535 suspected meningitis cases from various pediatric hospitals were considered for study. Among 535 cases 236 were pyogenic meningitis cases. Out of 236 cases, 40.3% parents were from urban and 59.7% were from rural areas of Gulbarga district. The most important signs of pyogenic meningitis were fever, headache, lethargy, irritability and vomiting. Respiratory signs were seen among 19.5% of cases. Seizures in 61.4%, petechial rashes in 7.6%, conjunctival discharge in 7.6% cases and bulging fontanella was observed in 3.4%.

Out of 236 pyogenic meningitis cases, 157 were male and 79 female. Male preponderance over female was observed. The most affected group was those between 4 months to 3 years. The mean age at presentation was 4.1±0.71 months.

Socio-economic status of the mother revealed that 58.3% of the parents of the children were from lower socio-economic group, followed by 33.7% middle, 5.8% upper middle and 2.2% upper income group.

In untreated cases, gram stained smear examination of the CSF samples showed positivity results in 208 of 236 samples. Gram-positive diplococci were the common organism to be observed (33.9%) (Table-1). The other organisms observed in the smear are Gram-negative bacilli in 70 cases (29.7%), Gram-negative diplococci in 22 (9.3%) cases, Gram positive cocci in clusters 10 (4.2%).
Incidence of culture positivity was 84.3%. Males were more prone for meningitis. *S. pneumoniae* was the common isolate 89 (44.7%) grown followed by *Haemophilus influenzae type b* 51 (25.6%), *Neisseria meningitides* was isolated in 12 (6.0%), Group B Streptococci in 19 (9.5%), *Staphylococcus aureus* in 11(5.5%), *Klebsiella pneumoniae* in 08 (4.0%), *E. coli* in 06 (2.5%) and *Pseudomonas aeruginosa* 03 (1.5%).

Latex agglutination test for detection of bacterial antigens were done for all 236 untreated cases, of which 214 (90.7%) of CSF samples were positive of one or other bacterial antigens. The most common bacterial antigens identified by LAT are 94 *Pneumococcal* meningitis, 61 *Haemophilus influenzae* meningitis, 29 Group B *Streptococci* meningitis 18, *Neisseria meningitidis*, and 12 *E. coli* meningitis. A total of 55 patients received medication prior to hospitalization. In treated group, the conventional bacteriological technique identified the causative organism in 5 (7.2%) cases whereas 16 (20%) cases were identified by LAT.

Among 236 untreated cases, 146 CSF samples were positive for CRP. Of which 102 samples were positive for both culture and CRP, 28 were positive only for CRP and Culture negative. 97 CSF sample which were culture positive were CSF-CRP negative.
Gram stain has 100% sensitivity and negative predictive accuracy in comparison with culture. Among 236 untreated cases, 208 CSF samples were positive for gram stain. Of which 199 samples were positive for both culture and gram stain, 09 were positive only for gram stain, but culture negative. 28 CSF sample which were both culture negative and gram stain negative. None of the samples which were positive for culture was negative for gram stain.

Latex agglutination has 100% sensitivity and negative predictive accuracy in comparison with culture. Positive predictive accuracy was 96%. Latex agglutination test for detection of bacterial antigens were done for all 236 untreated cases, of which 214 (90.7%) of CSF samples were positive of one or other bacterial antigens. 199 CSF samples were positive for both culture and LAT, 09 CSF samples were positive for LAT, but negative for culture.

CRP has 51.3% sensitivity, 24.3% specificity and 84.9% negative predictive accuracy in comparison with culture. Positive predictive accuracy was 78.5%. Among 236 untreated cases, 146 CSF samples were positive for CRP. Of which 102 samples were positive for both culture and CRP, 28 were positive only for CRP and Culture negative. 97 CSF sample which were culture positive were CSF-CRP negative.
There is an emergence of drug resistance in pyogenic meningitis. Majority of organisms were resistance to two or more antibiotics. In Gram-negative bacilli, 62.5% of *Klebsiella* sps, 66.7% of *H. influenzae*, 60.0% of *E. coli*, 66.7% of *Pseudomonas aeruginosa* were resistant to more then three antibiotics. Among Gram-positive cocci, 54.5% of *Staphylococci aureus* was resistant to more then three antibiotics. Methicillin resistant *Staphylococcus aureus* was detected in 34.6%. Penicillin resistant *Streptococcus pneumoniae* was seen 23.6% of the isolates, but these isolates were sensitive to ampicillin and cephoxitin. Extended spectrum β-lactamase medicated resistance was seen in 52.2% of isolates. *Klebsiella pneumoniae* was the predominant ESβL producer followed by *E. coli* and *Pseudomonas aeruginosa* and *H. influenzae*.

Information on the prevalence of Amp C β-lactamase producing strains in India is very limited and no data’s are available on the prevalence of Amp C production in pediatric group. In the present study 17.9% were AmpC producers and *H. influenzae* was the predominant Amp C producer followed by *Klebsiella pneumonia.*

In the present study 29.5% of gram negative bacilli were metallo β-lactamase producers. *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*, were the predominant MBL producers.
• The application of the multiplex PCR as an epidemiological tool for improved non-culture diagnosis was evaluated. This study outlines the evaluation of a single-tube multiplex real-time PCR for the simultaneous detection of *N. meningitidis*, *H. influenzae* and *S. pneumoniae* in clinical samples using the TaqMan system. The sensitivity and specificity for the detection of the three major meningitis-causing pathogens are assessed.

• The CSF which was negative in culture was positive in single tube multiplex real time PCR. By utilizing the available TaqMan technology, the introduction of a three-in-one multiplex PCR enables rapid identification and a high throughput of samples (130 min for 96 specimens), with a modest additional cost for primers and probes in each reaction. The multiplex PCR demonstrated that testing a large number of previously culture-negative specimens provides information on the incidence of meningococcal, *H. influenzae* and pneumococcal infections in clinical specimens originally referred for meningococcal PCR testing.

• In summary, the spectrum of bacteria causing pyogenic meningitis has remained more or less the same at our region compared to other data. It needs to be reiterated that simple, rapid, inexpensive tests like the Gram stain remain significant means of diagnosis of pyogenic meningitis in developing countries. To increase the cost-
effectiveness in a resource limited setting, LAT for pneumococcal antigen should be performed first, since it is the most common pathogen causing pyogenic meningitis in all age groups. Smear negative CSF samples with neutrophilic pleocytosis in patients with a clinical suspicion of bacterial meningitis warrant antigen testing with LAT.

- *Streptococcus pneumoniae* remains the major aetiological agent of pyogenic meningitis both in adults and children not only in India, but worldwide. High fatality rates have been reported by *Streptococcus pneumoniae* with long-term neurological sequelae in survivors. Meningitis caused by *H. influenzae* has almost been eliminated from the Western world following routine vaccination with Hib conjugate vaccine but not in India. *H. influenzae* is the second most predominant bacteria in our study. Enforcing mandatory Hib vaccine may reduce meningitis with *H. influenzae* and also introduction of conjugate vaccines against *Streptococcus pneumoniae* can reduce the burden of childhood meningitis and may produce herd immunity among adults.

- Theoretically, pneumococcal meningitis too is a vaccine preventable disease, but unfortunately, the currently available polysaccharide vaccine is not effective in children under two years of age. Universal use of meningococcal vaccine is unattractive as the disease is
generally sporadic and vaccine is of limited immunogenicity. However if *H. influenzae* and pneumococcal vaccines become a reality, the incidence of pyogenic meningitis in early childhood can reduce by more than 60%, with considerable reduction in the financial and emotional cost burden of the disease.

- The inclusion of the multiplex PCR in the routine molecular diagnostic screening regimen would provide an adjunct in improved non-culture diagnosis and case ascertainment of meningitis and septicemia.