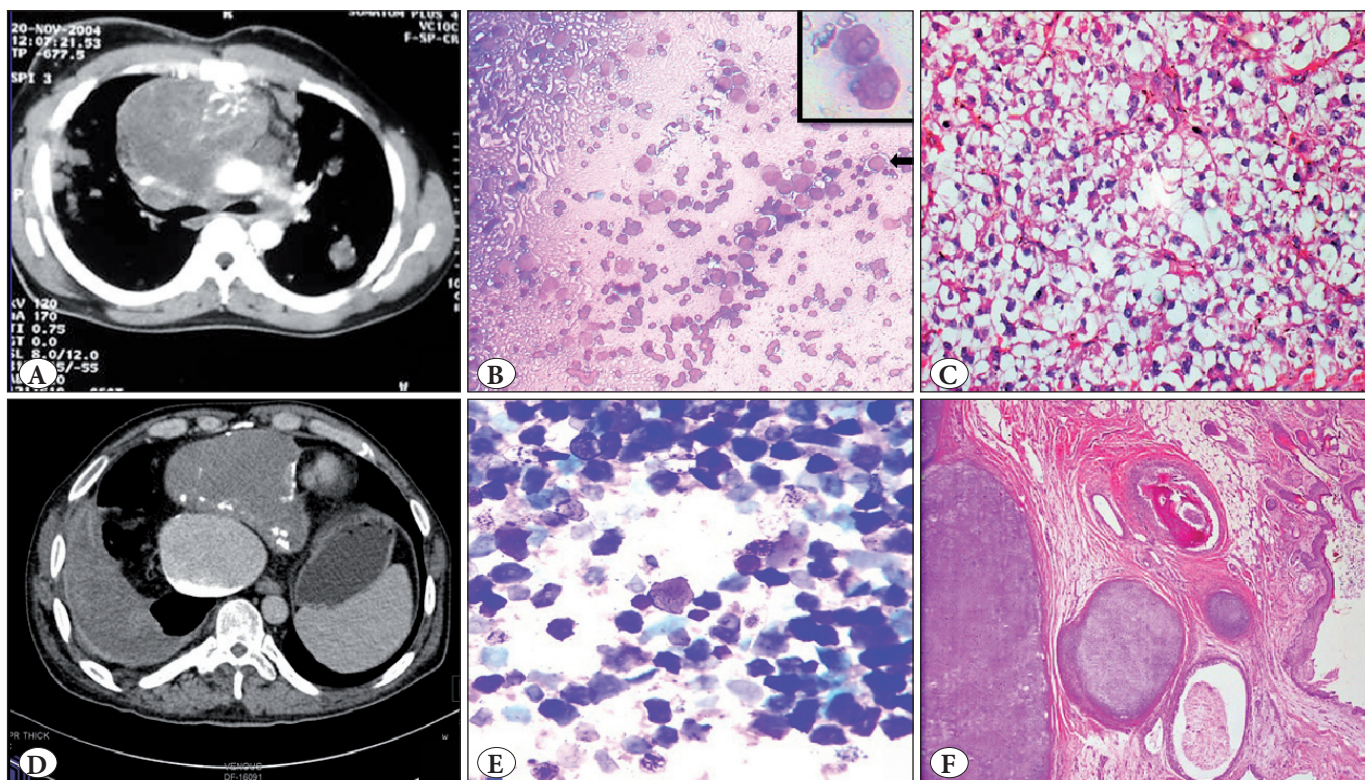


**Figure 4:** A) CT scan of a case of Lennert's lymphoma showing enlarged paratracheal and subcarinal nodes showing homogeneous enhancement. B) Diffuse effacement of lymph node architecture by clusters of epithelioid histiocytes (arrow) interspersed with lymphoid cells (H&E; x100). C) Higher magnification shows lymphoid cells with scant cytoplasm and round to oval hyperchromatic nuclei showing mild atypia. Few binucleate RS like cells are also seen (inset) (H&E ; x200). D) Lymphoid cells are positive for CD3 (CD3; x200). E) RS like cells are negative for CD 15 (CD15; x200) and F) CD30 (CD30; x200).

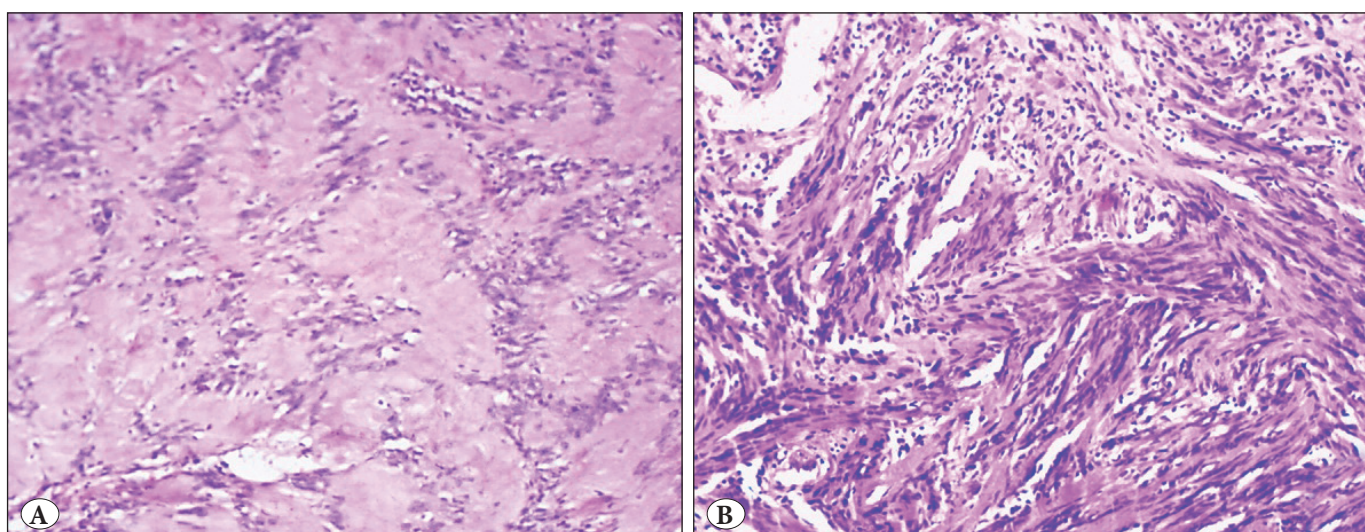


**Figure 5:** A) Epithelioid cell granulomas in a lymph node (H&E; x200). B) Stain for acid fast bacilli is positive (inset) diagnostic of tubercular lymphadenitis (Ziehl-Neelsen; x200)



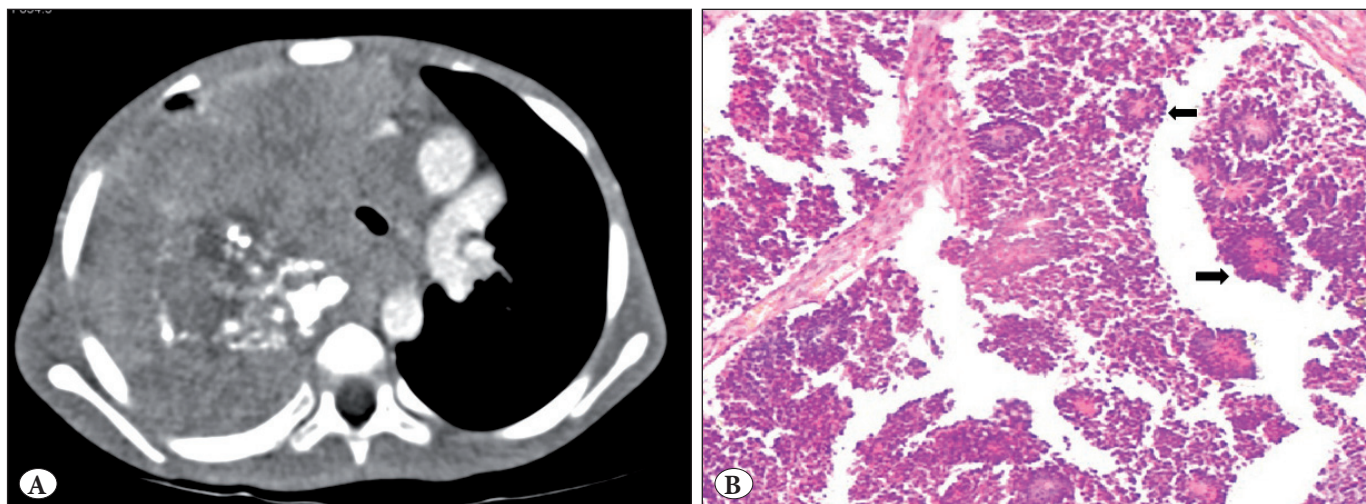


**Figure 6:** A) CT scan of a case of seminoma showing an anterior mediastinal mass with internal calcifications along with multiple nodular metastases seen in both lungs. B) FNA smear showing large atypical cells against a blood mixed tigroid background. The cells show vacuolated cytoplasm (arrow) and a vesicular nucleus with prominent 1-2 nucleoli (inset) (May Grünwald Giemsa; x200). C) Large tumour cells separated by thin fibrous septae infiltrated by lymphocytes. The tumour cells have clear cytoplasm and a central vesicular nucleus with prominent nucleoli favouring mediastinal seminoma (H&E; x200). D) CT scan of a case of teratoma showing soft tissue mass in anterior mediastinum with internal calcific foci. E) FNA smear shows anucleated squamous cells (May Grünwald Giemsa; x400). F) Keratinized stratified squamous epithelium with skin appendages, fat, cartilage and glands lined by tall columnar epithelium suggestive of mediastinal teratoma (H&E; x200).

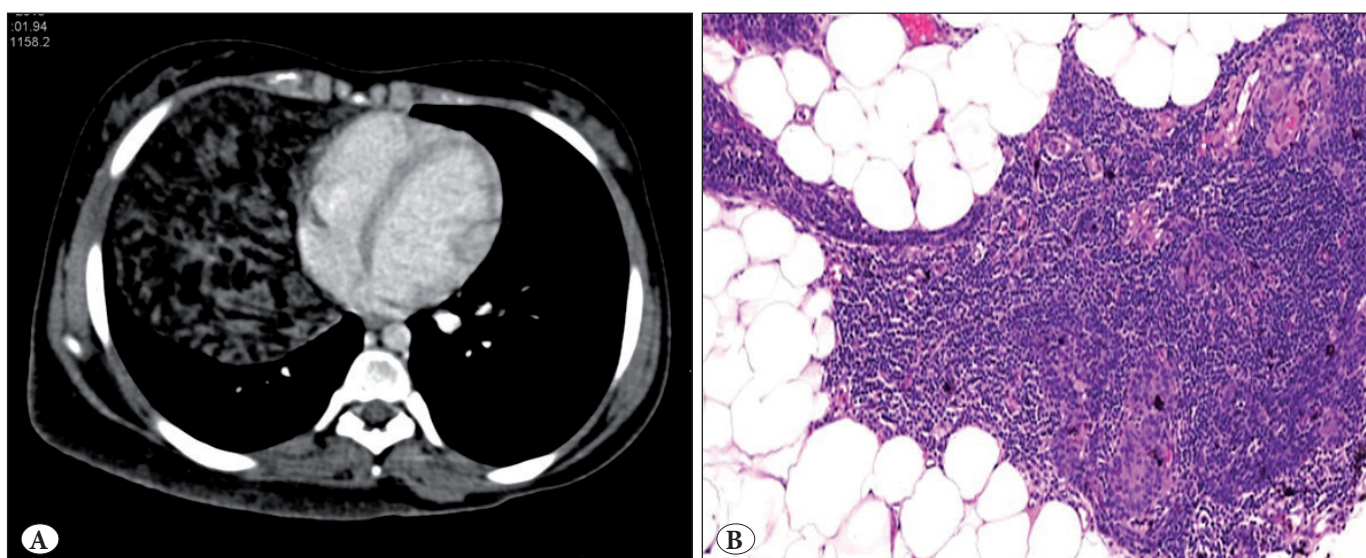


**Figure 7:** A) Numerous Verocay bodies in a case of schwannoma (H&E; x200). B) Neurofibroma showing spindle-shaped cells arranged in bundles with cells showing moderate eosinophilic cytoplasm with hyperchromatic wavy spindle-shaped nuclei (H&E; x200).





**Figure 8:** A) CT scan showing large heterogeneous mass with internal calcifications in the right hemithorax replacing the right lung. B) Diffuse sheets of small round blue cells with formation of rosettes (inset) suggestive of neuroblastoma (H&E; x200).



**Figure 9:** A) CT scan showing a large fat containing anterior mediastinal mass extending along the right pericardium. B) Thymic tissue intermixed with mature adipocytes suggestive of thymolipoma (H&E; x200).

histopathological examination of the excised specimen. FNAC was not performed in these cases owing to the cystic nature of the lesion. One case each of mediastinal lipoma was readily diagnosed on cytology and confirmed on histopathology. Also, a 22-year-old female patient showed a large fat-containing anterior mediastinal mass extending along the right pericardium on CT scan (Figure 9A). FNA showed features suggestive of lipoma. On histopathology, a well-encapsulated tumour was seen showing intermixed thymic tissue within mature adipocytes, suggestive of thymolipoma (Figure 9B).

## DISCUSSION

Primary mediastinal tumours are rare, accounting for 3% of tumours occurring within the chest (3). We studied 60 cases of mediastinal lesions received over a period of 5 years. The mean patient age was 37.5 years (range 2-61 years) with a male preponderance. Our findings were in concordance with the previously published literature (2, 4).

Nearly 50% of all mediastinal masses involve the anterior compartment, including thymoma, teratoma, lymphoma, thyroid and parathyroid lesions. Congenital cysts and



neurogenic tumours are most commonly observed in the middle and posterior mediastinum respectively (3). In our study, the anterior compartment (36.7%) was maximally involved followed by the middle mediastinum (33.3%) and posterior mediastinum (30%). Thymoma was the most common tumour of the anterior compartment followed by germ cell tumours. The middle mediastinum showed mainly nodal involvement with lymphoma most commonly followed by tubercular lymphadenitis. Neurogenic tumours were seen in the posterior mediastinum only. Studies conducted in the past showed nearly similar trends (5-7).

In the present study, 96.8% of the subjects were symptomatic at presentation. Dubashi et al. (5) reported a similar incidence of 97% with other studies reporting a range of 60-88% (2, 4).

In 1987 however, Davis et al. (4) reported a much higher incidence of asymptomatic cases (38%). This difference may be attributed to the fact that most of the patients in our setting visit the hospital for their symptoms rather than for routine examination. The most common symptoms reported in this study was cough followed by chest pain, dyspnoea and weight loss which was similar to the previously reported incidence (8, 9).

Pre-operative diagnosis of mediastinal masses is a challenging task owing to the non-specific clinico-radiological signs and technical difficulty in performing FNAC. The investigation of choice for the detection of mediastinal masses is CT scan (10). Magnetic resonance imaging (MRI) is of help in further categorisation of the lesions (11). In our study, CT scan identified mediastinal masses in all cases but was unable to categorise the lesion in any. CT scan thus helps in compartment localisation of the mediastinal mass. This further helps in narrowing the probable differential diagnosis.

CT-guided FNAC is a safe, useful and cost-effective method for the diagnosis of mediastinal masses. However, close proximity to vital organs can be a hindrance for this technique. In our study, FNAC was performed in 54 cases. There were 7 cases which revealed inadequate material. Thymic lesions often pose a diagnostic challenge for the reporting cytologist. While the lymphocyte-rich variant of thymoma (B1 subtype) can easily be misdiagnosed as lymphoma (11), epithelial rich variant (A subtype) on the other hand is a close mimicker for carcinoma (12). Similar to previous studies, our study also highlights the difficulty encountered in the FNA diagnosis of thymic lesions especially two cases of B1 subtype where the possibility of a lymphoma could not be ruled out and six cases of AB

subtype in which thymic hyperplasia versus thymoma could not be accurately diagnosed on FNAC. Thymic carcinoma on the other hand was readily diagnosed due to the frank malignant features.

Apart from the thymic lesions, neurogenic tumours are difficult to categorise on cytology. Schwannomas are usually difficult to differentiate from neurofibroma unless characteristic Verocay bodies are seen, while ganglioneuroma cannot be diagnosed till ganglion cells are aspirated. Both these findings were not seen in our cases and a final diagnosis of spindle cell tumour was rendered on FNAC. Further, some degree of cellular and nuclear atypia is commonly seen in neurogenic tumours. This warrants a careful search for mitotic figures and excision biopsy for proper histopathological evaluation. Thus characterisation of benign versus malignant on FNAC is a diagnostic difficulty which is then overcome on histopathology. On the other hand, germ cell tumours cannot readily be diagnosed on FNAC since tigroid background is not entirely specific for seminoma and when lymphocytes/ lymphoglandular bodies are present, a confusion with malignant lymphoma is possible, as seen in two of our cases. Furthermore, smears showing discohesive pattern on cytology should be differentiated from poorly differentiated adenocarcinoma, melanoma and lymphoma. In our study, the component of yolk sac tumour was missed in a single case of mixed germ cell tumour. This can be attributed to selective sampling which is a well-known hurdle in aspiration cytology. As reported earlier by Chhieng et al. particularly in case of mixed germ cell tumours may also pose diagnostic challenge (13). Similarly, fat fragments were selectively sampled from an anterior mediastinal mass in a 22-year-old female which was diagnosed later as thymolipoma on histopathological evaluation.

A rather rare case encountered in our study which deserves special mention was Lennert's lymphoma in a 33-year-old male patient with mediastinal lymphadenopathy involving the middle compartment. Lennert's lymphoma is a variant of peripheral T-Cell lymphoma, not otherwise specified in the WHO classification of 2008 (14). Its rarity and the lack of strict diagnostic criteria explain why only a few reports on Lennert's lymphoma have been published so far. Our case showed presence of RS like cells mimicking Hodgkin's lymphoma and posed the same diagnostic dilemma as the previously published cases. In 2013, Parimal et al. (15) reported a series of 5 cases of Lennert's lymphoma which constituted 0.71% of all peripheral T-cell lymphomas in their institution. They concluded that classical Hodgkin's lymphoma is a close mimicker of Lennert's lymphoma



and suggested T cell gene rearrangement studies when distinction cannot be made on morphology and IHC.

To conclude, we evaluated the role of clinico-radiological, cytological and histopathological findings in the differential diagnosis of mediastinal masses. Owing to rather non-specific clinical and radiological signs, histopathological examination remains the mainstay of diagnosis. Further, the proximity of this area to vital visceral organs decreases the usefulness of FNAC and leads to a failure to adequately describe cytomorphological characteristics of the mediastinal mass.

### CONFLICT OF INTEREST

The authors declared no conflict of interest.

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