Section: Pathology

Original Research Article

ROLE OF FINE NEEDLE ASPIRATION CYTOLOGY IN DIAGNOSIS OF TUBERCULOUS LYMPHADENOPATHY ALONG WITH UTILITY OF CBNAAT IN TERTIARY CARE CENTRE IN SOUTH INDIA

Ashok kumar S[1], Sanjeevitha D[2], Ravi S[3],Kalaivani S[4],Hemavathy N[5]

[1]Associate Professor, Department of Pathology, Chengalpattu Medical College, Tamilnadu, India.
[2]Assistant Surgeon, Thellar, Tamilnadu, India.
[3]Professor& Head, Department of Pathology, Chengalpattu Medical College, Tamilnadu, India.
[4]Post Graduate, Department of Pathology, Chengalpattu Medical College, Tamilnadu, India
[5]Associate Professor, Department of Pathology, Chengalpattu Medical College, Tamilnadu, India.

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Corresponding Author:
Dr. Hemavathy N,
Email: nphemaavathi@yahoo.co.in

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Abstract

Background: Tuberculosis is an important health problem in developing countries. Diagnosing Extra Pulmonary Tuberculosis (EPTB) is highly challenging due to low bacillary load and atypical presentation. Most of EPTB cases involve lymph nodes and present with lymphadenopathy. FNAC serves as first line test in diagnosing causes for lymphadenopathies but use of adjuvant technique like CBNAAT helps in confirming diagnosis and also helps in finding out sensitivity to Rifampicin. The objectives are to compare the findings of FNAC and correlation of results with CBNAAT results in clinically suspected cases of tuberculous lymphadenopathy. **Materials and Methods:** It is a descriptive cross-sectional study conducted in 100 suspected cases of tuberculosis lymphadenopathy at the Department of Pathology in Chengalpattu Medical College, Chengalpattu, Tamilnadu, for the study period of two years from October 2021 to October 2023. **Result:** In our study cytological diagnosis from FNAC showed a Sensitivity of 77.7%, Specificity of 80.82%, Positive likelihood ratio of 4.05, Negative likelihood ratio of 0.275, Positive predictive value(PPV) of 60%, Negative predictive value (NPV) of 90.76% and Percentage of false positive 19.17%,percentage of false negative 22.22%,Accuracy 80%. **Conclusion:** FNAC technique employed primarily in diagnosing suspected cases of Tuberculous lymphadenopathies showed a high sensitivity and high negative predictive value. CBNAAT being a molecular level diagnosis has advantages of identifying missed cases by FNAC and helps to find drug resistance and also for ruling out other probable causes of Granulomatous lymphadenitis. Our study showed that FNAC results done under experienced pathologist was on par with CBNAAT results in predicting extrapulmonary Tuberculous Lymphadenopathies. FNAC has further advantages of being cost effective, minimally invasive and rapid diagnostic test.

INTRODUCTION

Fine Needle Aspiration Cytology (FNAC) is widely used as a diagnostic test for the assessment of lymphadenopathy. Incidence of tuberculosis (TB) is high and it is the leading cause of lymphadenopathy. The estimated incidence of MDR-TB is 2% among new cases and 15% among previously treated cases. Superficial lymphadenopathy is the most common Extra Pulmonary manifestation of TB (EPTB) and its diagnosis is challenging because of its atypical clinical presentation, paucibacillary nature of mycobacteria at the infected sites, variation in strains of tuberculous bacteria, different levels of sensitivity of a test to specimens collected by different methods.[1] Ancillary tests like Acid fast bacilli (AFB) stain from FNAC material aid to some extent in indefinite diagnosis. Newer molecular methods like Cartridge Based Nucleic Acid Amplification Test (CBNAAT) can be done from FNAC material to confirm EPTB and act as an adjuvant for the diagnosis and can also detect Rifampicin resistant cases. Hence this study was undertaken to study the primary utilitywith adjuvant tool, CBNAAT for diagnosing suspected cases of tuberculous lymphadenopathies and also to find the

effectiveness of FNAC in diagnosing tuberculosis (EPTB)and to compare with positivity by CBNAAT technique.

MATERIALS AND METHODS

The present study is a descriptive cross-sectional study conducted at the Department of Pathology in Chengalpattu Medical College, Chengalpattu, Tamilnadu. This study was started after obtaining approval from the Institutional Ethics Committee. 100 cases of lymphadenopathy suspected to be EPTB were included in our study and under gone FNAC and CBNAAT testing during the study period of 2 years from October 2021 to October 2023. FNAC was done using 22-gauge needle / 5ml disposable syringe, under strict aseptic precautions.

In each case, part of the aspirate was used for preparing cytological smears and were stained by H&E stain. Remaining aspirated material has been mixed with reagent and sent for CBNAAT. Cytodiagnosis of tuberculous lymphadenitis was confirmed by demonstrating features of granuloma composed of epithelioid cells, Langhans giant cells with or without caseous necrosis.

For CBNAAT analysis, the sampleand reagent were added at a ratio of 3:1 with aspirated material & reagent and sent for analysis. The closed specimen container, manually agitated twice during a 15-minute period at room temperature and 2 ml of the inactivated material (equivalent to 0.5 ml of decontaminated pellet) was transferred to the test cartridge. CBNAAT purifies, concentrates, amplifies, and identifies the targeted rpoB nucleic acid sequences, and delivers the results in about 120 minutes.

Inclusion criteria being patients having clinical features of lymphadenopathy with high suspicious of extra-pulmonary tuberculosis and given consent for the study.

Exclusion criteria being Patients with features suggestive of other known causes of lymphadenopathy like lymphomas, secondary deposits, and nodal enlargement secondary to obvious infections and those who are taking anti tuberculosis treatment.

RESULTS

Our study included 100 suspected patients with clinical features of extrapulmonary tuberculosis (EPTB) predominantly involving cervical group of lymphnodes. Various cytological pattern observed in our study includes reactive lymphadenitis, Acute suppurative lymphadenitis and Granulomatous lymphadenitis.

The cytological pattern observed among granulomatous lymphadenitis was further divided in to granulomas associated with necrosis,granulomas without necrosis.Results of CBNAAT with respect to cytological parameters were also observed and tabulated. The sensitivity, specificity, predictive values and diagnostic accuracy of the CBNAAT was compared with FNAC cytodiagnosis based on cytomorphological pattern. In the present study,among 100 cases of EPTB studied,majority of the cases were in between 10-20 years age group with male to female ratio being 1:1in that age group [Table 1,2].In the present study majority of cases involved cervical lymphnode95 cases (95 %) and 3 cases(3%) showed involvement of axillary region,2cases(2%) showed involvement of supraclavicular region.

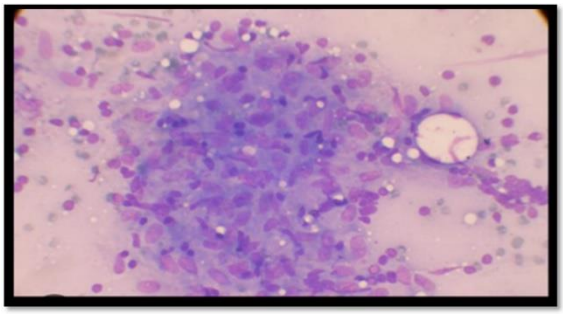

Figure 1: granulomatous lymphadenitis without necrosis [H&E 400x]

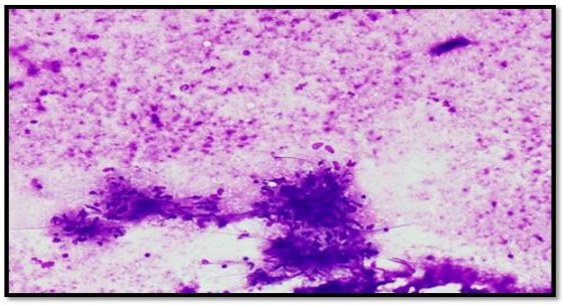

Figure 2: Granulomatous lymphadenitis with necrosis [GIEMSA 400x]

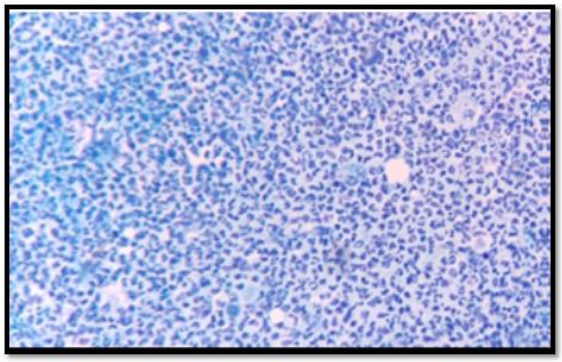

Figure 3: Suppurative lymphadenitis [H&E 400x]

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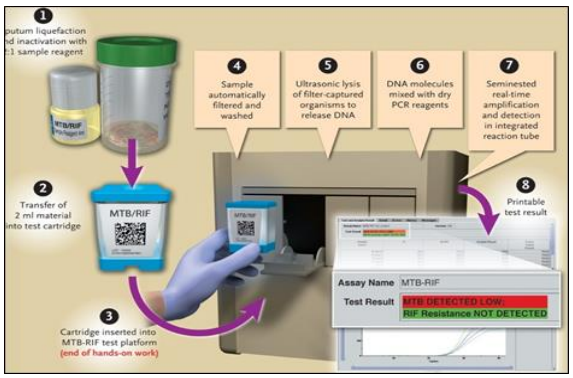

Figure 4: CBNAAT Technique

In the present study analysis of nature of aspirate showed, purulent aspirate had highest CBNAAT positivity, i.e., 34 /100cases (34%), followed by grey white tissue material and hemorrhagic aspirate each contributing 28 cases with a total of 56 cases (56%), Pus mixed with blood aspirate seen in10cases (10%) [Table 3].

Out of 100 cases, cytomorphological features (FNAC) consistent with Granulomatous lymphadenitis was noted in 35 cases (35%), Reactive lymphadenitis was diagnosed in 30 cases (30%) and Acute suppurative lymphadenitis accounted for 30cases (30%). Other 5 cases include 4 cases of malignant cytology [3cases of metastatic deposit, one Hodgkin's lymphoma] and other one case was diagnosed as pleomporphic adenoma of salivary gland [Table 4].

Table 1: age distribution of total cases

Age	No. of.Cases	Male	Female	Percentage (%)
1-20yrs	40	20	20	40%
21-40yrs	31	14	17	31%
41-60yrs	21	8	13	21%
61-85yrs	8	7	1	8%

Table 2: Sex distribution of total cases

Gender	No .of Cases	Percentage (%)
Male	43	43%
Female	46	46%
Male child	6	6%
Female child	5	5%
Total	100	100

Table 3: distribution of type of FNAC aspirates along with CBNAAT results

Type of Aspirate	No. of FNAC cases	Percentage (%)	CBNAAT(+)	CBNAAT(-)
Grey white	28	28%	7	21
Hemorrhage	28	28%	2	26
Purulent	34	34%	12	22
Pus blood	10	10%	6	4

Table 4: analysis of cytological pattern in study population (n=100)

Cytological Pattern	No .of FNAC cases	Percentage (%)
Granulomatous lymphadenitis	35	35%
Reactive lymphadenitis	30	30%
Suppurative lymphadenitis	30	30%
Others	5	5%

Table 5: analysis of different cytological pattern vs CBNAAT results (n=100)

Cytological Pattern	CBNAAT(+)	%	CBNAAT(-)	%
Granulomatous lymphadenitis	21	60%	14	40%
Reactive lymphadenitis	0	-	30	100%
Suppurative lymphadenitis	6	20%	24	80%

Table 6: distribution of cytomorphological patterns observed vs CBNAAT positivity

FNAC			CBNAAT	
Cytodiagnosis	No. of cases	%	Positivity	%
Granulomatous lymphadenitis with necrosis	14	40%	9	64%
Granulomatous lymphadenitis without necrosis	21	60%	12	58%
Suppurative lymphadenitis	30	30%	6	20%

Table 7: distribution of site of lymphadenopathy

Site of Lymphadenopathy	No. ofCases	Percentage (%)
Cervical	95	95%
Axillary	3	3%
Supraclavicular	2	2%

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Total	100	100.0

Table 8: distribution of laterality

Laterality	No. Of Cases	Percentage (%)
Unilateral	84	84%
Bilateral	16	16%
Total	100	100.0

Table 9: FNAC VS CBNAAT

FNAC	CBNAAT(+)	CBNAAT(-)
(Positive+) 35	21	14
(Negative-) 65	6	59

Table 10: comparison of cytodiagnosis in other studies

Pattern	Present study	Subhan Ali et al (11)	Bhavani et al (12)
Granulomatous Lymphadenitis	35%	39%	42%
Reactive Lymphadenitis	30%	37%	35%
Acute Suppurative Lymphadenitis	30%	7%	10%
Others	5%	17%	13%

Table 11: comparison of granulomatous pattern observed with other studies

Granulomatous pattern	Present study	Subhan Ali et al (11)	Bhavani et al (12)
Granuloma without necrosis	60%	51%	9.8%
Granuloma with necrosis	40%	34%	27%

Table 12: comparison of CBNAAT among various studies

Study	Sensitivity	Specificity
Present study	60%	90.77%
K Arpitha et al,[13]	80%	86
Komanapalli SK et al,[2]	84.25%	86.71%
Aruna L et al,[14]	65%	92.45%

Among 35 cases diagnosed as granulomatous lymphadenitis on cytology, 21 60%) cases were CBNAAT positive. Among 30 cases of suppurative lymphadenitis cases, 6 (20%) cases were positive for CBNAAT [Table 5, 6].

DISCUSSION

FNAC done in 100 suspected cases of tuberculous lymphadenopathy showed age of the patients ranged from 1years to 85yearswith increased incidence in 10-20yrs age group in concordant with study done by Komanapalli SK et al and Rock RB et al where they observed similar incidences among age groups of 11-30 yrs and 15-24 yrs respectively. M:F ratio in our study was 1:1.07 with slight female preponderance, in contrast to studies of Komanapalli SK et al and Rock RB et al., where they observed male preponderance.[2-9] In our study majority of patients presented with unilateral, single nodes 84 cases (84%), followed by16 cases (16%) showed bilateral/multiple matted nodes [Table8]. The most common site of presentation in our study was cervical region 95 cases (95%) and 3 cases(3%) showed involvement of axillary site,2cases(2%) showed involvement of supraclavicular region in concordant with study by Dr. Siddegowda et al where he also observed cervical region as most commonest site of involvement[Table 7].[10]Cytodiagnosis of Tuberculous lymphadenitis was established by demonstration of presence of granulomas composed of epithelioid cells, Langhans giant cells with or without caseous necrosis. Studies by Dr.Subhanali et al., also analysed similar cytological parameters.[11]Predominant pattern observed by FNAC in our study was granulomatous lymphadenitis which was similar to study of Subhan Ali et al., and Bhavani et al.[11,12] [Table 10]. The cytological picture in granulomatous lesion observed by FNAC in our study showed 21 cases (60%) of Granulomatous lymphadenitis without necrosis and 14 cases (40%) of Granulomatous lymphadenitis with necrosis. Similar observation was seen in study by Subhan Ali et al, and in contrast to study by Bhavani et al.[12] [Table 11].

Our study observed cytomorphological features by FNAC of 35cases were suggestive of Tuberculous lymphadenitis, of which 21 cases showed CBNAAT positivity of 60%[Table 9].While observation by Prayas et al.showed out of 80 cases,58 (72.5%) cases showed cytomorphological feature of Tuberculous lymphadenitis and 53 out of 58 cases showed CBNAAT positivity accounting to 91%correlation.[13]

In our study cytological diagnosis from FNAC showed a Sensitivity of 77.7%, Specificity of 80.82%, Positive likelihood hood ratio of 4.05, Negative likelihood ratio of 0.275, PPV of 60%, NPV of 90.76% and percentage of false positive 19.17%, percentage of false negative 22.22%,Accuracy 80%.Thus the FNAC technique employed primarily in diagnosing suspected cases of Tuberculous lymphadenopathies showed a high

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sensitivity and high negative predictive value.CBNAAT being a molecular level diagnosis has advantages of identifying FNAC missed cases, drug resistance and ruling out other probable causes of Granulomatous lymphadenitis like Churg-Strauss syndrome, Behçet disease, chronic granulomatous disease,sarcoidosis,cat-scratch disease, syphilis, leprosy, actinomycosis, rhinoscleroma, and fungal infections.

This study also describes the utility of CBNAAT in diagnosing those 6 cases (6%) with absent Tuberculous cytomorphological features and in excluding the 14 cases (14%) which presented with diagnosis of granulomatous lymphadenitis by FNAC.[13-15]On comparing with studies of sensitivity and specificity with CBNAAT testing by K Arpithaetal, Komanapalli SK et al., and Aruna L et al., the present study observed low sensitivity and high specificity [Table12].

No drug resistant case was observed in our study.

CONCLUSION

Our study observed utility of FNAC by observing various cytomorphological patterns in tuberculosis suspected patients and tried to compare its sensitivity, specificity, positive and negative predictive value with that of CBNAAT results. We observed that FNAC results done under an experienced pathologist was on par with CBNAAT results in predicting Extra Pulmonary Tuberculous Lymphadenopathies. FNAC has further advantages of being cost effective, minimally invasive and rapid diagnostic test especially when done in remote areas that lack the costlier equipment, uninterrupted electrical facility and necessity of providing ambient temperatures of cartridges and instruments for efficient working. So with a background clinical information and by visualisation with naked eye examination of nature of aspirate, it may be concluded that FNAC as atool for predicting tuberculosis etiology at the earliest. CBNAAT appears to have advantage of diagnosing paucibacillary and Drug resistance cases. Thus our study enlightens the diagnostic excellence of FNAC in clinically suspicious cases of extra pulmonary tuberculosis by being a cost effective outreach procedure aiding in rapid diagnosis and carrying a high predictable capacity equivalent to existing gold standard CBNAAT diagnostic test.

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