

A Study of Bacterial and Viral Meningitis in Pediatric Age Group in a Tertiary Care Hospital

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ABSTRACT

Background: Meningitis, the inflammation of meninges is one of the most severe and potentially life-threatening infectious diseases. Young children are mostly at risk, but clinical severity also depends on the causative organism. Viral meningitis is underdiagnosed and under-notified. Objectives of the study are to determine the frequency of bacterial and viral meningitis and the antibiotic susceptibility pattern of bacterial isolates

Methodology: A cross-sectional study was conducted on the CSF samples collected from children ≤ 14 years of age within a 2-years study period (August 2022 to July 2024) admitted with suspected meningitis in Pediatric department of SCSMSR, Solapur. Part of the sample was subjected to Gram staining and inoculated on chocolate agar, blood agar and MacConkey agar. Bacterial isolates were identified by biochemical reactions and antibiotic susceptibility test by

disk diffusion. Multiplex PCR (TRUPCR[®] Neuro Panel Kit) was employed for viral study with the second part of sample. Analysing the distribution of isolates and sensitivity pattern of bacterial isolates were done using Microsoft Excel.

Results: Out of 114 suspected cases, 13 (11.41%) were culture and smear positive for pyogenic meningitis and 17 (14.91%) showed viral etiology by RT-PCR. Gram positive isolates (55.56%) were more common among the bacterial isolates. Our study detected seven different types of viruses among which the most common were CMV and HSV1 (both 23.53%).

Conclusion: Meningitis is a medical emergency and hence early diagnosis and treatment, are lifesaving. Since clinical signs of meningitis are not always reliable, a laboratory support is imperative, to achieve an early diagnosis.

Keywords: meningitis, bacterial, viral, pediatric, CSF, PCR.

INTRODUCTION

The epidemiology of bacterial along with other kinds of meningitis has shifted significantly over this past century. The use of conjugate vaccines during the last few decades has radically transformed the incidence of the disease in high-income nations, resulting in the 'near elimination' of *Haemophilus influenzae* type b [Hib] meningitis and serogroup type C meningococcal meningitis. [1] Among bacterial causes, the previously infrequent group B Streptococcus (GBS) is now the most predominant pathogen causing purulent meningitis. [2] Despite breakthroughs in vaccine development and chemoprophylaxis, meningitis continues to be a leading cause of death and long-term neurological disability due to antibiotic resistance. Meningitis and its associated complications can be avoided with timely diagnosis and treatment. [3]

The disease imposes a significant worldwide burden. Other than epidemics, no less than 1.2 million cases of meningitis are anticipated each year, with 135000 deaths worldwide. [4,5] The mortality rate in India and other poor countries ranges between 16 to 32%. [6] Severe clinical manifestations are associated with high morbidity and mortality. [7,8] Because of poor educational facilities and almost no emphasis on facilitating a normal life, the impact of complications in children due to meningitis in countries such as India differs significantly from that in developed nations. [9]

A new international response to meningitis, WHO's Defeating Meningitis by 2030 Global Roadmap is now in place. Over the past few years, the expansion of meningitis-related vaccination programs for children under the age of five, increased research and intervention efforts, and fostering support on behalf of meningitis survivors along with their families have presented numerous possibilities for both meningitis prevention and improved quality of life for survivors. [10]

Meningitis is a medical emergency, and anyone suspected of having the disease should be evaluated and treated urgently. Delays in diagnosis and treatment can lead to a poor disease outcome. [11] The bacteria commonly causing meningitis in pediatric population are *H. influenzae*, *Neisseria meningitidis*, *Streptococcus pneumoniae*, Group B Streptococcus. [2] The most common viral agents of meningitis are non-polio enteroviruses (group b coxsackievirus and echovirus). Other viral causes: mumps, Parechovirus, Herpesviruses (including Epstein Barr virus, Herpes simplex virus, and Varicella-zoster virus), measles, influenza, and arboviruses (West Nile, La Crosse, Powassan, Jamestown Canyon). [12] CMV meningitis is an infrequent manifestation which can be difficult to discern in neonates who are preterm. [13]

Understanding the prevalence of meningitis in children is critical for developing effective public health policies, improving treatment protocols, and implementing preventive measures. Accurate data on incidence rates can help healthcare providers and policymakers allocate resources more effectively, potentially reducing the disease's burden.

MATERIALS & METHODS

Study design: A prospective observational cross-sectional study was carried out in the Microbiology department of Tertiary care hospital in Solapur, Maharashtra.

Study duration: 02 years Study was carried out from August 2022 to July 2024

Study population: Children ≤ 14 years of age, who were admitted to the tertiary care hospital, with signs and symptoms of meningitis.

Inclusion criteria- Patients suspected of meningitis, ≤ 14 years of age and who were admitted in the hospital.

Exclusion criteria- Patients who have neurological symptoms but are known case of seizure disorder or with brain space occupying lesion or with hydrocephalus with history of shunt surgery.

Sample size: The study comprised CSF samples received in the Microbiology department over a one-year period, from August 2022 to July 2023, that were suspected of meningitis. Meningitis was diagnosed based on a CSF examination, culture, and/or other relevant assays conducted during the patient's hospitalization. Following the application of inclusion and exclusion criteria, 114 study subjects were identified as eligible and included in our study. All CSF samples were tested for both bacterial and viral meningitis etiological agents.

Processing:

The 3-5 ml of CSF samples in sterile leak proof container of patients were received in the Microbiology department of Dr. VMGMC, Solapur for suspected meningitis were included in study after taking consent from their parents. Socio-demographic data, clinical history, immunization and relevant history were obtained from them. The volume and gross appearance i.e., consistency, presence of blood and the colour of CSF were noted. The sample was divided into three parts. The first part was used for bacteriological study.

A few drops of CSF were placed on a clean glass slide, then air dried and heat fixed for Gram staining. CSF samples were either heaped up or centrifuged and Gram staining was carried out on them, then observed under oil immersion objective of microscope. The gram-stained smear was assessed for presence of pus cells and microorganisms. Loopful of CSF was inoculated on chocolate agar, blood agar and MacConkey agar and incubated at 37°C for 48 hours in 5-10% CO₂. The candle jar was used for achieving 5-10% of CO₂. The second part of the sample was incubated as it is at 37°C. This sample was used for culture when the previous culture yielded no growth even after 48 hours of incubation. The growth obtained was identified by Gram stain and conventional biochemical tests.

Antibiotic sensitivity testing (AST) was performed on Mueller–Hinton agar plates by Kirby–Bauer disc diffusion method. For Gram positive catalase negative organisms blood agar plates were used for AST and colonies were swabbed after preparation of suspension in normal saline.

Gram negative organisms were inoculated in peptone water broth and incubated for two hours at 37°C. Turbidity of the peptone water broth was compared to 0.5 McFarland standard against a Wickerham card and lawn cultured on Mueller-Hinton agar plates using sterile swabs. Antibiotic discs were placed on these plates and they were incubated at 37°C for 18 hours. Readings were taken by measuring diameter with the help of Vernier callipers and interpreted as per the Clinical and Laboratory Standards Institute (CLSI) guidelines, of the year 2022 and 2023.

The various antibiotic discs used for AST of gram-positive organisms were Ampicillin (10µg), Chloramphenicol (30µg), Cotrimoxazole (1.25/23.75µg), Gentamicin (10µg), Linezolid (30µg), Levofloxacin (5µg), Oxacillin (1µg), Penicillin (10 units) and Vancomycin (30 µg- except in *Staphylococcus aureus*)

Antibiotic discs used for AST of gram-negative organisms were Amikacin (30µg), Ampicillin (10µg), Amoxicillin/Clavulanate (20/10 µg), Aztreonam (30µg), Chloramphenicol (30ug), Cefotaxime (30µg), Ceftriaxone (30µg), Cotrimoxazole (1.25/23.75µg), Gentamicin (10µg), Meropenem (10µg), Levofloxacin (5µg) and Piperacillin/Tazobactam (100/10 µg).

Methicillin resistance in *Staphylococcus aureus* (MRSA) was tested using Mueller-Hinton agar with Cefoxitin disc (30 µg) by Kirby-Bauer disc diffusion method. A zone size of ≥ 22 mm was considered methicillin sensitive and ≤ 21 was considered methicillin resistant.

High level Aminoglycoside resistance in *Enterococcus species* was tested using High level Gentamicin disc (120 µg).

Vancomycin susceptibility in MRSA isolates was measured with Vancomycin Ezy MIC™ strip (VAN) (0.016-256 µg/ml) by Epsilon meter test.

AST of Penicillin (0.016-256 µg/ml), Cefotaxime (0.016-256 µg/ml), Ceftriaxone (0.016-256 µg/ml) in *Streptococcus pneumoniae* isolates were also measured with E strips (Ezy MIC™ strip) by Epsilon meter test.

The third part of the CSF in the study was used for viral diagnosis. Samples were stored in a deep freezer (at -20°C). Multiplex PCR (TRUPCR® Neuro Panel Kit) was employed for diagnosis of 11 different viruses namely Human adenovirus, Enterovirus and Human parechovirus, Herpes simplex virus 1, Herpes simplex virus 2, Human parvovirus B19, Epstein-Barr virus, Varicella Zoster virus, Human Cytomegalovirus, Human Herpes virus 6 and Human Herpes virus 7.

For each sample four separate reaction mixtures were made-

- **Tube 1-** Human adenovirus, Enterovirus and Human parechovirus.
- **Tube 2-** Herpes simplex virus 1, Herpes simplex virus 2 and endogenous Internal control (IC).
- **Tube 3-** Human parvovirus B19, Epstein-Barr virus, Varicella Zoster virus.
- **Tube 4-** Human Cytomegalovirus, Human Herpes virus 6, Human Herpes virus 7.

Preparation of reaction mixture-

Name of the Reagent	Tube (Volume in µl)			
	1	2	3	4
Master mix	10	10	10	10
Primer Probe Mix-1	5	-	-	-
Primer Probe Mix-2	-	5	-	-
Primer Probe Mix-3	-	-	5	-
Primer Probe Mix-4	-	-	-	5
Total Reaction volume	15	15	15	15

Transfer of 15µl of this prepared reaction mix was done to 0.2 ml PCR tubes and then the tubes were closed. To this 10 µl of extracted nucleic acid or positive control (for all four tubes) or negative control (sterile water) was added to make a final volume of 25 µl.

Program was set up for temperature profile- 50°C for 15 minutes → 94°C for 10 minutes → 94°C for 15 seconds → 60°C for 45 seconds → 72°C for 15 seconds.

Channel selection setting was done for detection of reporter dyes like FAM/Green, HEX/Yellow and Tex Red/Orange for various viruses in each tube.

Result analysis was done at the end of 35 cycles (threshold cutoff cycle Ct) but the assay ran for 38 cycles. Presence of channel signal in any particular tube indicates the presence of genetic material of that particular virus.

CSF cytology results and CSF biochemical analysis of the samples were obtained from Pathology and Biochemistry departments respectively.

Statistical Analysis

Microsoft Excel 2023 and SPSS were used to analyse the distribution of various bacterial isolates, their sensitivity patterns, and viral etiological agents, as well as to interpret the data. The results were presented as frequency, percentages, mean, median, and mode.

RESULT

During the study period 114 CSF samples were analyzed, with one sample representing each patient.

Demographic characteristics:

In our study, majority i.e. 26.32% patients were from 1 Month to 1 Year age group followed by >5 Years to 10 Years and >1 Year to 5 Years age groups as shown in Table 1.

Table 1: Age wise demographical distribution of patients.

Age Group	No. of Patients(N)
<1 Month	22 (19.30%)
1 Month to 1 Year	30 (26.32%)
>1 Year to 5 Years	26 (22.81%)
>5 Years to 10 Years	27 (23.68%)
>10 Years to 14 Years	09 (07.89%)
Total	114 (100%)

Table 2 shows the positivity rate of meningitis among female patients was marginally more as compared to male children.

Table 2: Distribution of patients according to their gender.

Gender	Total specimens tested	Microbiologically diagnosed cases
Male	51(44.73%)	13 (25.49%)
Female	63 (55.27%)	17 (26.98%)
Total	114 (100%)	30 (26.32%)

Symptoms and signs on presentation:

All the patients were admitted with multiple signs and symptoms. The most common clinical presentation observed in our study

was vomiting (81.57%), followed by fever (77.19%) and convulsion (64.91%) as demonstrated in Table 3.

Table 3: Distribution of patients according to their symptoms.

Clinical Presentation	No. of patients (N=114)
Vomiting	93 (81.57%)
Fever	88 (77.19%)
Convulsions	74 (64.91%)
Irritability	51 (44.73%)
Crying	38 (33.33%)
Bulging fontanelle	17 (14.91%)
Neck rigidity	17 (14.91%)
Weight loss	05 (04.38%)

Types of meningitis:

Out of 114 cases, majority of patients were unproven cases of meningitis (73.68%). In present study, the frequency of bacterial

meningitis and viral meningitis was 11.41% and 14.91% respectively as shown by Figure 1.

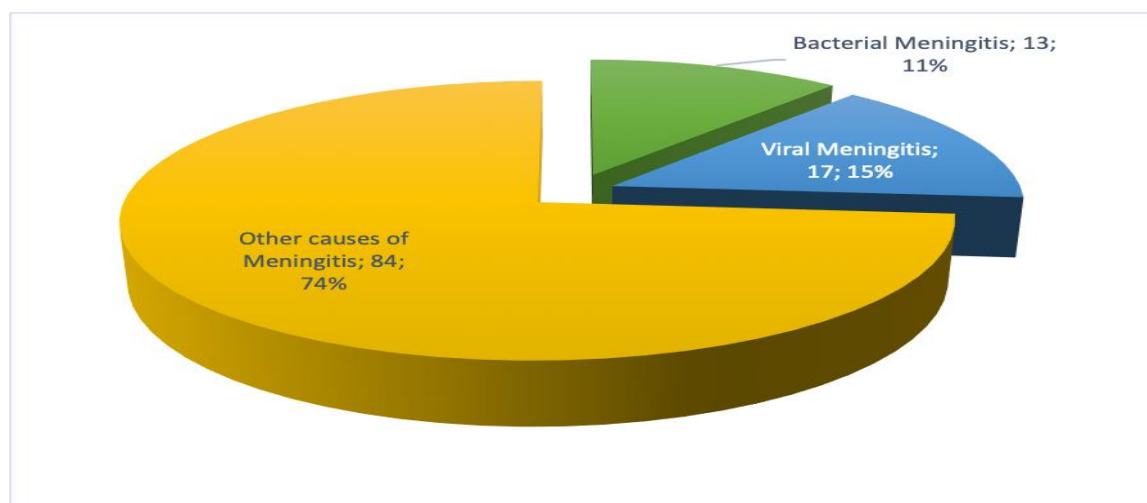


Figure 1: Distribution of patients according to frequency of bacterial and viral meningitis

All the viral meningitis cases were negative for bacterial culture. All the bacterial meningitis cases were negative for viruses by Multiplex PCR. Our study shows that bacterial meningitis was common in 10 year to 14 year age group (22.22%) while viral meningitis was common in 1 month to 1 year age group (23.33%). Overall highest

positivity of meningitis is shown by patients in the age group 1 month to 1 year (36.67%) followed by those in > 10 years to 14 years (33.33%) and > 1 Year to 5 Years (19.23%). In the present study, all (100%) the culture positive isolates were also positive by direct Gram staining.

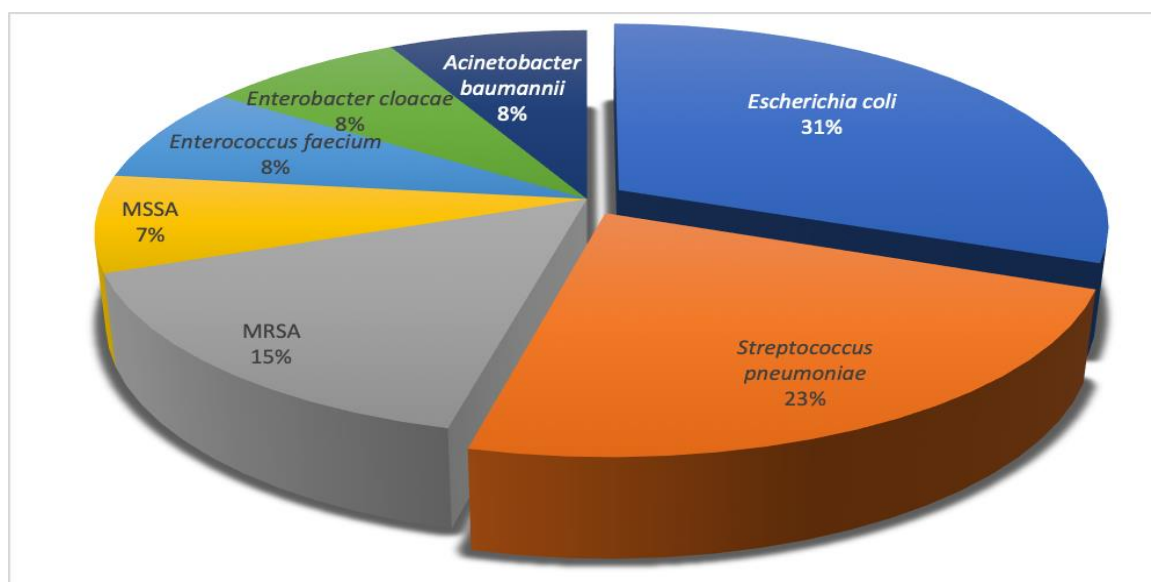


Figure 2: Distribution of patients according to etiological agents in bacterial meningitis.
Abbreviations: MRSA- Methicillin Resistant *Staphylococcus aureus*,
MSSA- Methicillin Sensitive *Staphylococcus aureus*.

Figure 2 shows that *Escherichia coli* (30.77%) was the most common bacterial agent in the present study, followed by *Streptococcus pneumoniae* (23.08%) and Methicillin Resistant *Staphylococcus aureus* (MRSA,15.39%). Each of these pathogens contributes significantly to the total number of cases. The other bacteria listed

(*Enterococcus faecium*, MSSA, *Enterobacter cloacae*, and *Acinetobacter baumannii*) each account for a smaller proportion (07.69%) of the total cases, representing the diversity of bacterial agents involved in meningitis infections. Only single organism was isolated in each of the positive specimens by culture.

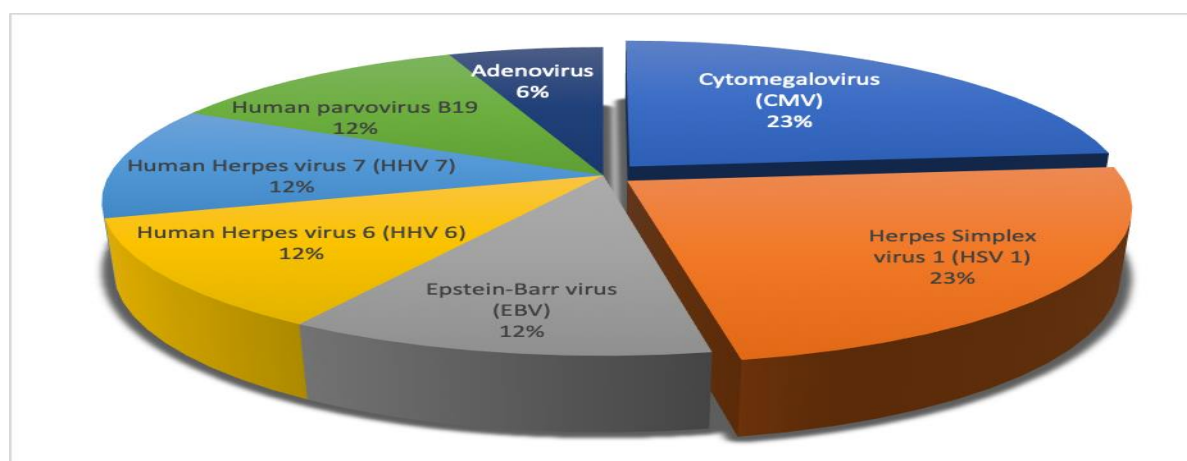


Figure 3: Distribution of patients according to positivity rate of viral meningitis

In patients with viral meningitis, most common viruses identified were CMV and HSV 1 each accounting for 23.53% as demonstrated by Figure 3. Other important etiological agents found in viral meningitis were EBV, HHV6, HHV7, Human Parvovirus B19 and Adenovirus. In the present study, Herpes virus group (82.35%) was found to be the most common cause of viral meningitis. Besides Herpes viruses other two viruses causing meningitis were Adenovirus and Human Parvovirus B19.

Only single virus was detected by PCR in each of the viral meningitis case.

CSF cytology and biochemistry:

Table 4 shows Differential leukocyte count (DLC) of CSF samples where neutrophils were predominant in bacterial meningitis ranging from 60-100% with mean $81.15\% \pm$ SD (standard deviation) of 11.92% , while lymphocytes were predominant in viral meningitis ranging from 0-100% with mean $82.35\% \pm$ SD of 31.92% .

Table 4: Correlation between microbiological finding and CSF Cytology in the present study.

Parameters in CSF Cytology (DLC)	Bacterial Meningitis	Viral Meningitis
Lymphocytes (%)	0-40	0-100
Range (Mean $\pm$ SD)	(18.84 ± 10.93)	(82.35 ± 31.92)
Median (Min, Max)	20 (10, 30)	90 (80, 100)
Neutrophils (%)	60-100	0-20
Range (Mean $\pm$ SD)	(81.15 ± 11.92)	(05.88 ± 07.95)
Median (Min, Max)	80 (70, 90)	0 (0, 10)

For CSF protein levels in our study, bacterial meningitis cases exhibited a wide range from 45 to 1400 mg/dl, with a median of 126 mg/dl and an IQR of 360 mg/dl, indicating significant variability and generally higher protein levels. In contrast, viral meningitis cases had a narrower range from 10 to 89 mg/dl, with a mean of 56 mg/dl and an IQR of 23 mg/dl, reflecting lower and more consistent protein levels.

In case of CSF sugar levels, bacterial meningitis cases had a range from 12 to 88 mg/dl, with a median of 23 mg/dl and an IQR of 15 mg/dl, demonstrating lower sugar levels compared to viral meningitis. Viral meningitis cases showed a sugar range from 33 to 67 mg/dl, with a median of 45 mg/dl and an IQR of 20 mg/dl, indicating relatively higher and more stable sugar levels.

Table 5: Distribution of bacterial and viral meningitis according to CSF Biochemistry in the present study.

Parameters in CSF Biochemistry	Bacterial Meningitis	Viral Meningitis
PROTEIN (mg/dl)	45-1400	10-89
Range (Mean $\pm$ SD)	(310.15 ± 379.88) mg/dl	(55.47 ± 21.17) mg/dl
Median (Min, Max)	126 (90, 450)	56 (45, 68)
SUGAR (mg/dl)	12-88	33-67
Range (Mean $\pm$ SD)	(27.38 ± 20.60) mg/dl	(48.29 ± 14.46) mg/dl
Median (Min, Max)	23 (14, 29)	45 (37, 57)
CHLORIDE (mEq/L)	110-189	108-137
Range (Mean $\pm$ SD)	(142.69 ± 24.90) mEq/L	(123 ± 09.17) mEq/L
Median (Min, Max)	144 (119, 156)	123 (119, 131)

In case of CSF chloride levels, bacterial meningitis cases had a range from 110 to 189 mEq/L, with a median of 144 mEq/L and an IQR of 37 mEq/L. In viral meningitis, the chloride levels ranged from 108 to 137 mEq/L, with a median of 123 mEq/L and an IQR of 12 mEq/L, showing

slightly lower and less variable chloride levels compared to bacterial meningitis.

Antibiotic susceptibility test:

In our study, both of the MRSA isolates were Vancomycin and Linezolid sensitive as shown in Table 6. MSSA was susceptible

to all antibiotics except for resistance to Penicillin. *Enterococcus faecium* was resistant to ampicillin but susceptible to high level gentamicin and vancomycin.

Streptococcus pneumoniae isolates were resistant to ampicillin and oxacillin. One out of them was linezolid resistant.

Table 6: Antibiotic resistance pattern of Gram-positive bacterial isolates in the present study.

Antibiotics	<i>Streptococcus pneumoniae</i> (n=03)	<i>Staphylococcus aureus</i> (n=03)		<i>Enterococcus faecium</i> (n=01)
		MRSA (n=02)	MSSA (n=01)	
Ampicillin	02	NT	NT	01
Cefotaxime	01	NT	NT	NT
Ceftriaxone	01	NT	NT	NT
Chloramphenicol	00	NT	NT	NT
Cotrimoxazole	03	01	00	NT
Gentamicin	01	01	00	NT
High level Gentamicin	NT	NT	NT	00
Linezolid	01	00	00	00
Levofloxacin	01	NT	NT	NT
Oxacillin	02	NT	NT	NT
Penicillin	03	02	01	01
Vancomycin	00	00	00	00

Abbreviations: NT- Not Tested, MRSA- Methicillin Resistant *Staphylococcus aureus*, MSSA- Methicillin Sensitive *Staphylococcus aureus*.

Table 7 shows the antimicrobial resistance pattern of Gram-negative bacterial isolates of our study. The *Acinetobacter baumannii* and *Enterobacter cloacae* isolates were resistant to all antibiotics tested. Most of the *Escherichia coli* isolates were resistant to Beta Lactam-Beta lactam inhibitor

combination agents (BL-BLI) like Penicillins and third generation cephalosporins. One of the isolates was resistant to meropenem. Among non-beta lactam antimicrobial agents, resistance to gentamicin was seen predominantly among the bacterial isolates.

Table 7: Antibiotic resistance pattern of Gram-negative bacterial isolates in the present study.

Antibiotics	<i>Escherichia coli</i> (n=04)	<i>Acinetobacter baumannii</i> (n=01)	<i>Enterobacter cloacae</i> (n=01)
Amikacin	01	01	01
Ampicillin	03	NT	NT
Amoxicillin Clavulanate	03	NT	NT
Aztreonam	03	NT	01
Chloramphenicol	00	NT	01
Ceftazidime	03	01	01
Cefotaxime	03	01	01
Ceftriaxone	03	01	01
Cotrimoxazole	02	01	01
Gentamicin	03	01	01
Meropenem	01	01	01
Levofloxacin	03	01	01
Piperacillin Tazobactam	03	01	01

Abbreviations: NT- Not Tested.

DISCUSSION

Meningitis is serious infectious disease characterized by inflammation of the meninges and remains an important cause of

morbidity and mortality among pediatric patients worldwide. [14] Bacterial and viral pathogens constitute the major infectious etiologies of acute meningitis, with their

clinical presentation often overlapping, making early etiological diagnosis challenging. Timely recognition and laboratory confirmation of meningitis are essential for appropriate patient management, particularly in bacterial meningitis, where prompt initiation of antimicrobial therapy is critical. [15] In this context, the present study was undertaken to evaluate the bacterial and viral etiological profile of meningitis among pediatric patients presenting to a tertiary care hospital.

In our study, the age group of 1 Month to 1 Year (19.30%) was the most common age group where meningitis was suspected. The findings in the present study are consistent with other studies by Bhatta M et al (2021) [16] (40.04%), Gahlot T et al (2021) [17] (54.60%) and Attia Bari FZ et al (2016) [18] (68.3%). While studies by Shrestha RG et al (2015) [19] and Khan N et al (2014) [20] shows that the meningitis cases were highest in age groups of >60 months (36.9%) and 0-5 years (86%), respectively. The comparison shows a consistent trend across different studies that infants, are the most affected age group in meningitis cases. The increased vulnerability of this age group may be due to various factors like immaturity of their immune system, vaccination status or transmission of infection from mother. Our study correlates well with these findings, reinforcing the importance of focusing on infants in

meningitis research and prevention strategies. Overall highest positivity of bacterial and viral meningitis collectively was shown by patients in the age group of 1 Month to 1 year (36.67%). The same age group also has more viral meningitis positivity (23.33%). Biaukula VL et al (2012) [21] showed that 41.7% of the children diagnosed with viral meningitis belonged to the age group of 1 month to 1 year.

In the present study, meningitis was suspected more in female patients (55.27%) and positivity was also slightly higher (26.98%) in female gender. In contrast to this, studies by Siyal Het al (2020) [22] (57%), Gahlot T et al (2021) [17] (61.7%), Attia Bari FZ et al (2016) [18] (63.8%), Basri R et al (2015) [23] (65.6%), Shrestha RG et al (2015) [19] (55.2%), Joardar S et al (2012) [24] (60.43%) and Debnath DJ et al (2012) [25] (64.56%) showed that males are more commonly affected than females. The data on gender distribution in meningitis cases across various studies reveals a consistent trend of higher incidence among males compared to females, with most studies reporting a male predominance. This difference may suggest potential regional, genetic, or socio-cultural factors influencing the gender distribution in meningitis cases, highlighting the importance of considering these variables in epidemiological studies [26,16,27].

Table 8: Percentage of most common presenting symptoms of meningitis in different studies

Study and Year	Most common presenting Symptom	Second most common Symptom	Third most common Symptom
Present study	Vomiting (81.57%)	Fever (77.19%)	Convulsion (64.91%)
Bhatta M et al (2021) [16]	Fever (87%)	Vomiting (71%)	Convulsion (64%)
Raj KA et al (2021) [28]	Fever (96%)	Vomiting (77%)	Altered sensorium (56%) and neck stiffness (56%)
Siyal Het al (2020) [22]	Fever (100%)	Headache (67%)	Vomiting (44%)
Basri R et al (2015) [23]	Fever (73.6%)	Poor oral intake (48%)	Convulsion (36%)
Khan N et al (2014) [20]	Fever (90.7%)	Altered sensorium (78%)	Abnormal movements (60%)
Bhagawati G et al (2014) [29]	Fever (92.72%)	Altered sensorium (74.36%)	Headache (61.71%)
Khan F et al (2011) [30]	Fever (96%)	Altered sensorium (62%)	Neck stiffness (51%)
Michos AG et al (2007) [31]	Fever (98%)	Headache (94%)	Vomiting (67%)

Most of the studies show fever to be the most common presenting symptom as demonstrated in Table 8; however, in our study we found vomiting (81.57%) to be the most common presenting symptom.

In the present study, the confirmed cases of bacterial meningitis were 11.41% which is comparable to the studies conducted by Bhagawati G et al (2014) [29] (13.72%) and Abedin MZ et al (2021) [32] (14%). Some studies have reported much higher percentages of bacterial meningitis. Studies by Raj KA et al (2021) [28], Debnath DJ et al (2012) [25] and Yerramilli A et al (2017) [33] have shown 18%, 20.25% and 28% confirmed cases of bacterial meningitis respectively. While studies conducted by Iregbu KC et al (2015) [34] (5.2%), Fitzwater SP et al (2013) [35] (1.98%) and Mohammadi SF et al (2013) [36] (1.7%) have shown lower values.

Our study shows that the microbiologically confirmed viral meningitis cases were 14.91%. Study conducted in Fiji by Biaukula VL et al (2012) [21] found viral positivity of 17.14% while study by Liu H et al (2022) [37] showed 11.8% positivity. Other authors like Salavatiha Z et al (2024) [38] (55.17%), Habiba U et al (2023) [39] (59.13%) have shown much higher positivity values.

The low positivity in the present study may be due to difficulty in diagnosing clinical meningitis in very young children and different population of patients. Fluctuations in virus prevalence like sporadic cases or outbreaks, and changes in diagnostic and reporting methods over time contribute to these discrepancies. Variations in how aseptic meningitis is classified and investigated can also affect the observed frequency of viral meningitis [40,41,31].

In present study, *Escherichia coli* (30.77%) was the leading cause of bacterial meningitis, followed by *Streptococcus pneumoniae* (23.08%) and MRSA (15.38%). Each of these pathogens notably contributes to the total number of cases. The other bacteria listed (*Enterococcus faecium*, MSSA, *Enterobacter cloacae*, and

Acinetobacter baumannii) each account for a smaller proportion (07.69%) of the total cases, collectively representing the diversity of bacterial agents involved in meningitis infections. Adhikary M et al (2013) [42] also found in their study that, most common organism isolated was *Escherichia coli* (25.6%), followed by *Staphylococcus aureus* (15.4%). In study by Raza MS et al (2019) [43], most prevalent Gram negative organisms were *Escherichia coli* (27.08%), followed by *Acinetobacter baumannii* (25%). Gram positive organisms detected in his study were *Staphylococcus aureus* (8.33%), followed by *Enterococcus faecium* (6.25%). Moafi N et al (2016) [44] found that the, *H. influenzae* was the most prevalent isolate from newborns while *E.coli* was in the second place.

Study by Raj KA et al (2021) [28] showed, most common causative bacteria to be *Streptococcus pneumoniae*, *Staphylococcus aureus*, *Escherichia coli*, *Enterobacter cloacae*, *Streptococcus viridians*, *Brucella* species, and *Salmonella typhimurium* in the order of occurrence. Gahlot T et al (2021) [17] in their study showed that the most common bacteria found in culture was *Acinetobacter* species (30.6%) followed by *Enterococcus* species isolated in 18.4% of the cases and *Klebsiella pneumoniae* was isolated in 16.3% of the cases. Basri R et al (2015) [23] found that, Coagulase negative *Staphylococcus* (21.6%), was the most common pathogen in their study, followed by *Acinetobacter* species (17.6%), *Staphylococcus aureus* (13.6%), *Streptococcus* species (11.2%) and *Klebsiella pneumoniae* (6.4%). In the study conducted by Ali SA et al (2021) [45] he found *Streptococcus pneumoniae* (14.7%) to be the most common bacteria followed by, *Staphylococcus aureus* (6.9%), *Neisseria meningitidis* (5.7%), *Haemophilus influenzae* (2.5%), *Escherichia coli* (4.5%), and *Klebsiella pneumoniae* (1.9%).

In the present study among patients of viral meningitis, most common virus diagnosed was CMV and HSV 1 each found in 23.53% of cases. Other important viral etiological

agents found were EBV, HHV6, HHV7, Human Parvovirus B19 (each 11.76% of cases) and lastly Adenovirus (05.88%). Multiple literature suggests Enterovirus as the most common etiological agent, while our study showed that Herpesviridae family was more common. This can be attributed to asymptomatic shedding of virus from mother to child, geographic and demographic differences and variations in diagnostic techniques. Additionally, factors like study population characteristics, sample sizes, and regional outbreaks contribute to differing prevalence rates of viral infections [15,16,42].

Limitations

A cross-sectional study design provides a snapshot at one point in time and does not account for temporal changes. The frequency of meningitis can vary over time due to changes in population immunity, seasonal factors, pathogen prevalence, and healthcare practices. The findings are based on data from a single tertiary care hospital, which may not be representative of other regions. This limits the generalizability of the results to broader populations. Differences in diagnostic methods and criteria over time or between microbiologists can introduce variability in identifying isolates and classifying cases of bacterial and viral meningitis. The diagnostic tests used to identify bacterial and viral pathogens may have varying sensitivity and specificity, leading to potential misclassification or missed diagnoses. Bacterial resistance patterns can change rapidly, which may make the results less relevant over time. Furthermore, only a limited range of antibiotics was tested, which may not provide a comprehensive view of resistance profiles. These limitations highlight the need for further research with larger, more diverse samples, advanced methodologies, and longitudinal analyses to better understand objectives of study.

CONCLUSION

Meningitis remains a critical can't-miss diagnosis for children where early recognition and appropriate treatment can improve survival and neurological outcomes of it. Signs and symptoms of meningitis may be non-specific and require high index of suspicion to recognize potential meningitis and perform the cerebrospinal fluid studies necessary of diagnosis. In the present study, viral meningitis was predominant and seen in 14.91% of patients while bacterial meningitis was seen in 11.41% patients. Overall, the most commonly affected age group was 1 month to 1 year (36.67%), followed by 10 year to 14 year age group (33.33%). Also, it was found that 1 month to 1 year age group (23.33%) was predominantly affected by viral meningitis. With the introduction of pneumococcal vaccine in the National Immunization Schedule, a lesser number of pneumococcal meningitis was expected in the pediatric age group. But our study underscores that *Streptococcus pneumoniae* is still a major cause of bacterial meningitis in children. Given the relatively high frequency of viral meningitis in our study, particularly CMV and HSV 1, there is a need for enhanced surveillance and early diagnostic efforts in pediatric patients especially in 1 month to 1 year age group presenting with symptoms of meningitis.

Declaration by Authors

Ethical Approval: Approval (Ref O.No/Pharma Dept/IEC/Approval Letter/190/22) from the Institutional Ethics Committee (IEC- Dr. Vaishampayan Memorial Government Medical College, Solapur) was obtained. The hospital's department of paediatrics had given permission to use patient records, assess patients, and interview patient informants. While the Department of Pathology and Biochemistry assisted in the collection of CSF cytology and biochemistry data. The extracted data was treated discreetly, with no identifying information recorded on the data sheet.

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