A STUDY OF A SPECTRUM OF LESIONS IN FINE NEEDLE ASPIRATION SMEARS OF CERVICAL LYMPH NODE AMONG WORKERS IN A TEXTILE INDUSTRY AREA
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ABSTRACT

BACKGROUND
Textile industry workers face so many health-related challenges compared to the general population. Lymphadenopathy is one of the most common presenting symptoms of these people. This study is done in inhabitants of Tiruppur, a major textile industry area in the state of Tamil Nadu, India, where people work in garment factories for low wages.

The aim of the study is to-
1. Evaluate the usefulness of Fine Needle Aspiration Cytology (FNAC) as a diagnostic tool in cases of cervical lymphadenopathy.
2. Analyse the various cytomorphological presentations of tuberculous lymphadenopathy.

MATERIALS AND METHODS
In this study, a spectrum of lesions was diagnosed by fine needle aspiration of lymph nodes of 56 patients presenting with cervical lymphadenopathy. The diseases were categorised into suppurative lymphadenitis, granulomatous lymphadenitis, metastatic tumour deposits and lymphoproliferative disorder. Various morphological presentations of tuberculous lymphadenitis were also analysed.

RESULTS
From this study, it is observed that the most common diagnosis of cervical lymphadenopathy is reactive lymphadenitis (34%), followed by granulomatous lymphadenitis (30%). Also, among the tuberculosis cases, it is found that majority of cases had only granulomas (45%) followed by caseous necrosis with granulomas (28%).

CONCLUSION
Knowing the usefulness of FNAC, it is concluded that it is a valuable, noninvasive, reliable and cost-effective technique in such a high-risk population. It also helps the clinician to start treatment based on FNAC findings and do the necessary at the earliest.

KEYWORDS
Textile Industry Workers, Lymphadenopathy, FNAC, Suppurative Lymphadenitis, Tuberculosis, Metastatic Deposits, Lymphoproliferative Disorder.

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BACKGROUND
Workers in the textile industry are exposed to a number of health hazards.¹ Exposure to chemicals like Benzidine, solvents and fixatives, formaldehyde lead to nose and lung cancer,² carcinoma of buccal cavity, oesophageal cancer, etc. Basically, there are three types of interactions in the working environment. They are: i) Man and physical agents, chemical and biological agents; ii) Man and machine, iii) Man and man. Industrial health hazards can be classified into three classes - chemical hazards, in which the body absorbs toxin; ergonomic hazards, which involves improper lifting or repetitive stress and physical hazards, in which the body is exposed to extreme temperature, pressure or noise. Garment workers constitute a major share of the total labour force in the country, which bring much of the country's foreign currency. Many patients who work in these industries present with lymph node enlargement, most commonly cervical lymph nodes. Cervical lymph nodes can be categorised into inflammatory, neoplastic and degenerative diseases. More common causes include acute bacterial infection, tuberculosis, sarcoidosis, chronic nonspecific lymphadenitis, lymphoproliferative disorder and metastatic tumour deposits. In India, more common aetiology is tuberculosis. Tuberculosis is caused by Mycobacterium tuberculosis bacilli. These are classified as acid-fast bacilli because of their unique cell wall structure crucial to their survival. The disease is spread by small air borne droplets called droplet nuclei. The disease transmission is influenced...
by number of bacilli in the droplets, virulence of the bacteria and degree of ventilation. Although, the pulmonary system is most commonly involved, more than 20% of the patients are affected by extrapulmonary tuberculosis. Lymphatic system is the most common extrapulmonary organ involved and cervical lymphadenopathy occurs in most cases. People who work in textile industries have increased risk of tuberculosis compared to general population. Tuberculosis has been called 'modern epidemic' since it is closely associated with industrial workers where people crowd together in an environment, which can easily spread tubercle bacilli from one person to another. Patients usually present with fever and multiple matted lymph nodes on examination. But, different presentations have emerged with various clinical signs like solitary lymphadenopathy and their cytological presentations also differ. Not all cases present with caseous necrosis and epithelioid cell granulomas. Atypical presentations have become more common.

Fine Needle Aspiration Cytology (FNAC) is a technique introduced in Europe in the 1950s by Lopez-Cardozo in the Netherlands and Soderstrom in Sweden. Ublication by Zajicek from Karolinska Hospital brought aspiration cytology to international alterations. It is a relevant, simple and diagnostic investigation in cases of cervical lymph node enlargement because of ease accessibility. The procedure can be done in ease in paediatric age group patients to avoid surgery procedures. It is worth mention about Polymerase Chain Reaction (PCR), which is a diagnostic molecular technique, which has been so widely publicised during the past decade. It can be done on sputum smears, blood mononuclear cells and fresh lymph node aspirates. This is a retrospective study of 56 patients in Tiruppur, a well-known industrial city in the country, who were clinically diagnosed to have cervical lymphadenopathy. Fine needle aspiration was done in all cases of cervical lymph node enlargement and smears reported were analysed.

MATERIALS AND METHODS
This study was conducted over a period of 1½ years from January 2015 to July 2016 in Tiruppur inhabitants who work in textile industries. The study included aspirates from 56 cases who presented with cervical lymphadenopathy to the outpatient department. A detailed history was taken including history of fever, weight loss, loss of appetite, any other family members having similar symptoms, etc. A relevant clinical examination was done to assess single or multiple cervical lymph nodes, matted or discrete nodes and other clinical findings if any. Relevant investigations were taken like chest x-ray, blood investigations and ESR. FNAC was done using a 22-gauge needle and smears fixed in alcohol. Slides were stained with Haematoxylin-Eosin stain, analysed and cytological diagnosis was made.

Steps Used in Haematoxylin and Eosin Staining Method-
1. Slides fixed in alcohol.
2. Stained in haematoxylin for 3 minutes.
3. Washed in tap water.
4. Two changes in alcohol.
5. Stained with eosin for 30 seconds.
6. Dehydrated.
7. Clearing and mounting done.

Inclusion Criteria
1. All patients with significant cervical lymph node enlargement.
2. All age group.

Exclusion Criteria
1. Smears with inadequate material.
2. Insignificant lymph node.
3. Lymph nodes other than cervical region.

Cases were categorised into reactive lymph node, granulomatous lymphadenitis, supplicative lymphadenitis, lymphoproliferative disorder and metastatic tumour deposits. Cases which were diagnosed as tuberculosis were separated into supplicative, granulomatous and granulomas with caseous necrosis and analysis was done.

PCR was done in cases, which were doubtful of tuberculosis, especially which presented with supplicative lymphadenitis. For this, the aspiration material was immediately frozen at -20 degree Celsius and sent for analysis.

PCR was performed using primers 59-TCCGTGCCAGTCGTTCC-39 and 59-GTCTCCTCCGAGTCTA GGCCA-39 to amplify a 240 bp region (460-700) from the gene encoding the MPB 64 kDa protein. Amplification of the gene was done in 25 l reaction mixture containing 50-100 ng of genomic DNA, 10 mM Tris/HCl (pH 8.0), 1.5 mM magnesium chloride, 50 mM potassium chloride, 200 M of each dNTP (dATP, dGTP, dCTP and dTTP), 10 pmol of each primer and 0.5 U Taq DNA polymerase. DNA was first denatured at 94 8C for 4 mins., then followed by 29 cycles of denaturation at 94 8C for 30s, primer annealing at 55 8C for 30s and extension at 72 8C for 30s. In the final cycle, extension at 72 8C was allowed for 4 mins. Positive (mycobacterium tuberculosis DNA) and a negative (distilled water) controls were included for each experiment. Amplified DNA was electroforesed on a 3% Ethidium bromide-stained agarose gel along with HaeIII-digested X174 DNA molecular mass markers and photographed under a UV transilluminator. Amplicon identity was confirmed in each instance by Southern hybridisation. The sequence of the probe was chosen from the middle part of the amplified sequence of the gene (59-CTTCAACCGGGGAGT-39).

Observation
Among all 56 cases who presented with cervical lymphadenopathy, 39 % were males and 61% were females. The youngest patient in this study was 9-month-old female child and the oldest patient was 85 years. Hence, it is observed that there is wide variation in the age of presentation and predominant patients were females.

Out of 56 cases studied, following diagnoses were obtained (Table 1) (Figure 1 and Figure 2).
Table 1. Spectrum of Cervical Lymph Node Lesions in FNA Aspirations of Cervical Lymph Node

<table>
<thead>
<tr>
<th>Lesions</th>
<th>Number of Cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Suppurative lymphadenitis</td>
<td>14</td>
</tr>
<tr>
<td>Granulomatous lymphadenitis</td>
<td>17</td>
</tr>
<tr>
<td>Reactive lymphadenitis</td>
<td>19</td>
</tr>
<tr>
<td>Lymphoproliferative disorder</td>
<td>02</td>
</tr>
<tr>
<td>Metastatic tumour deposits</td>
<td>04</td>
</tr>
</tbody>
</table>

FNA smears showing plenty of polymorphs in a necrotic background were diagnosed as suppurative lymphadenitis. Those showing epithelioid cell granulomas were diagnosed as granulomatous lymphadenitis. Lymphoproliferative disorder showed either monotonous population of lymphoid cell population or polymorphous population of lymphocytes, plasma cells, eosinophils, histiocytes and Reed-Sternberg cells. Metastatic tumour deposits had clusters or scattered large epithelial cells in a lymphoid background.

Figure 3. Proportion of Lesions of Cervical Lymphadenopathy Diagnosed by FNAC

Cases of lymphoproliferative disorder were proceeded with histopathological examination for confirmation and characterisation. Metastatic lymph nodes included primary most commonly from the head and neck region.

It is found that the most common presentation of cervical lymphadenopathy in this study is reactive lymphadenitis (34%) followed by granulomatous lymphadenitis (30%) (Figure 3).

In this study, it is also found that presentations of tuberculosis in patients with cervical lymph node greatly differ and the variations are as follows.
malignant lesions. Textile industry is a highly labour-intensive industry throughout the world. Occupational health hazards depend on the type of the material used in the industry. Tiruppur is a major textile industry city in India. Exposure to bleaching, washing, textile dyes, various acids and dye setting media leads to major and minor health hazards. Exposure to some of the chemicals lead to increased risk of cancer. Studies on occupational cancer show that cotton industry is classified among those with increased cancer incidence. People who work in textile industries and live in the surrounding areas have more risk of developing signs of cervical lymphadenopathy. They are subjected to FNAC as a primary investigation since it is an easy, simple, noninvasive and cost-effective method of screening diseases. The diagnosis given in FNAC is often accepted as final diagnosis when there is no further correlation in many cases. Causes of cervical lymph node in such population include acute bacterial infection, tuberculosis, nonspecific reactive causes, lymphoproliferative disorder and metastatic tumour deposits. Acute supplicative lymphadenitis shows numerous neutrophils. Bacteria maybe seen. Reactive lymph nodes show hypercellular smears with numerous small round lymphocytes, admixed with medium and large non-cleaved and cleaved lymphoid cells. Characteristic follicular centre cell fragment (germinal centre) is seen with tingible body macrophages. The lymphocytes are activated and there is often an increase in the numbers of immunoblasts. The smears show other cell types in the background- plasma cells, neutrophils, eosinophils and histiocytes. If abundant plasma cells are seen, it may suggest syphilitic lymphadenitis, SLE or rheumatoid arthritis. Other specific entities include cat-scratch disease, Kikuchi’s lymphadenitis and Rosai-Dorfman disease. Lymph nodes with follicular hyperplasia, if managed properly, usually subside within few weeks. The diagnosis of metastatic tumour deposits in lymph node is crucial, but highly reliable. In most cases, the primary tumour is clinically known and FNAC is used for follow up of the patients. However, sometimes, it would be a sole indication for searching the primary tumour, especially in cases of occult carcinoma. In cases of lymphoid malignancies, FNAC is used mainly to assess the staging and to recognise the residual and recurrent lymphoproliferative disorder. Among the lymphoid malignancies, FNAC is said to have a higher diagnostic accuracy in case of Hodgkin’s lymphoma. It shows the presence of classical Reed-Sternberg cells along with larger atypical mononuclear cells in a background of mixed population of lymphoid cells comprising of lymphocytes, plasma cells and histiocytes. Sometimes, prominent eosinophilic infiltrates are seen. Diagnosis of non-Hodgkin’s lymphoma by FNAC is at times difficult when biopsy is needed for confirmation and subtyping. Tuberculosis is the most common cause of cervical lymphadenopathy in countries like India where it continues to be a major health problem with enormous social and economic implications. In cities like Tiruppur, which remains a soil for employment of low socioeconomic people, this disease is highly prevalent due to migration of

From the study, it is observed that the most common cytological presentation of tuberculosis is lymphadenitis with only granulomas (45%), followed by caseous necrosis with granulomas (28%) (Figure 4 and Figure 5).

10 cases of tuberculosis presented with acute onset of lymph node enlargement and showed suppurative lymphadenitis in the FNA smears. The fresh lymph node aspirates were sent for Tb-PCR examination and six were diagnosed to be tuberculous lymphadenitis (Table 2).

<table>
<thead>
<tr>
<th>Results of Tb-PCR</th>
<th>Number of Cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Suppurative Lymphadenitis with Positive Tb-PCR</td>
<td>6</td>
</tr>
<tr>
<td>Suppurative Lymphadenitis with Negative Tb-PCR</td>
<td>4</td>
</tr>
</tbody>
</table>

**Table 2. Results of Tuberculosis Cases Proven by PCR Method**

The cases which were positive for Tb-PCR were started antituberculosis treatment and those, which were negative were treated as acute bacterial infection.

**DISCUSSION**

Enlarged cervical lymph nodes are easily accessible for FNAC and are important in diagnosing malignant and non-

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population and also due to higher prevalence of HIV infection. During the past decade, there is a dramatic change in clinical pattern and presentation of tuberculosis. Atypical presentations of tuberculous lymphadenopathy have increased. Cytological variations in FNA aspiration smears have also increased in recent times. Follow up of the patients were often missed in such population and so histopathological examination was rarely done.

From this study, it is observed that the most common lesion diagnosed in FNAC of cervical lymphadenopathy is reactive lymphadenitis (34%), followed by granulomatous lymphadenitis (30%). Most common cytological presentation of tuberculous lymphadenitis is granulomas without caseous necrosis (45%). In a study by Kumar H,7 the most common pathological lesion was tuberculous lymphadenitis (47.67%), followed by reactive lesions (44.39). Hirachandr8 in his study found that the most common lesion in FNA was reactive hyperplasia (41.55%), followed by tuberculosis (28%). Fine needle aspiration is found to be very useful in diagnosis of tuberculous lymphadenopathy, which had various cytomorphological presentations in this study. In a study by Manitchotpisit,9 FNA in combination with PCR is found to be a fast and effective clinical approach to a case of tuberculosis, especially in cases, which show supplicative lymphadenitis. In a study by Tilak V, it was confirmed that FNAC is a highly diagnostic procedure for assessing swellings of head and neck region.10 Alam K et al in his study has shown the usefulness of FNAC in the diagnosis of metastatic lymphadenopathy.11 This procedure has now become widely used because of its easy availability of results, minimal trauma and complications.12 In all studies, it is observed that FNAC is a valuable test both for screening and follow up and even as a diagnostic tool if biopsy confirmation is not possible and in cases of tuberculosis, which present as supplicative lymph nodes without caseous necrosis or granulomas, PCR was also found to be useful in several cases, in which culture and smears showed negative results. It is a sensitive and specific diagnostic tool to diagnose tuberculosis. In a study by Iqbal S, it is found that is more sensitive than microbiological Zeel Nielson smear examination and LJ medium culture.13

CONCLUSION
FNAC is a very important and reliable technique for diagnosis and treatment of enlarged cervical lymph nodes when histopathological correlation is difficult. In developing countries like India, it serves a cost-effective procedure to suggest a preliminary diagnosis, which in many cases helpful in treatment. In addition, occupational safety and health is important for moral, legal and financial reasons. This study concludes that a spectrum of lesions can occur in FNAC of cervical lymph nodes in textile industry workers. Hence, cytopathology plays a vital role in diagnosis of cervical lymph node lesions. FNAC also helps the clinician to select appropriate treatment and modify planning in patients who require surgery.

REFERENCES