e-ISSN: 0975-1556, p-ISSN:2820-2643

Available online on www.ijpcr.com

International Journal of Pharmaceutical and Clinical Research 2022; 14(9); 848-855

Original Research Article

Clinical Profile of Childhood CNS Tuberculosis and Comparison of CSF-CBNAAT & Culture-Sensitivity as a Method of Diagnosis

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Received: 24-07-2022 / Revised: 23-08-2022 / Accepted: 20-09-2022

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Conflict of interest: Nil

Abstract

Introduction: Central Nervous System tuberculosis, particularly tuberculous meningitis, is the severest form of mycobacterium tuberculosis infection, causing death or severe neurological defects. The recent introduction of Cartridge based nucleic acid amplification test (CBNAAT) has significantly transformed the diagnostics of tuberculosis in adults but its application for the diagnosis of paediatric tuberculosis is under evaluation. Therefore, this study was conducted on CSF samples to compare CBNAAT and culture-sensitivity as diagnostic methods in the diagnosis of childhood CNS tuberculosis.

Methods: A prospective study was carried out between December 2018 to November 2019 consisting of 65 randomly selected patients suspected of CNS tuberculosis who had their CSF tested for CBNAAT and culture-sensitivity along with Mantoux test and other routine investigations. Statistical analysis was done using Chi square test.

Results: The sensitivity, specificity, positive predictive value and negative predictive value of CBNAAT was 70.6%, 90%, 70.6% and 89.6%, respectively.

Conclusion: CBNAAT is a rapid, sensitive, highly specific and accurate method for diagnosis of CNS tuberculosis in children. A positive CBNAAT should warrant commencement of full course of anti-tubercular therapy as per schedule. A negative CBNAAT does not necessarily exclude the diagnosis of tuberculosis and treatment should be guided based on clinical assessment of the patient as well.

Keywords: Childhood tuberculosis, Central Nervous System, CBNAAT

Introduction

Despite the ongoing tuberculosis control activities from more than 50 years, tuberculosis continues to be India's severest health crisis. Tuberculosis kills an estimated 480,000 Indians every year and more than 1,400 every day. This suffering and tragic loss of lives need to end with concerted efforts from the entire nation [1]. Central nervous system (CNS) tuberculosis, particularly Tuberculous Meningitis (TBM), is the severest form of mycobacterium tuberculosis infection, causing death or severe neurological defects in more than half of those affected, in spite of recent advancements in available anti-tubercular treatment. TBM

results in highest rates of morbidity and mortality amongst all forms of tuberculosis.

The definitive diagnosis of CNS tuberculosis depends upon the detection of Mycobacterium tuberculosis bacilli in the cerebrospinal fluid (CSF). In most settings, the diagnosis in children is traditionally based on contact tracing and very few attempts have been made for active case detection. This is mainly due to lack of pathognomonic clinical presentation in pediatric tuberculosis and lack of sensitive diagnostic tools [2-3]. Clinical diagnosis has low specificity, radiological interpretation is subjected to inter-observer variability and the tuberculin skin test is a marker of exposure, not disease.

With limited tools for confirmatory laboratory diagnosis, evaluation of medical history, chest radiography and lack of response to antibiotics help in making clinical diagnosis. Smear microscopy and microbiological confirmation are rarely achieved due to poor sensitivity of these tests owing to paucibacillary nature of the disease. At present, the diagnosis of CNS tuberculosis remains a complex issue because the most widely used conventional "gold standard" based on bacteriological detection methods, such as direct smear and culture identification, cannot rapidly detect Mycobacterium tuberculosis bacilli in CSF specimens with sufficient sensitivity in the acute phase of tuberculous meningitis. Depending on the diagnostic test used, these samples have shown variable sensitivity and specificity. It has been shown that CSF provides the highest (76-99%) detection rate, depending on sensitivity of the laboratory test adopted. Recently, instead of the conventional "gold standard", the various molecular based methods including Nucleic Acid Amplification (NAA) Assay Technique, particularly polymerase chain reaction (PCR) assay, have emerged as promising new methods for the diagnosis of CNS tuberculosis due to rapidity, sensitivity and specificity [4].

In a policy statement, World Health Organization (WHO) in 2011, recommended the use of Cartridge based nucleic acid amplification test (CBNAAT) as a preliminary diagnostic tool among children[16]. Subsequently, several workers have started using CBNAAT for the diagnosis of pediatric tuberculosis [5-8]. However, the widespread use of cartridge based nucleic acid amplification test for the diagnosis of pediatric TB on a routine basis still remains a distant option. To date, there are only a few studies on application of cartridge based nucleic acid amplification test for the diagnosis of pediatric TB. Therefore, it was planned to conduct a study on role of CSF examination in detection of Mycobacterium tuberculosis by CBNAAT and Culture & Sensitivity in the diagnosis of childhood CNS Tuberculosis with the following objectives:

1. To evaluate the role of CB-NAAT on Cerebrospinal Fluid specimen in children suspected to be suffering from CNS Tuberculosis.
2. To evaluate the correlation between Mantoux test and positivity of CB-NAAT on Cerebrospinal fluid.
3. To evaluate the correlation between Mantoux test and positivity of Culture & Sensitivity on cerebrospinal fluid.
4. To evaluate the correlation between results of Culture & Sensitivity and CB-NAAT on CSF.
5. To analyze the clinical profile of CNS Tuberculosis and compare the clinical profile and CBNAAT on CSF specimen.

Methods

A prospective hospital-based study conducted from December 2018 to November 2019 in the Department of Paediatrics, JLN Medical College and associated group of Hospitals, Ajmer. A total of 65 patients were included in the study. The study group enrolled was drawn from the patients aged < 18 years, attending the indoor and outdoor of Paediatrics Department. Patients with

clinical features of CNS TB, history of TB contact, Tuberculin skin test positivity, CSF or Neuroimaging findings suggestive of tuberculosis or history of Antiretroviral Treatment (ART) were included in study. Children aged <6 months or those already on ATT were excluded. Detailed history was obtained from all patients. Apart from routine haematological examination, Mantoux test was performed by intradermal injection of 0.1 ml purified protein derivative (PPD). CSF sample was collected and sent for biochemical analysis, CBNAAT and Culture & Sensitivity. The data was arranged in tabular form and is expressed in the form of self-explanatory tables. The data were analysed using IBM SPSS software version 22, by applying appropriate tests of significance.

Results

A total of 65 randomly selected patients of age < 18 years of either sex were included in this study. Maximum no. of cases belonged to lower middle class (40%) and upper lower class (32.3%), and only 3.1% cases were from upper class. 76.92% cases presented with Tubercular Meningitis, 4.62% with TB Vasculopathy, 3.08% with CNS Tuberculoma and 1.54% with Pott's spine & Pott's paraplegia, 9.2% with viral meningoencephalitis and 4.6 with bacterial meningitis.

History of contact was present in 56.9% of the cases. Majority of the cases (64.6%) were having BCG scar. A reactive Mantoux test was observed in 41.5% of the cases. 78.4% of the cases presented with altered sensorium, 69.2% of the cases with seizures, 50.9% of the cases with the complain of fever>2 weeks and 44.6% of the cases with meningeal irritation as shown in Table1. Neuroimaging studies (CT/MRI) were done for 46 cases which showed evidence of Hydrocephalus (32.61%), Basal Exudates (19.57%), Infarct/Vasculopathy (17.39%) or Tuberculoma (4.35%).

RpoB gene was detected by CBNAAT in 16 out of 45 (35.6%) children who had Seizure as presenting symptom as shown in table2. In the absence of seizure, RpoB gene was detected by CBNAAT in only 1 (5%) case. The difference was statistically significant. RpoB gene was detected by CBNAAT in 15 out of 51 (29.4%) children who had altered sensorium as shown in table3. In the absence of altered sensorium, RpoB gene was detected by CBNAAT in 2 (14.3%) cases. The difference was statistically non-significant. RpoB gene was detected by CBNAAT in 14 out of 33 (42.4%) children with fever as shown in table4. In the absence of fever, RpoB gene was detected by CBNAAT in 3 (9.4%) cases. The difference was statistically significant. RpoB gene was detected by CBNAAT in 7 out of 42 (16.6%) children with BCG scar as shown in table5. In the absence of BCG Scar, RpoB gene was detected by CBNAAT in 10 (43.5%) cases. The difference was statistically significant. RpoB gene was detected by CBNAAT in 16 out of 37 (42.2%) children with positive history of contact as shown in table6. In cases where there was no history of contact, RpoB gene was detected by CBNAAT in only 1 (5.9%) case. The difference was statistically significant. RpoB gene was detected by CBNAAT in 10 out of 27 (37.03%) children with positive Mantoux test and in 11 out of 38 (18.42%) cases with negative Mantoux test as shown in table7. The difference was statistically non-significant.

Culture and sensitivity were positive in 12 out of 42 (28.6%) children with BCG scar as shown in table8. In the absence of BCG scar, culture and sensitivity was positive in 5 (21.7%) cases. The difference was statistically not significant. Culture and sensitivity were positive in 13 out of 37 (35.1%) children with positive history of contact as shown in table9. In cases where there was no history of contact, culture and sensitivity was positive in 4 (14.3%) cases. The difference was statistically non-significant. Culture and sensitivity were positive in 8 out of 27 (29.6%) children

with positive Mantoux test and in 9 out of 38 (23.7%) cases with negative Mantoux test as shown in table10. The difference was statistically non-significant.

The difference between CBNAAT and culture-sensitivity was found to be statistically significant as shown in table 11. The sensitivity, specificity, positive predictive value and negative predictive value of CBNAAT was 70.6%, 90%, 70.6% and 89.6%, respectively as shown in table 12.

Table 1: Complaints at the time of presentation

S. No.	Complaints	Status	No. of Patients	Percentage
1.	Fever >2 weeks	Yes	33	50.8
		No	32	49.2
2.	Meningeal irritation	Yes	29	44.6
		No	36	55.7
3.	Altered sensorium	Yes	51	78.4
		No	14	21.6
4.	Headache	Yes	28	43.1
		No	37	56.9
5.	Seizures	Yes	45	69.2
		No	20	30.8
6.	Decreased appetite	Yes	29	44.6
		No	36	55.4
7.	Weight loss / no weight gain	Yes	34	52.3
		No	31	47.7
8.	Nausea/Vomiting	Yes	30	46.1
		No	35	53.9

Table 2: Association of CBNAAT and Seizure as presenting complaint

		H/O SEIZURE		Total
		Yes	No	
CBNAAT	Detected	16	1	17
	Not Detected	29	19	48
p value: 0.0096 (S)				

Table 3: Association of CBNAAT and Altered Sensorium as presenting complaint.

		ALTERED SENSORIUM		Total
		Yes	No	
CBNAAT	Detected	15	2	17
	Not Detected	36	12	48
p value: 0.2540 (NS)				

Table 4: Association of CBNAAT and Fever as presenting complaint.

		FEVER > 2 WEEKS		Total
		Yes	No	
CBNAAT	Detected	14	3	17
	Not Detected	19	29	48
p value: 0.0024 (S)				

Table 5: Association of CBNAAT and BCG Scar

		BCG SCAR		Total
		Present	Absent	
CBNAAT	Detected	7	10	17
	Not Detected	35	13	48
p value: 0.0187 (S)				

Table 6: Association of CBNAAT and History of contact

		H/O CONTACT		Total
		Yes	No	
CBNAAT	Detected	16	1	17
	Not Detected	21	27	48
p value: <0.001 (S)				

Table 7: Association of CBNAAT and Mantoux test

		Mantoux Test		Total
		Positive	Negative	
CBNAAT	Detected	10	7	17
	Not Detected	17	31	48
p value: 0.0924 (NS)				

Table 8: Association of culture & sensitivity for MTB and BCG Scar

		BCG SCAR		Total
		Present	Absent	
Culture & Sensitivity	Detected	12	5	17
	Not Detected	30	18	48
p value: 0.5490 **(NS)**				

Table 9: Association of culture & sensitivity for MTB and History of contact

		H/O CONTACT		Total
		Yes	No	
Culture & Sensitivity	Detected	13	4	17
	Not Detected	24	24	48
p value: 0.05822 **(NS)**				

Table 10: Association of culture & sensitivity for MTB and Mantoux test

		Mantoux Test		Total
		Positive	Negative	
Culture & Sensitivity	Detected	8	9	17
	Not Detected	19	29	48
p value: 0.5909 **(NS)**				

Table 11: Association of CBNAAT and Culture-Sensitivity

		CBNAAT		Total
		Positive	Negative	
Culture & Sensitivity	Positive	12	5	17
	Negative	5	43	48
p value: <0.001 **(S)**				

Table 12: Sensitivity, Specificity, Positive Predictive Value and Negative Predictive Value of CBNAAT

Sensitivity	70.6%
Specificity	90 %
Positive Predictive Value	70.6 %
Negative Predictive Value	89.6 %

Discussion

Our study showed a sensitivity of 70.6% and specificity of 90 % for Cartridge Based Nucleic Acid Amplification Test (CBNAAT) on CSF samples of children suspected to be suffering from CNS tuberculosis. Positive Predictive Value (PPV) and Negative Predictive Value (PPV) were 70.6% and 89.6% respectively (Table no.12).

In a previous study by Deepa Kumari *et al.* (2018) [9]**,** among 53 patients, who were included in the study period, 5 patients were culture positive and 48 were culture negative for M. tuberculosis. GeneXpert MTB/RIF test showed a sensitivity of 60% and specificity of 97.9% for diagnosing mycobacterium tuberculosis, the negative predictive value was 95.9% and positive predictive value was 75% [9].

In a study by Amit Singh *et al*. (2016) [10], among 75 clinically suspected cases, 6 (8.0%) belonged to 'Definite' TBM group and 69 (92.0%) belonged to 'Probable' TBM group. Out of total 75 cases, 6 CSF samples were positive by culture on LJ medium. CSF from control group was all culture negative. Overall, LJ culture had Sensitivity, Specificity, PPV and NPV of 8%, 100%, 100% and 33.01% respectively and overall, nPCR had Sensitivity, Specificity, PPV, NPV and diagnostic accuracy of 89.34%, 97.06%, 98.53%, 80.5% and 91.74% respectively [10].

In a study by RS Solomons *et al*. (2015)[11], Xpert MTB/RIF assay of 101 meningitis suspects, 55 were diagnosed with TBM. Culture, Geno Type and Xpert combined performed best with 56% Sensitivity and 98% Specificity [11].

In a study by Sheena Gupta *et al*. (2016) [12] a total of thirty-three subjects were enrolled in the study of which 18 were in Group I and 15 were in Group II. The sensitivity and specificity of TB PCR for diagnosis of TBM was 77.78% & 100%, respectively and positive predictive value was 100% and the negative predictive value was 78.95% [12]. Comparison of sensitivity and specificity of CBNAAT from CSF sample with other studies is shown in table13.

Table 13: Comparative data showing Sensitivity and Specificity of Cartridge Based Nucleic Acid Amplification Test (CBNAAT) from CSF Sample

Study	Sensitivity	Specificity
Arzu *et al.* 2011[13]	70	100
Deepa Kumari *et al.* 2018 [9]	60%	97.9%
Nhu *et al*. 2014[14]	59	99
Pai *et al*. 2003[15]	56	98
Sheena Gupta *et al.* 2016[12]	77.78%	100%
Solomon *et al.* 2015[11]	56%	98%
Present Study	70.6%	90%

The findings in our study were in accordance with the previous studies. Not many studies are available which define the positive association between Culture &

Sensitivity and CBNAAT. The studies have mainly been descriptive and have not looked at many significant factors.

The paucibacillary nature of samples from young children partly explains the lower sensitivity of culture than that observed in older children. People with low socioeconomic status typically live in poor housing and environmental conditions, have greater food insecurity and have less access to quality health care relative to those from higher socioeconomic groups. All of these social determinants related to tuberculosis often work together to put the poor at greater risk of disease by acting on different stages in the pathogenesis pathway.

A history of tuberculosis exposure increases the likelihood of occurrence of tuberculosis in a child with suspected CNS tuberculosis. A positive Tuberculin sensitivity test response is a marker of tuberculosis exposure and also increases the likelihood of occurrence of tuberculosis in a child with suspected CNS tuberculosis.

Conclusion:

Our study prospectively assessed Cartridge Based Nucleic Acid Amplification test for the rapid diagnosis of childhood tuberculosis using CSF samples in a high tuberculosis endemic setting. The study has two key findings:

- Cartridge Based Nucleic Acid Amplification Test performs well in rapid, accurate and highly specific diagnosis of childhood tuberculosis.
- Results of CBNAAT relate better with clinical parameters like history of contact and presence of BCG scar (statistically significant results) while the same parameters did not relate with the results of CSF Culture and Sensitivity.

In a nutshell, CBNAAT is a rapid, sensitive, highly specific and accurate method for diagnosis of CNS tuberculosis in children. A positive CBNAAT should warrant commencement of full course of anti-tubercular therapy as per schedule. A negative CBNAAT does not necessarily exclude the diagnosis of tuberculosis and treatment should be guided based on clinical assessment of the patient as well.

References

1. Revised National Tuberculosis Control Programme, National strategic plan for Tuberculosis elimination 2017–2025; March 2017. Central TB division, directorate general of health services, Ministry of Health with Family Welfare, Nirman Bhavan, New Delhi – 110 108.
2. Zar HJ, Hanslo D, Apolles P, Swingler G, Hussey G. Induced sputum versus gastric lavage for microbiological confirmation of pulmonary tuberculosis in infants and young children: a prospective study. Lancet. 2005; 365:130–4.
3. WHO (2011a) WHO Report Global Tuberculosis Control. WHO, Geneva.
4. Nicol MP, Workman L, Isaacs W, Munro J, Black F, Eley B, et al. Accuracy of the Xpert MTB/RIF test for the diagnosis of pulmonary tuberculosis in children admitted to hospital in Cape Town, South Africa: a descriptive study. Lancet Infect Dis. 2011; 11:819–24.
5. Bates M, O'Grady J, Maeurer M, Tembo J, Chilukutu L, Chabala C, *et al*. Assessment of the Xpert MTB/RIF assay for diagnosis of tuberculosis with gastric lavage aspirates in children in sub-Saharan Africa: a prospective descriptive study. Lancet Infect Dis. 2013; 13:36–42.
6. Sekadde MP, Wobudeya E, Joloba ML, Ssengooba W, Kisembo H, Bakeera-Kitaka S, *et al*. Evaluation of the Xpert MTB/RIF test for the diagnosis of childhood pulmonary tuberculosis in Uganda: a cross-sectional diagnostic study. BMC Infect Dis. 2013; 13:133.
7. Nhu NTQ, Ha DTM, Anh ND, Thu DDA, Duong TN, Quang ND, *et al*. Evaluation of Xpert MTB/RIF and

MODS assay for the diagnosis of pediatric tuberculosis. BMC Infect Dis. 2013; 13:31.
8. K. S. Sahana, Anitha S Prabhu, Prakash RM Saldanha. Usage of Cartridge Basesd Nucleic Acid Amplification Test (CB-NAAT/ GeneXpert) test as diagnostic modality for pediatric tuberculosis; case series from Manglore, south India. http://doi.org/10.1016/j.jctube.2017.12.002
9. Deepa Kumari, Shampa Anupurbha, Manish Gupta, Sarita Kumari, & Anup Singh. (2018). Correlation of GeneXpert and cerebrospinal fluid culture in patients of tubercular meningitis. Asian Pacific Journal of Health Sciences, 5(2), 148–151.
10. Amit Singh *et al*., Rapid Diagnosis of Pediatric TBM by Nested PCR National Journal of Laboratory Medicine. 2016 Jul, Vol-5(3): MO05-MO10.
11. RS Solomons, DH Visser, SO Friedrich, AH Diacon, KGP Hoek, BJ Marais, JF Schoeman, AM van Furth, Improved diagnosis of childhood tuberculous meningitis using more than one nucleic acid amplification test, The International Journal of Tuberculosis and Lung Disease 19 (1), 74-80, 2015.
12. Gupta S, Gupta A, Wasim S, Kotwal A, Bhat NK. A Study of Polymerase Chain Reaction in Cerebrospinal Fluid for Diagnosis of Tuberculous Meningitis. Ntl J Community Med 2016; 7(3):212-215.
13. Zeka AN, Tasbakan S, Cavusoglu C. Evaluation of the GeneXpert MTB/RIF assay for rapid diagnosis of tuberculosis and detection of rifampin resistance in pulmonary and extrapulmonary specimens. J Clin Microbiol 2011; 49:4138- 41.
14. Nhu NT, Heemskerk D, Thu DD, Chau TT, Mai NT, Nghia HD, *et al*. Evaluation of GeneXpert MTB/RIF for diagnosis of tuberculous meningitis. J Clin Microbiol 2014; 52:226-33.
15. Pai M, Flores LL, Pai N, Hubbard A, Riley LW, Colford JM. Diagnostic accuracy of neucleic acid amplification tests for tuberculous meningitis: A systematic review and meta-analysis. Lancet Infect Dis 2003; 3:633-43.