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BACTERIAL PROFILE AND ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF BACTERIAL MENINGITIS AMONG NEONATES AND CHILDREN AT GONDAR UNIVERSITY HOSPITAL, NORTHWEST ETHIOPIA

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Abstract

Bacterial meningitis account for the highest proportion among cause of morbidity and mortality in neonatology and pediatrics patients and posing considerable challenge in developing countries like Ethiopia. However, information on prevalence of bacterial meningitis and antimicrobial susceptibility pattern among neonates and children in Gondar University hospital is very scarce. This study was aimed to assess the prevalence and antimicrobial susceptibility pattern of bacterial meningitis among childhood attending at University of Gondar Teaching Hospital from 2002-2012. This laboratory based retrospective analysis of 2170 cerebrospinal fluids culture and antimicrobial susceptibility tests were conducted in the Bacteriology laboratory of the department of microbiology at University of Gondar Teaching Hospital within a ten year period between September 2002-2012. Bacterial pathogens were isolated from 97 patients with an isolation rate of 4.5%. The predominant bacteria were *Streptococcus pneumoniae* 35(36.1%), followed by *Nessieria meningitides* 28(28.9%) and *Haemophilus* species 9 (9.3 %). The gram positive bacteria were constituted 52 (54.7%). Among gram positive organism S. *pneumoniae* showed a high level of drug resistance against co-trimoxazole 24 (68.6%), gentamycin 23(65.7%), tetracycline 14(40 %), penicillin 12(34.3%) and chloramphenicol 9 (25.8%). Of the gram negative bacteria, N. *meningitides* was found to be resistant to co-trimoxazole 24(85.8%), tetracycline 19(67.8 %), ampicillin 9(32.2%), gentamycin 7 (25%) and penicillin 6 (21. 5%). Multidrug resistance was observed in 61.9% of the isolates. The predominant causes of meningitis in this study are S. pneumoniae, N. meningitides and Haemophilus species. The frequency of single as well as multidrug resistance was very high among the bacterial isolates. Based on the findings a longitudinal study is recommended to determine bacterial meneginitis and antimicrobial susceptibility at large scale to target interventions.

Keywords: Bacterial meningitis, Childhood, Antimicrobial susceptibility.

Introduction

Bacterial meningitis is an inflammation affecting the pia, arachnoids and subarachnoid space that happens in response to bacteria and bacterial products[1]. Bacterial meningitis can be quite severe and may result in brain damage, hearing loss or learning disability and death if not treated early[2].

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Now a day's despite increase availability of potent anti microbial and sophisticated intensive care units, bacterial meningitis continues to be a significant cause of morbidity and mortality. This is reflected by the fact that the global burden of the disease in childhood is at least 1.2 million cases of bacterial meningitis every year with 1, 35,000 deaths [3,4.] The highest burden of meningococcal meningitis occurs in sub- Saharan Africa, in an area that is referred to as the meningitis belt which has an estimated total population 300 million[5]. Ethiopia is one of the countries in ``meningitis belt``, recently there has been an report on an outbreak of meningococcal meningitis in southern Ethiopia[6].

The relative frequency of isolation of various bacterial species as a cause of meningitis varies with age and geographic regions. S. *pneumonia*, N. *meningitides*, and H. *influenza* type b are the most common worldwide causes of bacterial meningitis in infants and young children[7,8]. Other bacteria that causes meningitis includes S. *aureus*, S. *agalactiae*, E.*coi*, K. *pneumonia*, S. *pyopens*, *Salmonella* species, P. *aeruginosa*, *Enterobecter* species, *Acinetobacter* species, L. *monocytogens* and other gram negative bacilli[9-14].

Antimicrobial resistance of bacteria is a worldwide problem however, the situation in developing countries like Ethiopia is particularly serious.[12] Bacterial meningitis has for a long time been treated with a combination of penicillin or ampicillin and chloramphenicol, and this combination is still the widely recommended as a first choice in most of Africa countries. However the increasing frequency of reports of bacterial resistance in vitro to this drug has raised concern that this choice may not be longer appropriate[15-17].

Because of the absence of well organized laboratories, inadequacy of vaccination, unavailability of antimicrobial agents and lack of proper surveillance exists on bacterial meningitis and antimicrobial resistance continues to be a great challenge in Ethiopia[18].

Since the presence of drug resistance of bacteria in the community is a great threat for public health and because of ever increasing number of drug resistant strain with time. Up to date information on pathogens and their antimicrobial resistance pattern is very essential for empirical treatment of patients, to monitor drug resistance, for planning the effective use of drugs in the region. The purpose of this study was, therefore, to assess the profile of bacteria that cause meneginitis and antimicrobial susceptibility pattern among meningitis suspected neonates and children.

Materials and methods

Study design, area and period, population

A retrospective study was conducted at University of Gondar Teaching Hospital. This zone is located in the highland of Northwest Ethiopia which lay with on the eastern end of the traditional meningitis belt in Africa (Zonal statistical office). Gondar has an elevation of 2135 meter above sea level. According to the National population survey of 2008 the city has a population 231,977 people. This study was conducted from November 2012-April 2013. A total of 2170 CSF specimen from neonates and children patients who visited University of Gondar Teaching Hospital Laboratory and for bacteriological culturing and antimicrobial susceptibility testing were included in the study from September 1, 2002 to January 30, 2012.

Data collection method

Data was collected manually by using a pre-prepared data abstraction format from the department of clinical bacteriology registration book on which laboratory findings after investigation of CSF are recorded.

Specimen collection and processing

CSF samples were collected as part of the routine clinical management of patients admitted in Pediatric and Neonatal wards of the hospital. The samples were collected in sterile containers by attending physicians and delivered to the Bacteriology Laboratory within quarter of an hour after collection. The samples were processed following the standard microbiological procedures by inoculating on blood agar, chocolate agar, and MacConkey agar plates (Oxoid Ltd, Hampshire, UK) prepared as per the manufacturers instruction and incubated at 35-37°C aerobically.

The chocolate agar plates were incubated by putting them in the candle jar, which provided 5-10% CO_2 concentration to create a microaerophlic condition for fastidious bacteria. After 18-24 hours of incubation, the plates were examined for the presence of bacterial colonies. Plates, which did not show any growth, were farther incubated for an additional 24 hours. Organisms were identified by standard microbiological methods, which include colony morphology, as well as Gram staining, biochemical and serological tests.[1, 18,19, 20]

Antimicrobial susceptibility test

Antibiotic susceptibility tests were conducted on pure culture isolates employing the disk diffusion method. For the commonly used antibiotics: ampicillin (10 µg), gentamycin (10 µg), penicillin (10 IU), tetracycline (30µg), erythromycin (15µg), chloramphenicol (30 µg), ciprofloxacin (5 µg) and co-trimoxazole (25 µg) (Oxoid Ltd). The diameter of growth inhibition around the disk were measured and interpreted as sensitive, intermediate or resistant as per the guideline set by Bauer, et al.[21] Reference strain: E.coli ATCC 25922 and *S. aureus* ATCC 25923 were tested as controls according to Clinical and Laboratory Standards Institutions (CLSI).[22]

Data processing and analysis

Data was checked, coded and entered to SPSS 16 software version. Checked data was analyzed for descriptive statistics and the finding was presented in the text and tables.

Ethical consideration

Ethical clearance was obtained from Schools of Biomedical and Laboratory Science. Permission was obtained from the hospital officials at the time of study.

Result

Two thousand one hundred seventy of meningitis suspected patient`s CSF culture were processed routinely from September 2002 to January 2012 in Microbiology Laboratory. Of these 1304 (60.1%) and 866 (39.9%) were male and female; respectively. The age range was from 0 day- 14 years). Majority of patients 672 (31%) were in the age range of 1-5 yrs age. Male to female ratio was 1.5 to 1 (Table 1).

Table No. 01: Age and sex frequency distribution of pediatric patients requested for CSF culture in University of Gondar Teaching Hospital September 1, 2002- 2012).

	Frequency		
	Sex		
Age in year	**Male**	**Female**	**Total**
<28day	235(10.8)	249(11.5)	**484(22.3)**
28-day-1	359(16.5)	203(9.4)	**562(25.9)**
1-5Yrs	410(18.9)	262(12.1)	**672(31)**
5-10Yrs	212(9.8)	117(5.4)	**329(15.2)**
10-14Yrs	88(4.1)	35(1.6)	**123(5.7)**
Total	**1304(60.1)**	**866(39.9)**	**2170(100)**

Out of 2170 clinically suspected cases of bacterial meningitis admitted over a span of 10 years, diagnosis of bacterial meningitis were confirmed by culture in 97 (4.5%) cases. About 52 (54.7%) of all the isolated were gram positives while 43(45.3%) were gram negatives. S. *pneumoniae* were the predominant group of organism recovered from CSF culture 35 (36.1%), followed by N. *meningitidis* 28(28.9%), *Haemophilus* species 9(9.3%), *S. agalactiae* 8(8.2%), S. *aureus* 4 (4.1) (Table 2).

Table No. 02: Bacterial profile isolated from culture of CSF, among meningitis suspected patients of Neonates and Children at University of Gondar Teaching Hospital September 1, 2002- January 30, 2012.

Organisms	Number (%)	Patient category & isolates frequency					
		Neonate (<28 days) n			Children (>28days_14 years) n		
		M	F	T	M	F	T
S. *pneumonia*	35(36.1)	3	0	3	19	13	**32**
N. *meningitides*	28(28.9)	0	2	2	19	7	**26**
Streptococcus species	8(8.2)	0	0	0	4	4	**8**
S. *aureus*	4(4.1)	0	1	1	2	1	**3**
***Haemophilus* species**	9(9.3)	0	0	0	7	2	**9**
S. *epidermidis*	2(2.1)	0	0	0	1	1	**2**
E. *coli*	3(3.1)	1	1	2	0	1	**1**
L. *monocytognes*	2(2.1)	0	0	0	0	2	**2**
S. *agalactiae*	2(2.1)	0	0	0	0	2	**2**
***Pseudomonas* species**	1(1)	0	0	0	1	0	**1**
***Salmonella* species**	3(3.1)	0	1	1	1	1	**2**

M=male, F=Female, T=total

Among gram positive organisms S. *pneumoniae* showed a high level of drug resistance against co-trimoxazole 24 (68.6%), gentamycin 23 (65.7%), tetracycline 14 (40%), penicillin 12 (34.3%) and chloramphenicol 9 (25.8%). S. *aureus* also showed a high level of drug resistance against both ampicillin, penicillin and chloramphenicol 3(75%) each, tetracycline 2(50%) and no S. *aureus* strain were resistant to gentamycin and ciprofloxacin. S. *epidermidis* was found to be resistant only to penicillin. Of the gram negative bacteria N. *meningitides* was found to be resistant to co-trimoxazole 24(85.8%), tetracycline 19(67.8%), ampicillin 9(32.2%), gentamycin 7(25%) and penicillin 6 (21.5%) (Table 3).

Table No. 03: Antimicrobial susceptibility pattern of bacterial isolates from CSF of pediatric patients University of Gondar Hospital September 1,2002- January 30, 2012.

Organisms		**Antimicrobial agent**							
		AMP	**TTC**	**SXT**	**CAF**	**PEN**	**GN**	**CIP**	**E**
S. *pnumoniae*	S	30(85.7)	21(60)	11(31.4)	26(74.2)	23(65.:	12(34.3)	28(80)	**31(88.5)**
	R	5(14.3)	14(40)	24(68.6)	9(25.8)	12(34.3)	23(65.7)	7(20)	**4(11.5)**
N. *meningitides*	S	19(67.8)	9(32.2)	4(14.2)	23(82.1)	22(78.5)	21(75)	19(67.8)	**21(75)**
	R	9(32.2)	19(67.8)	24(85.8)	5(17.9)	6(21.5)	7(25)	9(32.2)	**7(25)**
Streptococcus spp.	S	5(62.5)	5(62.5)	4(50)	6(75)	5(62.5)	4(50)	6(75)	**7(87.5)**
	R	3(37.5)	3(37.5)	4(50)	2(25)	3(37.5)	4(50)	2(25)	**1(12.5)**
s. *aureus*	S	1(25)	2(50)	3(75)	1(25)	1(25)	4(100)	4(100)	**3(75)**
	R	3(75)	2(50)	1(25)	3(75)	3(75)	0	0	**1(25)**
***Haemophilus* spp.**	S	5(55.5)	5(55.5)	6(66.6)	9	9	9	9	**7(77.7)**
	R	4(44.5)	4(44.5)	3(33.4)	0	0	0	0	**2(12.3)**
S. *epidermidis*	S	2	2	2	2	0	2	2	**2**
	R	0	0	0	0	2	0	0	**0**
E. *coli*	S	0	2(100)	2	2	0	3	3	**0**
	R	3(100)	1	1	1	0	0	0	**0**
	S	2(100)	0	2	2	2	2	0	**2**
S. *agalactae*	R	0	0	0	0	0	0	0	**0**

Among gram positives organisms S. *aureus* were a higher level of multi drug resistance 3(75%) followed by S. *pneumoniae 24(68.5%) and* streptococcus spp. 5 (62.5%) while *Salmonella* species 3(100%) showed high level of multidrug resistance among gram negatives (Table 4).

Table No. 04: Lists of multi drug resistance pattern for each bacterial isolates of pediatric patients at University of Gondar Teaching Hospital September1, 2002- January 30, 2012.

Isolated organism	**Resistance type (n %)**						
	R0	R 1	R2	R 3	R 4	R 5	**R 6**
S. *pneumoniae*	5(14.3)	6(17.1)	9(25.7)	5(14.3)	3(8.6)	1(2.9)	**4(11.4)**
N. *meningitidis*	3(10.7	7(25)	5(17.9)	0	8(28.6)	3(10.7)	**0**
Streptococcus spp.	0	3(37.5)	1(12.5)	2(25)	1(12.5)	0	**1(12.5)**
S. *aureus*	1(25)	0	0	2(50)	0	0	**1(25)**
***Hemophilus* spp.**	2(22.2)	1(11.1)	2(22.2)	0	0	1(11.1)	**1(11.1)**
S. *epidermidis*	0	1(50)	0	0	0	0	**0**
E. *coli*	0	1(33.3)	0	1(33.3)	1(33.3)	0	**0**
L. *monocytogens*	0	0	0	0	0	0	**0**
***Salmonella* spp.**	0	0	0	0	0	2(66.6)	**1(33.3)**
***Pseudomonas* spp.**	**0**	**0**	**0**	**1(1)**	**0**	**0**	**0**

Key: R0 – no resistant to any of antibiotics
R1 – resistant to only one antibiotics
R2 – resistant to two antibiotics
R3 – resistant to three antibiotics
R4 – resistant to four antibiotics
R5 – resistant to five antibiotics
R6 – resistant to six and above

Discussion

Now a day's despite increase availability of potent antimicrobial and sophisticated intensive care units, bacterial meningitis continues to be a significant cause of morbidity and mortality. This is reflected by the fact that the global burden of the disease in childhood is at least 1.2 million cases of bacterial meningitides every year with 1,35,000 death. [3,4] The highest burden of meningococcal meningitis occurs sub-Saharan Africa, in an area that is referred to as the meningitis belt which has an estimated total population 300 million.[5]

The successful management of patients suffering from bacterial illness depends up on the identification of the types on organism that cause the disease and the selection of an effective antibiotic against the organism in question.[17] Thus, the data presented in this study could provide information of immediate public health importance to clinicians in northwest Ethiopia on the selection of antimicrobial agents for the treatment of patients suffering from acute bacterial meningitis.

The isolation rate of bacteria found in the present study is comparable to a previous study conducted in Gondar, where an isolation rate of 5.2, 5.6%.[9, 18] However, this finding is lower than studies done in Chittagong medical college, India, Uganda and B.S medical college India, where an isolation rate of 14.37% ,44.12% , 62.5% , and 69.26% were reported respectively.[23, 24 , 25, 26] The reason for low yield of bacterial isolates on culture in this study might be due to over clinical diagnosis of meningitis of all febrile neonates, neonates and children has done unnecessary lumbar puncture, delay in transport of specimens to the laboratory, non availability of special media for specific pathogens in the laboratory setting, fastidious nature of organisms, and antibiotic treatment prior to lumbar puncture. On the contrary, the rate of isolation in our study is higher than studies conducted at Tehran university of medical science and Lokmanya Tilak municpal medical college where isolation rate of 2.9 % and 1.7% respectively.[24, 25] The reason for this is not clear but, it might be epidemiological nature of the organisms and inaccessibility of vaccination of S. pneumonia, Neisseria meningitides in our country.

The predominant aetiological agents for meningitis in our study were Gram positive organisms. This is similar to a study in Iran.[24] To the contrary of our study, Gram negative bacteria were predominant isolate in CSF culture of the study in Gondar.[18] This may be due to the latter study done in neonates, children and adults.

S. *pneaumoniae* and N. *meningitides* were the commonest pathogens causing meningitis which is similar with the previous study at University of Gondar and Chittagong medical college.[18, 19] S. *pneaumoniae* and N. *meningitides* and Haemophilus influenzae type b were commonly reported from CSF culture in most cases of meningitis in Ethiopia.[18, 27] Recently, Haemophilus influenza type b (Hib) once a common cause of meningitis, has disappeared in developed nations, reflecting that the success of Hib vaccination. However, Hib remains a significant public health problem in medical supply constraint countries like Ethiopia.

In this study, *S. pneumoniae* was the predominant bacteria unlike studies in Gondar, N. meningitides, in Uganda, Haemophilus spp. and in Iran, Coagulase negative staphylococci.[18, 24, 26] Of 9.3% meningitis case was caused by *Haemophilus* species in the present finding. This is much higher than a study reported in Gondar, which was a single isolate 1(0.45%).[18] The reasons for the higher prevalence of *Haemophilus* spp. in the present study might be due to large sample size, availability of enriched culture media for the growth of this fastidious organism, observer's skill and experience.

The study also provides insights into the resistance pattern of bacterial isolates in CSF. Resistance of S. *pneumoniae*, N. *meningitides*, and H. *influanzae* to commonly prescribed antibiotics has been reported in different areas of the country.[23] But much is not known about the drug resistance behavior of S.*pneumoniae* and N. *meningitides* from northwest Ethiopia.[18] S. *pneumoniae* in the present study were found to have high levels of drug resistance against co-trimoxazole, gentamycin, tetracycline, penicillin and chloramphenicol (Table 2). We found that 34.3% of S. pneumoniae isolates were resistant to penicillin. However, no S. pneumoniae was resistant to penicillin in a study conducted in Gondar previously.[18] This is due to may be bacteria

Mulat Dagnew. et al., Int. J. Pharm. & H. Care Res., Vol.–01 (02) 2013 [46 - 52]

through time developing resistance for drugs or miss use of penicillin in the study area.

Only one N. *meningitides* strain was resistant to ciprofloxacin in the present study which is similar with a study reported at Chittagong Medical College.[24] The susceptibility of the organism isolated to the fluoroquinolones was generally good in the present study. Although there is limited experience with the use of fluoroquinolones in childhood meningitis as these are still not generally recommended for use in early children in our country. It may be used as potential second line drugs.

A study of in vitro antimicrobial susceptibility profile of the aetiological agents of meningitis has revealed that there is a growing emergence of multi-drug resistant microbes. Majority of the isolates 61. 9% showed multiple antibiotic resistance (Table 3). Multidrug resistances were observed in the present study which is inconsistent with a study at India.[25] The highest resistance rate was noted for co-trimoxazole and tetracycline. The reason may be due to, Co-trimoxazole in our hospital has been repeatedly prescribed empirically for children and extravagantly used. The highest multiple drug resistance was observed among Gram negative bacteria. Salmonella species showed highest multiple antibiotic resistance which were 100%. In our study, similar to the previous report, antibiotic resistance among gram negative bacteria were frequent.[17]

The increasing number of multi resistant strains of bacteria is currently one of major challenges. This situation raises serious concern. This suggests a very high resistance gene pool due perhaps to gross misuse and inappropriate usage of the antibacterial agents.

Conclusion and recommendation

S. pneumoniae, Neisseria meningitides and *Haemophilus* species were the most common bacterial aetiologic agents for meningitis isolated from CSF culture in this study. The frequency of multidrug resistance is alarmingly high. This might be reflection of the inappropriate use of antibiotics, or unavailability of a guide line regarding the selection of drugs. Producing updated information on local pathogens and their susceptibility patterns is a prime tool in combating drug resistance problem in a region. We recommended further study with a better design, and survey of antimicrobial susceptibility at large scale to control the spread of antibiotic resistance bacteria at national level.

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References

1. Cheesborough M. District laboratory practice in tropical countries part II Cambridge university press 2002, 116-124.
2. Richard EB, Nelson textbook of pediatrics 16th ed. W.B Saundes company .New York USA, 2000.P.707.
3. Weber Mw, Herman J, Jeffers. Clinical pediatrics of bacterial meningitis in infants and young children in Gambia. Trop med Int Health 2002; 7:722-31.
4. Akepeda G, Adeyemi O, Abba A, sykes RM. Pattern and antibiotic susceptibility of bacterial in pynogenic meningitis in a children's emergency room population in Maiduguri, Nigeria, 1988-1992. Acta padiater 1994; 83:719-23
5. World Health organization. Control of epidemic meningococcal disease. WHO practical Guidelines 2ed .1998. Geneva Switzerland.
6. Foreign & Commonwealth Office (FCO)/Health. Report on Meningococcal Meningitis outbreak in southern Ethiopia. http://www.fco.gov.uk/en/travel-and-living-abroad/travel-advice-by-country/sub-saharan-africa/ethiopia accessed on March 20,2013.
7. Schuchet A, Robihsonk, Wenger ID. Bacterial meningitis in the united states in 1995 active surveillance team N Engl J med 1997;337:970-6.
8. Dawson KG, Emerson Jc, Aurns Jl. Fifteen years of experience with bacterial meningitis peadiatr Dis j infect 1999;18:816-22.
9. Centers for Disease Control and Prevention. Diminishing racial disparities in early onset neonatal group B streptococcal disease United States, 2000-20003. MMWR Morb Mortal WKLY Rep2004; 53:502 - 5.

10. Moreno MT, Vargas S, Poveda R, Saez-llovens X. Neonatal Sepsis and meningitis in a developing Latin American country. Perditra infect Dis J 1994, 13, 516-20.
11. Laving Am, Musoke RN, Wasunn Ao, ARevathi G. Neonatal bacterial meningitis at the new born unit of Kenyatta National Hospital. East Afr med J2003; 80:456-62.
12. Osrin D, vergnano S, fanaroff AA, Lemons JA. Entrobacter Sakazakii is a rare cause of neonatal septicemia or meningitis in VLBW infants. Jpediatra 2004; 144:821-3.
13. Center for Disease control and prevention. Entrobacter sakazakii infection associated with the use of powdered infant formula-Tennessee, 2001. JAMA2002; 287:2204-5.
14. Costello A. Serious bacterial infection in new born infants in developing countries. Infect Dis 2004; 17:217-24.
15. World Health Organization. Control of epidemic meningococcal disease. WHO Guidelines 2ed 1998.Genava Switzerland.
16. Aseffa A, Yohannes G. Antibiotic sensitivity pattern of prevalent bacterial pathogens in Gondar Ethiopia. East Afar Med J. 1996; 73(1):67-71.
17. Golnez R, Babak P, Mohammed H. Antimicrobial susceptibility of bacterial isolated from CSF in an Iranian referral pediatric center, 1998-2008. A journal of clinical medicine 2012; 7(2):131-137.
18. Mulu A, Kassu A, Tessema B. Bacterial isolates from CSF and their antibiotic susceptibility patterns in University of Gondar Teaching Hospital, Northwest Ethiopia. Etiop. J health dex 2005; 19(2):161 - 164.
19. Jawetz MA. Medical Microbiology. 19th ed. Prentice-hall International Inc: Antibacterial and antifungal chemotherapy pp.149 - 179.
20. World Health Organization. Basic laboratory procedures in clinical bacteriology, WHO, Geneva, Switzerland, 1991:78-95.
21. Bauer AW, Kirby WM, Sherirs JC, Turck M. Antibiotic susceptibility testing by standard single disk method. Am J Clin Pathol, 966, 45(4), 433-496.
22. National Committee for Clinical Laboratory Standards. Performance standards for antimicrobial disc susceptibility tests. Approved standard, ASM-2(2ded.) NCCLS, Villanovan, pa.1979.
23. Yashpal C, Arun k, Anodita B. Antimicrobial sensitivity pattern of gram positive CSF isolates in children with septic meningitis in Tertiary Care Hospital. Internet Journal of medical up date publisher 2011, 6(2), 1 - 13.
24. Alka E, Son V, Baradkar VP, M mathus. Pattern and antibiotic susceptibility of bacteria isolated in clinically suspected cases of meningitis in children. J peditra neurosci 2008; 3:131 -133.
25. Modi GB, Patel KD, Soni ST, Patel KJ, Mangukiya JD, Jain PS. Bacteriological profile of pyogenic meningitis in Tertiary Care Hospital, Ahmedabad. National Journal of medical research 2012, 2(3),313 - 317.
26. Julius P, Juliet M. Childhood bacterial meningitis in Mubarara Hospital, Uganda; anti microbial susceptibility and outcome of treatment. African health sciences 2001; 1(1):9 - 11.
27. Hailu M, Muhe L. Childhood meningitis in a tertiary hospital in Addis Ababa: Clinical and epidemiological features. Ethiop Med J 2001; 39:29-38.