Original Research Article

CORRELATION OF CD4 COUNTS WITH THE FNAC PATTERNS OF TUBERCULOUS LYMPHADENITIS IN PATIENTS WITH HIV

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Abstract

Background: Tuberculosis is among the most common opportunistic infections in people living with HIV/AIDS and is also the most common cause of peripheral lymphadenopathy. These lymph nodes revealed four different patterns on FNAC, which indirectly reflected the immune status in these people. CD4 counts, being a primary marker for immunological status in HIV people, were used to initiate ART, monitoring disease progression and management. The present study was aimed to correlate these four cytomorphological patterns with CD4 counts.

Results: In the present study we observed that CD4 counts varied significantly with different cytological patterns. Pattern 1, having a lower CD4 counts, reflects a poor immune response, whilst pattern 4, having higher CD4 counts reflected a better immune response.

Conclusion: FNAC patterns can be used to predict the CD4 counts where flow cytometry facilities for CD4 count estimation may not be available.

Keywords: Tuberculosis, lymphadenopathy, HIV/AIDS, cytomorphology, CD4 counts

Introduction

The total number of people living with HIV/AIDS (PLHA) in India was estimated to be 21.17 lakhs in a survey undertaken by National AIDS Control Organisation, Ministry of Health and Family Welfare, Government of India [1]. India was estimated to have around eighty-six thousand new HIV infections and 67.6 thousand people died of AIDS-related causes in 2015 [1].

Opportunistic infections (OIs), leading to significant morbidity and mortality might grossly affect the health and quality of life of people living with HIV. Their incidence increases with the degree of immunosuppression [2]. In India, Tuberculosis (TB) is the most common

opportunistic infection in people living with HIV/AIDS and is the common cause for peripheral lymphadenopathy [3] The peripheral lymph nodes are easily accessible for fine needle aspiration cytology (FNAC), which is an office procedure for the diagnosis of tuberculosis. FNAC of tuberculous lymph nodes in HIV people supported by Ziehl-Neelsen staining, revealed four cytomorphological patterns, based on the extent of necrosis, the presence of granulomata and number of acid-fast bacilli [3]. These four patterns had indirectly reflected the immune status of people living with HIV/AIDS. The CD4 count is the primary marker of immunologic status in these individuals and used as the main criterion for initiation of Anti-Retroviral Therapy (ART) ($<$500 cells/mm^3) according to 2013 WHO guidelines [4, 5]. These CD4 counts correlate with the cytomorphological patterns of tuberculous lymphadenitis in people living with HIV/AIDS. If such a correlation exists, the FNAC patterns can be used to predict the CD4 counts where flow cytometry facilities for CD4 count estimation may not be available [3].

Aim and objective

To correlate CD4 counts with the FNAC patterns of tuberculous lymphadenitis in people living with HIV/AIDS (PLHA).

Material and Methods

This study was carried out on 121 HIV people with tuberculous lymphadenopathy, who were referred from nodal ART centre, in the department of pathology, Rangaraya medical college, Kakinada. This study was carried out over a period of 2 years (July 2016 to June 2018).

Inclusion exclusion criteria: Study included cases of cytologically diagnosed tuberculous lymphadenopathy in HIV patients. Previously diagnosed or incompletely treated tuberculous lymphadenopathy cases were excluded.
FNAC was performed under strict aseptic precautions. Material from the palpable lymph node was aspirated with the help of syringe and needle (23 G needle attached to 10 ml disposable syringe). Smears were prepared from the aspirate, fixed in alcohol and stained with hematoxylin and eosin. Air dried smears Ziehl Neelsen stain.
Different patterns of tuberculosis based on necrosis, granulomas and AFB were studied in stained smears.

Table 1: Cytomorphological patterns of tuberculous lymphadenitis in PLHA

Morphology	Distinct granuloma	Epithelioid cells	Necrosis	AFB
Pattern 1	Absent/Occasional	ILL formed	Predominant, many macrophages, Presence of neutrophils	3+
Pattern 2	Present	Well formed	Predominant	2+
Pattern 3	Prominently noted	Well formed	Present	1+
Pattern 4	Prominently noted	Well formed	Absent	1+

Results

Out of 121 cases of PLHA with tuberculous lymphadenitis pattern 1 is the most common followed by pattern 4, pattern 2 and pattern 3.
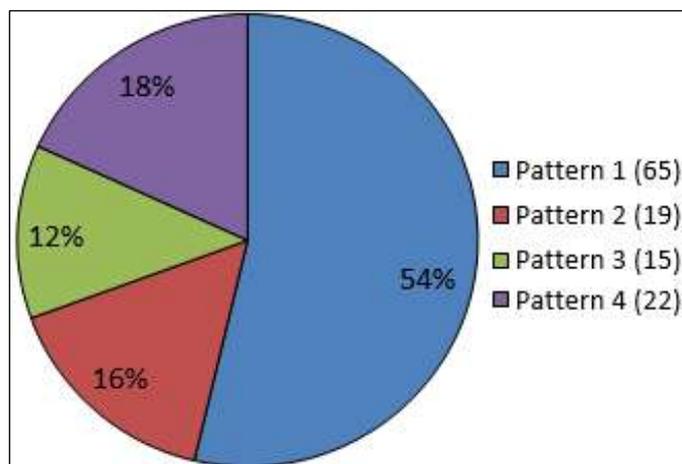
Graph 1: Cytomorphological patterns of tuberculous lymphadenitis PLHA (n=121)

Most patients were in age group of 31-40 years followed by 21-30 years. Cervical lymph nodes were the most commonly involved group followed by axillary and supraclavicular group of lymph nodes.

Table 2: Correlation of mean CD 4 counts with cytological patterns

| Morphology	No. of cases (n)	Mean CD4 counts ± 1SD
Pattern 1	54% (65)	107 ± 40
Pattern 2	16% (19)	212 ± 59
Pattern 3	12% (15)	326 ± 41
Pattern 4	18% (22)	474 ± 70
Total	n= 121	

The mean CD4 count of PLHA having pattern 1 cytomorphology was found to be 107 with a standard deviation of 40 whilst that in PLHA having pattern 4 morphology was 474 with a standard deviation of 70. The mean CD4 count of PLHA having pattern 2 and pattern 3 morphologies was 212 with a standard deviation of 59 and 326 with a standard deviation of 41 respectively.

Using ANOVA, present study was noted to had a p-value of (<0.05 is significant), which is highly significant.

In the present study we observed that CD4 counts varied significantly with different cytological patterns. Pattern 1, having a lower CD4 counts, reflects a poor immune response, whilst pattern 4, having higher CD4 counts reflected a better immune response.
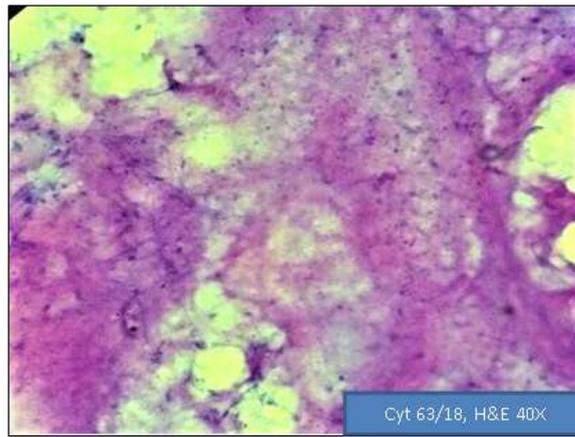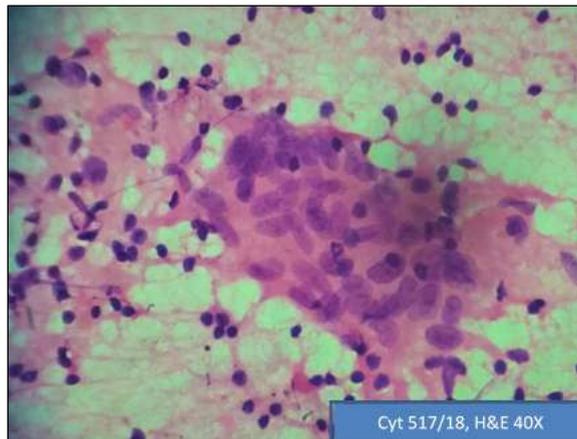
Pattern 1: Smear showing predominant caseous necrosis without granulomas
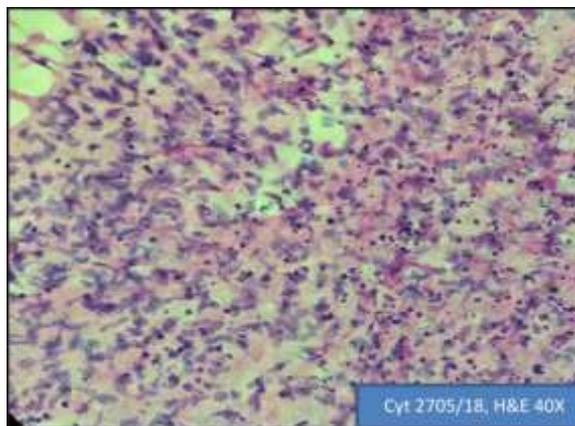
Pattern 2: Smear showing granulomas with ill-defined epithelioid cells but necrosis is still predominant
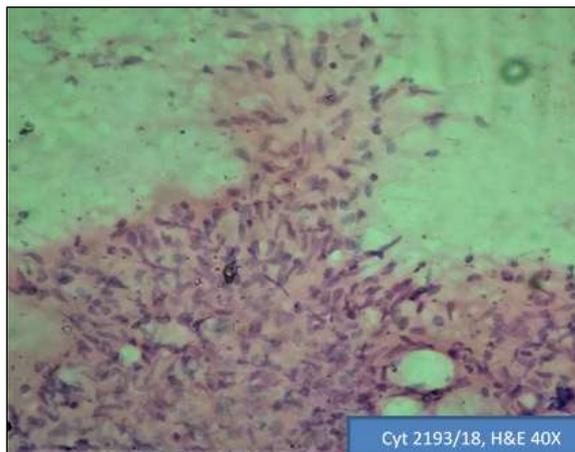
Pattern 3: Smear showing granulomas with well-formed epithelioid cells and necrosis

Cyt 517/18, H&E 40X

Pattern 4: Smear showing granulomas with well-defined epithelioid cells without necrosis

Discussion

Tuberculosis is among the most common opportunistic infections in people living with HIV/AIDS and is also the most common cause of peripheral lymphadenopathy. These lymph nodes revealed four different patterns on FNAC, which indirectly reflected the immune status in these people. CD4 counts, being a primary marker for immunological status in HIV people, were used to initiate ART, monitoring disease progression and management [6, 7].

The present study was aimed to correlate these four cytomorphological patterns with CD4 counts and compare our findings with other similar studies.

The cytomorphological patterns seen in tuberculous lymphadenitis have been categorized into 3 or 4 types, in various studies, based on the presence of caseous necrosis, epithelioid granulomas and secondary suppuration [8, 9, 10].

In the present study these patterns were categorized into 4 types similar to the study done by Rao *et al*., in people living with HIV/AIDS. Present study found that most of the cases to have pattern 1 morphology characterized by predominant necrosis, absent or occasional granulomas with ill formed epithelioid cells along with neutrophils and macrophages. Cases in whom ART was initiated, also showed predominance of pattern 1, which was often seen in severely immunocompromised individuals.

Similar predominance of pattern 1 was also observed by Nayak *et al*., Kumarguru *et al*., and Neelima *et al*., in their study on lymphadenopathy in PLHA.

Unlike the picture seen in PLHA, patients without HIV infection, often showed well defined granulomas with or without necrosis.

This reflects that immune status play a major role in determining the patterns of tuberculous lymphadenitis in PLHA.

In present study, CD4 counts were compared with four patterns of tuberculous lymphadenitis. We found significant variation in the counts among these patterns.

Table 3: Comparison of mean CD4 counts with other studies

Morphology	Mean CD4 count ±1SD (present study)	Mean CD4 counts ±1SD (Rao *et al.*) [3]	Mean CD4 counts (Kumarguru *et al.*) [8]
Pattern 1	107 ± 40 (65)	54 ± 26(10)	101.40(44)
Pattern 2	212 ± 59 (19)	137 ± 24 (10)	110.50(45)
Pattern 3	326 ± 41 (15)	343 ± 41 (10)	142.50(4)
Pattern 4	474 ± 70 (22)	466 ± 26 (10)	273.33(3)

The mean CD4 count found in pattern 1 of our study was similar to that observed by Kumarguru *et al.*, but was higher than that obtained by Rao *et al*.

The mean CD4 count in pattern 2 was much higher in our study when compared to the other two studies.

Whilst in patterns 3&4, the mean CD4 counts were similar to that observed by Rao *et al.*, they were higher than that obtained by Kumarguru *et al*.

The uniformly low values in all the four morphological patterns obtained by Rao *et al.*, in their study, could be because of inclusion of only newly diagnosed cases by them.

The cytomorphological patterns clearly reflect the immune status of the patients in their study.

We obtained higher mean values of CD4 counts in all the four categories, unlike that obtained by Rao *et al*. This could probably be due to the inclusion of cases in whom FNA was done within one month after starting ART along with newly diagnosed cases and the fact that an accelerated CD4 response is seen within the first 3 months of initiation of ART.

Decreased CD4 count leads to impairment of migration and activation of macrophages resulting in ill-defined granulomata. This explains the frequent occurrence of pattern 1 morphology in these individuals.

Conclusion

By studying the patterns of tuberculous lymphadenitis in PLHA, Cytologists can not only diagnose tuberculosis, but can also suggest the severity in CD4 count depression, and thus recommend initiation of ART in addition to ATT in these cases. This would be of great help in countries where there is a lack of facilities for CD4 count estimation.

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