Research Article
Open Access
CrossMark

Gram negative acute bacterial meningitis an emerging entity in a tertiary care centre, New Delhi

Keywords: acute bacterial meningitis, *listeria monocytogenes*, *streptococcus species*, cerebrospinal fluid, p yogenic meningitis

Abbreviations: BHI, brain heart infusion; CSF, cerebrospinal fluid; ABM, acute bacterial meningitis; LAT, latex agglutination tests

Volume 3 Issue 4 - 2016

Dimple Kasana
Department of Microbiology,VMMC & Safdarjung Hospital, India

Correspondence: Dimple Kasana, Senior Specialist & Associate professor,VMMC & Safdarjung Hospital, New Delhi, India, Email dimplekasana@gmail.com

Received: September 09, 2016 | **Published:** December 20, 2016

Introduction

The irritation or inflammation of the meninges, the covering of brain and spinal cord, is known as meningitis. Acute bacterial meningitis (ABM) is a medical emergency, which needs an early diagnosis and an aggressive management. ABM remains a major cause of mortality and long-term neurological sequelae worldwide. Despite the availability of potent newer antibiotics, the mortality rate due to ABM remains significantly high in India and other developing countries, ranging from 16-32%.[1–4] Most often, therapy for ABM has to be initiated before the aetiology is known. The emergence of antimicrobial resistance has added to this problem, and current recommendations are to identify targets for immunization, formulate preventive strategies and to carry out a rational empirical treatment.[5–9] Among neonates, Group B and non-Group B *Streptococcus species* (49%), *Escherichia coli*(18%) and *Listeria monocytogenes*(7%) are the most common pathogens which are seen. The meningitis-causing pathogens which are found in infant and child age groups are *Haemophilus influenzae* (40-60%), *Neisseria meningitidis* (25-40%) and *Streptococcus pneumoniae* (10-20%). The common pathogens which are encountered in adult meningitis are *Streptococcus pneumoniae* (30-50%), *Neisseria meningitidis* (10-35%), *Staphylococci*(5-15%), other *Streptococcus species*, *Haemophilus influenzae* (1-3%), Gram negative *bacilli*(1-10%) and *Listeria monocytogenes*.[10–14] A delay in diagnosis and initiation of antimicrobial therapy can result in a poor outcome of the disease. The aim of the present study was to analyze the emerging pathogen, bacterial profiles and antimicrobial susceptibility patterns of the isolates which were obtained from CSF of patients with ABM in the given area.

Material and methods

Retrospective study was conducted in the Department of Microbiology, at a tertiary care hospital in New Delhi, during the period of 3years from june2012-june2015. Six thousands suspected cases of ABM, who were admitted to various wards of our hospital, were included in this study. All cerebrospinal (CSF) fluid was send to the department of microbiology. Criteria used for inclusion of cases in the study were the presence of a positive CSF culture for bacterial pathogens and/or a positive latex agglutination test for antigen detection.

Macroscopic examination

All CSF samples were macroscopically examined for turbidity, haemorrhagic CSF and cobweb appearance for suspected case of bacterial etiology, traumatic injury to spinal cord and tubercular meningitis respectively.

Cell counts, smear and culture

All CSF samples received at the microbiology department were processed immediately. Direct microscopy by doing Gram's stain was done for pus cells, red blood cells and micro-organisms. Culture was put on chocolate agar, blood agar and Mac Conkey agar in the candle jar for 48hours. The rest samples were incubated in the brain heart infusion (BHI) broth for 48hours. A battery of biochemical tests were done for identification of the organism.[15]

Antigen detection

Detection of soluble antigens of *H. infl uenzae* (type b), *S. pneumoniae* and *N. meningitides* in CSF was performed by latex agglutination tests (LAT), using commercial kits Tulip Diagnostics Pvt. Ltd, India. Antimicrobial sensitivity test was performed on Mueller Hinton agar by the Kirby Bauer disk diffusion method.[16] The antibiotics which were used like ampicillin (10µg), cefoxitin (30µg), ceftazidime (30µg), amikacin (10µg), ciprofloxacin (30µg), erythromycin (30µg), penicillin G (10units), piperacillin+tazobactam (100/10µg), imipenem (10µg), meropemen (10µg), chloramphenicol (30µg) and vancomycin (30µg), which were obtained from Hi Media Laboratories, Mumbai, India. The results were interpreted as per NCCLS-2000 recommendations.[16]

Results

Of the total enrolled population of patients (6000), 1320 were fulfilled the inclusion criteria of this retrospective study as 1320 (22%) were culture and/or antigen confirmed cases of bacterial meningitis (Figure 1). Nine hundred and ninety were male (75%) and 330 (25%) were female (Figure 2). Two sixty four (20%) patients belonged to the paediatric age group (less than 12years) while 1056 (80%) were adults (Figure 3). Pus cells could be demonstrated by the Gram stain in the CSF samples of 462/1320 (35%) patients, while 858/1320 (65%) were Gram stain negative for pus cells. All smear positive for pus cells were also culture positive culture for bacteria (Table 1). The bacterial pathogen could be demonstrated by the Gram stain in the CSF samples of 198/1320 (15%) patients, while 1122/1320(85%) were Gram stain negative for bacterial pathogen. All smear positive for bacteria were also culture positive culture (Table 2). *Staphylococcus aureus* was the

most common pathogen in adult population (1056) which was isolated in 316 (30%) culture positive cases, followed by *Acinetobacter baumanii* which was isolated in 264(25%) of patients further followed by *Pneumococcus pneumoniae* (15%), *Klebseilla pneumoniae* (10%), *E. Coli* (10%) and α *Streptococci* (10%). There was no isolation of *Haemophilus influenzae and Neissaria* meningitides (Figure 4). For paediatric population *Staphylococcus aureus* and *Acinetobacter baumannii* is the most common isolated pathogen 66(25%) followed by *E.coli* (20%), *Pneumococcus pneumoniae* (10%), *Klebsiella pneumoniae* (10%) and α *Streptococci* (10%). There was no isolation of *Haemophilus influenzae and Neissaria meningitides* (Figure 5). The methicillin resistant *staphylococcus aureus* (MRSA) is 287(75%) of total isolated *staphylococcus aureus* (382) species while 95(25%) were methicillin sensitive *staphylococcus aureus* (MSSA). All Gram negative organisms was showing 91% sensitivity to cefoperazone+Salbactum combination followed by imipenem (85%), piperacillin+salbactum(78%), meropenem (78%), netilimicin (50%), amikacin (50%) and ceftazidime (50%) as shown in Figure 6. For Gram positive organisms all were sensitive to vancomycin and chloramphenicol followed by ciprofloxacin (85%), gentamicin (75%), erythromicin (40%), cefoxitin (25%), optochin (10%) and penicillin only (3%) as shown in Figure 7.

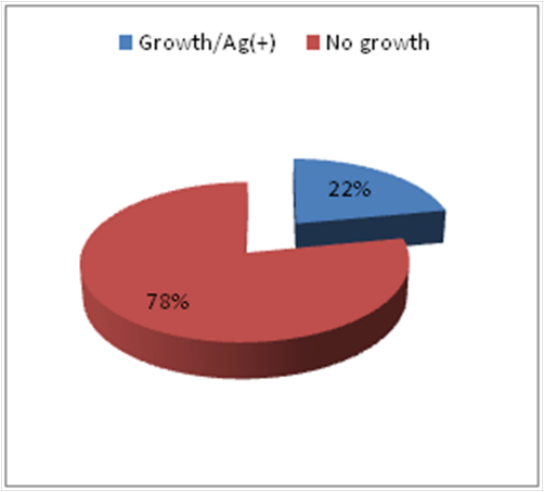

Figure 1 Percentage wise distribution of culture positive cases (1320/6000).

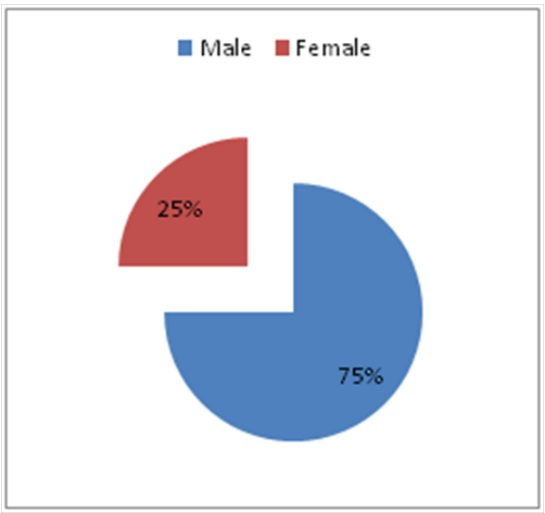

Figure 2 Percentage distribution of patients of ABM based on sex (Total patients= 1320).

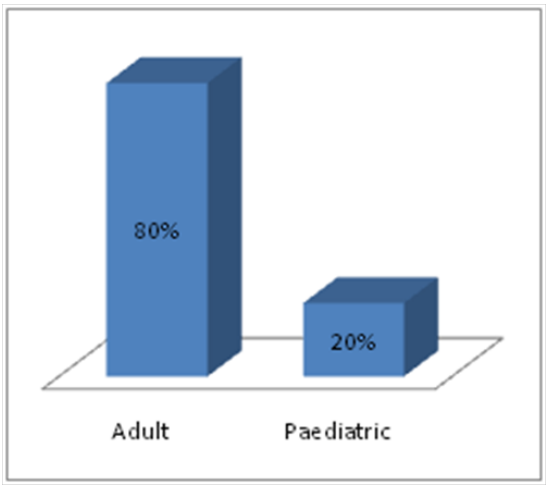

Figure 3 Percentage distribution of patients of ABM based on age (Total patients= 1320).

Table 1 Showing correlation of pus cells in Gram's stain and culture in bacterial meningitis

Culture (+)	Smear (-) for pus cell	Smear (+) for pus cells
1320	858(65%)	462(35%)

Table 2 Showing correlation of bacteria in Gram's stain and culture in bacterial meningitis

Culture (+)	Smear (+) for bacteria	Smear (-) for bacteria
1320	1122 (85%)	198(15%)

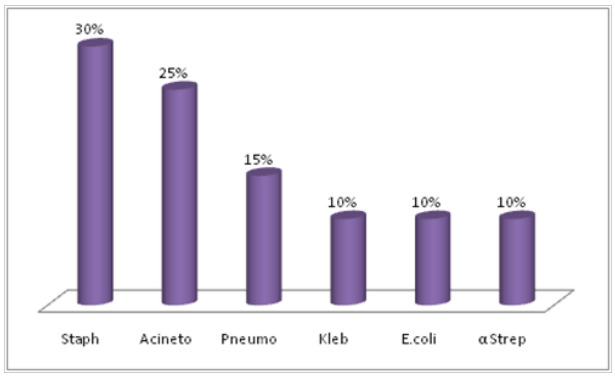

Figure 4 Distribution of organisms in adult bacterial meningitis cases.

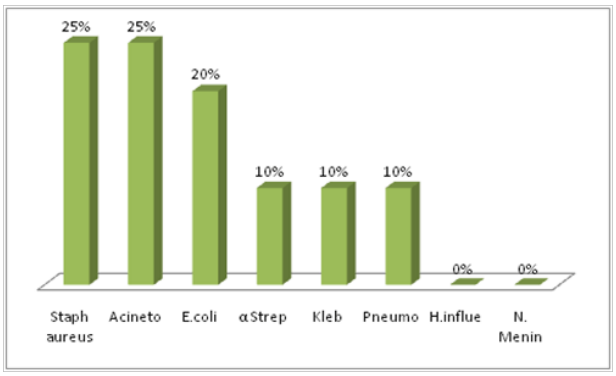

Figure 5 Distribution of organisms in paediatric bacterial meningitis cases.

Citation: Kasana D. Gram negative acute bacterial meningitis an emerging entity in a tertiary care centre, New Delhi. *J Bacteriol Mycol Open Access.* 2016;3(4):267–271. DOI: 10.15406/jbmoa.2016.03.00068

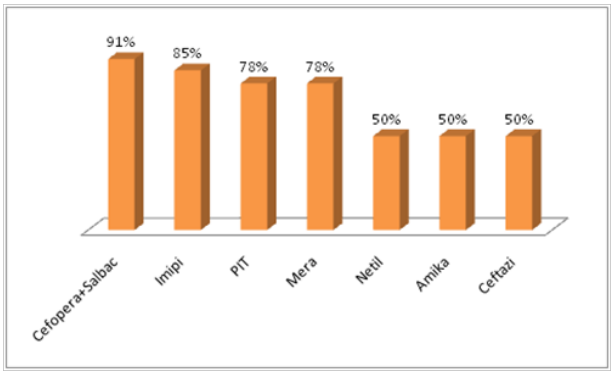

Figure 6 Showing sensitivity pattern of Gram negative organisms.

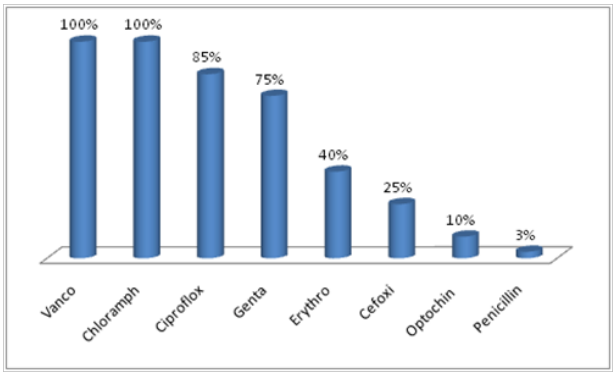

Figure 7 Showing sensitivity pattern of Gram positive organisms.

Discussion

Meningitis can be broadly classified as pyogenic, granulomatous, and lymphocytic. Acute bacterial or pyogenic meningitis is a potentially life threatening disease that consists of inflammation of the meninges and the underlying subarachnoid CSF. ABM is a medical emergency, which warrants early diagnosis and aggressive therapy. The choice of antimicrobial therapy is based on the most common pathogen prevalent in a particular geographical area and age group and their antibiotic susceptibility pattern. Though the common pathogens associated with bacterial meningitis in the west are *H. influenzae, N. meningitidis, S. Pneumoniae* 9 and *Listeria monocytogenes*.[17–19] However, in developed countries with effective vaccination programmes, the incidence of *H. influenzae* and *N. meningitidis* showed decreased trends.[20,21] In our study the culture confirmed and/or antigen positive cases were 1320 i.e. only 22% of total study population (6000) showed growth and rest were sterile for any bacterial pathogen. The male: female ratio is 4:1. This male preponderance which is seen with this disease, has also been reported in several previous studies.[22–24] A simple Gram stained smear can offer immediate clues to aid a diagnosis of pyogenic meningitis. Some studies have reported a CSF Gram stain sensitivity of 60-90% and a high speciÞ city of >97%, stressing its importance in the rapid and accurate diagnosis of the causative bacteria.[25,26] The yield of bacteria on a Gram stain depends on several factors like the number of organisms present, prior use of antibiotics, technique used for smear preparation (centrifuged deposit, cytospin, direct smear etc.), staining techniques and the observers skill and experience. Despite low Gram stain smear positivity from CSF samples and the fact that a negative Gram stain does not rule out infection, the importance of a positive smear cannot be over-emphasized, especially in developing countries where financial constraints limit the use of other rapid diagnostic tests to diagnose this potentially fatal infection. In our study Gram's stain provides 35% positivity for pus cells and 15% positivity for bacteria. Most Indian studies report only culture findings or low smear positivity.[27]

In our study the culture positivity rate was 22% (1320/6000). Various studies report culture negative cases of meningitis or a low CSF culture positivity, ranging from 6 to 50%.[25,27,28] Various reasons cited in the literature for a low yield of bacteria on culture are prior antibiotic therapy, delay in transport of specimens to the laboratory, non availability of special media for specific pathogens, presence of autolysis enzymes in CSF and lack of a 24hour facility for processing CSF samples. *Staphylococcus aureus* was the most common pathogen in adult population (1056) which was isolated in 316 (30%) culture positive cases, followed by *Acinetobacter baumanii* which was isolated in 264(25%) of patients further followed by *Pneumococcus pneumoniae* (15%), *Klebseilla pneumoniae* (10%), *E. Coli* (10%) and α *Streptococci* (10%). There was no isolation of *Haemophilus influenzae and Neissaria* meningitides. For paediatric population *Staphylococcus aureus* and *Acinetobacter baumannii* is the most common isolated pathogen 66(25%) followed by *E.coli* (20%), *Pneumococcus pneumoniae* (10%), *Klebsiella pneumoniae* (10%) and α *Streptococci* (10%). There was no isolation of Haemophilus influenzae and *Neissaria* meningitides. The MRSA is 287(75%) of total isolated *staphylococcus aureus* (382) species while 95(25%) were MSSA. In our study we observed that *S. aureus* and *Acinetobacter bowmanii* has been emerged as the most common pathogen causing ABM in all age groups followed by other gram positive and gram negative *bacilli*. A study in Taiwan showed an increase in the incidence of staphylococcal infection which rose from 15% to 23% and a decrease in the incidence of *Streptococcus pneumoniae* which fell from 10.6% to 3.6%. Staphylococcal strains replaced *S. pneumoniae*, becoming the most common Gram positive pathogen of acute ABM in their hospital.[29,30] In our study *Acinetobacter bowmanii* emerged as an emerging pathogen in both age group (Adult:Paediatric/30%:25%). Multidrug-resistant *Acinetobacter baumannii* is a rapidly emerging pathogen in the health care setting, where it causes infections that include bacteraemia, pneumonia, meningitis, urinary tract infection, and wound infection. The organism's ability to survive under a wide range of environmental conditions and to persist for extended periods of time on surfaces make it a frequent cause of outbreaks of infection and an endemic, health care-associated pathogen.[31,32] In many studies *Acinetobacter baumannii* has been reported as a cause of meningitis as hospital acquired infection or acquired after neurosurgical procedures.[33–38]

All Gram negative organisms showing 91% sensitivity to cefoperazone+salbactum combination followed by imipenem (85%), piperacillin+salbactum(78%), meropenem (78%), netilimicin (50%), amikacin (50%) and ceftazidime (50%). For Gram positive organisms all were sensitive to vancomycin and chloramphenicol followed by ciprofloxacin (85%), gentamicin (75%), erythromicin (40%), cefoxitin (25%), optochin (10%) and penicillin only (3%). These results signify the varying levels of drug resistance amongst the gram positive and the gram negative microbes, and the need to control the spread of these resistant strains before they reach the alarming levels in this region. Among the gram negative *bacilli* a general decline in the sensitivities to all groups of drugs was noticed. The simultaneous decline in sensitivities to different group of drugs can be correlated to the rampant indiscriminate use of antibiotics leading to a large scale drug resistance. As observed with the gram negative *bacilli*, a similar pattern of increasing drug resistance was seen among the

Citation: Kasana D. Gram negative acute bacterial meningitis an emerging entity in a tertiary care centre, New Delhi. *J Bacteriol Mycol Open Access.* 2016;3(4):267–271. DOI: 10.15406/jbmoa.2016.03.00068

Staphylococcal species, Streptococcus species. However, fortunately no vancomycin and chloramphenicol resistance were detected. As in Gram negative *bacilli*, cefoperazone+salbactum combination and carbapenems had very good sensitivity but they have side effects also. However seeing the predominance of gram positive cocci particularly *Staphylococcus aureus* with alarming rates of methicillin resistance (75%), suitable antibiotics should be given after sensitivity report. Stress should be given on the restrained and rationale use of antimicrobials both in and outside the hospital.

Conclusion

The spectrum of organisms which cause acute bacterial meningitis varies with time, geography and patients' ages. Since clinical signs of meningitis are not always reliable, a laboratory support is imperative, to achieve an early diagnosis and proper treatment. The emergence of antimicrobial resistance has added to this problem, and current recommendations are to identify targets for immunization, formulate preventive strategies and to carry out rational empirical treatment, especially for potentially fatal bacterial meningitis. Antimicrobial resistance is increasing, likely as a result both of the emergence of resistance in the context of antimicrobial pressure and of health care-associated transmission of drug-resistant strains. *Acinetobacter* infection poses a formidable threat to patients and is an emerging pathogen for meningitis in this study.

Acknowledgements

None.

Conflict of interest

The author declares no conflict of interest.

References

1. Kabra SK, Praveen Kumar, Verma IC, et al. Bacterial meningitis in India: An IJP survey. *Indian J Pediatr*. 1991;58(4):505–511.
2. Tang LM, Chen ST, Hsu WC, et al. Acute bacterial meningitis in adults: a hospital–based epidemiological study. *QJM*. 1999;92(12):719–725.
3. Ayaz C, Mehmet FG, Hosoglu S, et al. Characteristics of acute bacterial meningitis in Southeast Turkey. *Indian J Med Sci*. 2004;58(8):327–333.
4. Chinchankar N, Mane M, Bhave S, et al. Diagnosis and outcome of acute bacterial meningitis in early childhood. *Indian Pediatr*. 2002;39(10):914–921.
5. Mani R, Pradhan S, Nagarathna S, et al. Bacteriological profile of community–acquired acute bacterial meningitis: a ten year retrospective study in a tertiary neurocare centre in South India. *Indian J Med Microbiol*. 2007;25(2):108–114.
6. Schuchat A, Robinson K, Wenger JD, et al. Bacterial meningitis in the United States in 1995. Active Surveillance Team. *N Eng J Med*. 1997;337(14):970–976.
7. Wenger JD, Broome CV. Bacterial meningitis: epidemiology. In: Lambert HP, editor. *Infections of the central nervous system*. USA: BC Decker; 1996. p. 16–31.
8. Das BK, Gurubacharya RL, Mohapatra TM, et al. Bacterial antigen detection test in Meningitis. *Indian J Pediatr*. 2003;70(10):799–801.
9. Madhumita P, Gupta N. Clinical and bacteriological spectrum of community acquired acute bacterial meningitis in adults at a tertiary care hospital in northern India. *IJNPND*. 2011;1(2):194–200.
10. Chandramukhi A. Neuromicrobiology. *In: Neurosciences in India: Retrospect and Prospect*. India: The Neurological Society of India, CSIR; 1989. p. 361–395.
11. Chinchankar N, Mane M, Bhave S, et al. Diagnosis and outcome of acute bacterial meningitis in childhood. *Indian Padiatr*. 2002;39(10):914–921.
12. Sonavane A, Baradkar VP, Mathur M. Bacteriological profile of pyogenic meningitis in adults. *Bombay Hosp J*. 2008;50(3):452–455.
13. Khan F, Rizvi M, Fatima N, et al. Bacterial meningitis in North India: Trends over a period of eight years. *Neurology Asia*. 2011;16(1):47–56.
14. Van de Beek D, de Gans J, Tunkel AR, et al. Community–acquired bacterial meningitis in adults. *N Engl J Med*. 2006;354(1):44–53.
15. Forbes BA, Sahm DF, Weissfeld AS. *Bailey and Scott's Diagnostic Microbiology*. 11th ed. USA: Mosby; 2002.
16. CLSI. *Performance standards for antimicrobial susceptibility testing; twentieth informational supplement*. CLSI document M100–S20. USA: Clinical and Laboratory Standards Institute; 2010:30(1).
17. Schlech WF, Ward JI, Band JD, et al. Bacterial meningitis in the United States, 1978 Through 1981. The National Bacterial Meningitis Surveillance Study. *JAMA*. 1985;253(12):1749–1754.
18. Schuchat A, Robinson K, Wenger JD, et al. Bacterial meningitis in the United States in 1995. Active Surveillance Team. *N Eng J Med*. 1997;337:970–976.
19. Tang LM, Chen ST, Hsu WC, et al. Acute bacterial meningitis in adults: a hospital based epidemiological study. *QJM*. 1999;92(12):719–725.
20. Chong HT, Tan CT. Epidemiology of central nervous system infections in Asia, recent trends. *Neurology Asia*. 2005;10:7–11.
21. Chan YC, Wilder Smith A, Ong BKC, et al. Adult Community acquired bacterial meningitis in a Singaporean teaching hospital. A seven year overview (1993–2000). *Singapore Med J*. 2002;43(12):632–636.
22. Khan F, Rizvi M, Fatima N, et al. Bacterial meningitis in North India: Trends over a period of eight years. *Neurology Asia*. 2011;16(1):47–56.
23. Tang LM, Chen ST, Hsu WC, et al. Acute Bacterial Meningitis meningitis in adults: a hospital–based epidemiological study. *QJM*. 1999;92(12):719–725.
24. Pfister HW, Feiden W, Einhaupl KM. Spectrum of complications during bacterial meningitis in adults: results of a prospective clinical study. *Arch Neurol*. 1993;50(6):575–581.
25. Chinchankar N, Mane M, Bhave S, et al. Diagnosis and outcome of acute bacterial meningitis in early childhood. *Indian Pediatr*. 200239(10):914–921.
26. Van de Beek D, de Gans J, Spanjaard L, et al. Clinical features and prognostic factors in adults with bacterial meningitis. *N Engl J Med*. 2004;351:1849–1859.
27. Das BK, Gurubacharya RL, Mohapatra TM, et al. Bacterial antigen detection test in Meningitis. *Indian J Pediatr*. 2003;70(10):799–801.
28. Kabra SK, Praveen Kumar, Verma IC, et al. Bacterial meningitis in India: An IJP survey. *Indian J Pediatr*. 1991;58(4):505–511.
29. Tang LM, Chen ST, Hsu WC, et al. Acute bacterial meningitis in adults: A hospital based epidemiological study. *QJM*. 1999;92(12):719–25.
30. Liu CC, Chen JS, Lin CH, et al. Bacterial meningitis in infants and children in southern Taiwan: emphasis on *Haemophilus infl uenzae* tybe B infection. *J Formos Med Assoc*. 1993;92(10):884–888.
31. Fournier PE, Richet H. The epidemiology and control of *Acinetobacter baumannii* in health care facilities. *Clin Infect Dis*. 2006;42(5):692–699.
32. Jawad A, Heritage J, Snelling AM, et al. Influence of relative humidity and suspending menstrua on survival of *Acinetobacter* spp. on dry surfaces. *J Clin Microbiol*. 1996;34(12):2881–2887.
33. Huttova M, Freybergh PF, Rudinsky B, et al. Postsurgical meningitis caused by *Acinetobacter baumannii* associated with high mortality. *Neuro Endocrinol Lett*. 2007;28(Suppl 2):15–16.

Citation: Kasana D. Gram negative acute bacterial meningitis an emerging entity in a tertiary care centre, New Delhi. *J Bacteriol Mycol Open Access*. 2016;3(4):267–271. DOI: 10.15406/jbmoa.2016.03.00068

34. Metan G, Alp E, Aygen B, et al. *Acinetobacter baumannii* meningitis in post–neurosurgical patients: clinical outcome and impact of carbapenem resistance. *J Antimicrob Chemother*. 2007;60(1):197–199.

35. Rodriguez GA, Blanco A, Asensi V, et al. Multidrug–resistant *Acinetobacter* meningitis in neurosurgical patients with intraventricular catheters: assessment of different treatments. *J Antimicrob Chemother*. 2008;61(4):908–913.

36. Chen SF, Chang WN, Lu CH, et al. Adult *Acinetobacter* meningitis and its comparison with non–*Acinetobacter* gram negative bacterial meningitis. *Acta Neurol Taiwan*. 2005;14(3):131–137.

37. Denton M, Wilcox MH, Parnell P, et al. Role of environmental cleaning in controlling an outbreak of *Acinetobacter baumannii*on a neurosurgical intensive care unit. *Intensive Crit Care Nurs*. 2005;21(2):94–98.

38. Yang M, Hu Z, Hu F. Nosocomial meningitis caused by *Acinetobacter baumannii*: risk factors and their impact on patient outcomes and treatments. *Future Microbiol*. 2012;7(6):787–793.

Citation: Kasana D. Gram negative acute bacterial meningitis an emerging entity in a tertiary care centre, New Delhi. *J Bacteriol Mycol Open Access*. 2016;3(4):267–271. DOI: 10.15406/jbmoa.2016.03.00068