

## Prevalence and antibiotic resistance profiles of cerebrospinal fluid bacteria in children- Benghazi

\*Noor-Alhoda Milood Al-Awkally<sup>1</sup>, Maree Dokally Ali<sup>2</sup>, Rabea Bashir Al- Fakhari<sup>3</sup>, Alaaedin.M.S.Eldrolli<sup>4</sup>, Nesrine Miloud Alawkally<sup>5</sup>, Miftah S. M. Nag<sup>6</sup>, Alreda Miloud Al-Awkally<sup>7</sup>, Abeer Miloud Al-Awkally<sup>8</sup>, Muftah Abdulwahed Nasib<sup>9</sup>, Mohamed Ahmed Ahwidy Ali<sup>10</sup>

<sup>1</sup>Medical Laboratory Department, High institutes for comprehensive professions, Suluq

<sup>2</sup>Medical Laboratory Department, High institutes for comprehensive professions, Suluq

<sup>3</sup>Medical laboratory, Paediatric Hospital, Benghazi

<sup>4</sup>Department of Computer Technology, High institutes for comprehensive professions, Gamins

<sup>5</sup> Surgery Department, Al haowari hospital, Benghazi

<sup>6</sup>Faculty of Biomedical Technology, University of Benghazi

<sup>7</sup>Pharmacy Department, Ministry of health, Darna

<sup>8</sup>Veterinary Clinic, Ministry of Agriculture, Livestock- Darna

<sup>9</sup>Medical laboratory, High institute of comprehensive professions, Gamines

\*Corresponding author: [noornoor1973@gmail.com](mailto:noornoor1973@gmail.com)

**Abstract** Bacterial meningitis is a fatal, restricting endemic disease requiring rapid antibiotic controlling. The objective of this study was to evaluate the antimicrobial sensitivity patterns of microorganisms isolated from CSF to antibiotics. Of 3184 CSF were cultured and sensitivity tests were performed in microbiology department, Pediatric hospital, Benghazi, Libya. The study period was during January 2016 to December 2017. A total 3184 CSF samples, 3149 (99%) CSF samples had no bacterial growth, while 35 (1%) CSF samples showed bacterial growth. 20 (57%) were males and 15 (43%) were females. *Staph epidermidis* 9 (26%) was more bacteria isolated than patient samples, followed by Gram negative bacteria 7 (21%). Most of the cases were recorded in 28 (80%) winter, followed by 7 (20%) autumn. The bacteria have recorded high sensitivity to chloramphenicol by 23%, followed by Ciprofloxacin 14%, while the highest bacterial resistance was recorded to Septrin by 31%. The early diagnosis and providing treatment early are lifesaving and they reduce chronic morbidity.

**Keywords:** Cerebrospinal fluid, Chloramphenicol, Antimicrobial resistance, Meningitis, Paediatric hospital, Benghazi city.

## ملاحق انتشار ومقاومة المضادات الحيوية لبكتيريا السائل النخاعي عند الأطفال - بنغازي

\*نور الهدى ميلود العوكلي<sup>1</sup> و مرعي الدوكالي علي<sup>2</sup> و ربيعة بشير الفاخري<sup>3</sup> و علاء محمد الدرولي<sup>4</sup> و نسرين ميلود العوكلي<sup>5</sup> و مفتاح

سعد نجم<sup>6</sup>، الرضا ميلود العوكلي<sup>7</sup> و عيبر ميلود العوكلي<sup>8</sup> و مفتاح عبد الواحد نصيب<sup>9</sup> و محمد احمد اهوودي<sup>10</sup>

<sup>1</sup> قسم المختبرات الطبية-المعهد العالي للمهن الشاملة، سلوق، ليبيا

<sup>2</sup> قسم المختبرات الطبية-المعهد العالي للمهن الشاملة، سلوق، ليبيا

<sup>3</sup> قسم المختبرات-جامعة بنغازي، ليبيا

<sup>4</sup> الصيدلانية المركزية-وزارة الصحة، درنه، ليبيا

<sup>5</sup> العيادة البيطرية-وزارة الثروة الطبية، مستشفى الاطفال، بنغازي

<sup>6</sup> قسم تكنولوجيا الحاسوب، المعهد العالي للمهن الشاملة، قمينس، ليبيا

<sup>7</sup> قسم الجراحة-مستشفى الهوارى، بنغازي، ليبيا

<sup>8</sup> كلية التقنية الحيوية الحيوانية والبحرية-درنه، ليبيا

<sup>9</sup> قسم الاحياء الدقيقة-كلية العلوم-جامعة عمر المختار، البيضاء، ليبيا

<sup>10</sup> قسم المختبرات الطبية، المعهد العالي للمهن الشاملة، قمينس، ليبيا

**المخلص** التهاب السحايا الجرثومي هو مرض مستوطن مميت ويتطلب السيطرة السريعة عليه باستخدام المضادات الحيوية. كان الهدف من هذه الدراسة هو تقييم أنماط تحسس البكتيريا المعزولة من السائل النخاعي للمضادات الميكروبية. تم زرع 3184 عينة سائل نخاع شوكي واجراء اختبار الحساسية في معمل الاحياء الدقيقة بمستشفى الأطفال ، بنغازي ، ليبيا. فترة دراسته كانت ما بين يناير 2016 إلى ديسمبر 2017. إجمالي 3184 عينة من السائل الدماغى النخاعي، 3149 (99%) عينة من السائل الدماغى النخاعي لم يكن بها نمو بكتيري، بينما 35 (1%) كان بها نمو بكتيري. إجمالي عدد الاصابات كانت 35 حالة، الذكور كان عددهم 20 (57%) ذكور والاناث

15 (43%). كانت بكتيريا *Staph epidermidis* أكثر عزلاً للبكتيريا من عينات المرضى بنسبة 9 (26%)، تليها البكتيريا سالبة صبغة جرام 7 (21%). تم تسجيل معظم الحالات في الشتاء بنسبة 28 (80%)، تليها 7 (20%) في فصل الخريف. سجلت البكتيريا حساسية عالية للكلورامفينيكول بنسبة (23%)، تليها سيبروفلوكساسين (14%)، بينما سجلت البكتيريا أعلى مقاومة للسبترين بنسبة 31%. التهاب السحايا الجرثومي هو حالة طبية طارئة وإجراء التشخيص المبكر وتقديم العلاج في وقت مبكر ينقذ الحياة ويقلل من معدلات الاعتلالات المزمنة.

**الكلمات المفتاحية:** سائل النخاع الشوكي، مقاومة المضادات الحيوية، كلورامفينيكول، السحايا، مستشفى الأطفال، مدينة بنغازي.

## Introduction

Acute bacterial meningitis is a primary cause of death and debility worldwide. [1] Every year, over one million people are affected, with the incidence are greater among developing countries and in specific geographic areas. Meningitis is a widespread disease in Egypt; *S. pneumonia* meningitis is now the prominent cause of meningitis in Egypt and has the highest death rates between meningitis patients especially in patients less than one year of age. [2-3] Differentiating bacterial from nonbacterial meningitis is very important in determining treatment. Bacterial meningitis is a lethal neurological illness and needs quick antibiotics treatment, acute meningitis is caused by a variety of infectious agents. Pyogenic bacteria, such as *S. pneumoniae*, *N. meningitidis*, and *H. influenza* are the most causes of meningitis. [4] Gram stain smears of the CSF permits a rapid, correct method of diagnosis of bacterial meningitis in 60%–90% of patients. [6] CSF culture is highly specific. [5] The common CSF anomalies in bacterial meningitis are a polymorphonuclear leukocytosis, increased protein concentration and reduced glucose concentration. In viral meningitis, the usual CSF anomalies are a lymphocytic pleocytosis, a normal glucose concentration, and a normal or slightly elevated protein concentration. [7] Diagnostic tests are needed to differentiate between bacterial and viral meningitis. The peripheral WBC count, CRP, and ESR are usually elevated in patients with bacterial meningitis. [8] Aims of the present study was to evaluate of the bacterial meningitis and the antibiotic resistance of alarmed bacteria over the last two years in Pediatric hospital- Benghazi.

## Material and Methods

This Laboratory-based retrospective analysis of 3184 CSF cultures and sensitivity tests was conducted in Paediatric hospital Laboratory- Benghazi with- in a two year period between January 2016 and December 2017. The samples were collected from different wards of the hospital in sterile containers by physicians and delivered to the bacteriology department within half an hour collection and samples were processed following the standard microbiological procedures by inoculating on blood agar, chocolate agar, and Macconkey agar plates prepared as per the manufacturer instruction and incubated at 35- 37C° aerobically. The chocolate agar plates were incubated by putting them in a candle jar, which provided 5-10% CO2 concentration to create a microaerophilic condition for fastidious bacteria. After 20-24 hours of incubation, the plates were examined for

the presence of bacterial colonies. Plates, which did not show any growth, were further incubated for an additional 24 hours. Organisms were identified by standard microbiological methods, which included colony morphology, as well as staining, biochemical and serological tests. [15] The Kirby-Bauer disk diffusion method was performed to determine the antibiotic susceptibility in vitro susceptibility of Gram positive bacteria and Gram negative bacteria isolates against antibiotics were determined by the standard disc diffusion procedure. [15] Commercially prepared antibiotic discs (Oxoid, UK). (6 mm in diameter) belonging to different groups antibiotics were used: A-ampicillin (10 µg), Ofloxacin (5 µg), AMC-Amoxicillin Clavulonic Acid (30ug), G-Gentamicin (10Fg), Va-Vancomycin (25 µg), AK-Amikacin (30µg), CAR-Carbapenem (30µg), E-Erythromycin (15 µg), PRL-Plaritromicin (100µg), Ox-Oxacillin (6 µg), SXT-Sulfamethoxazole (25µg), CT-Colistin (10Fg), Cip- ciprofloxacin (5µg), Ch-chloramphenicol (10 µg) (Oxoid Ltd). The diameters of inhibition zone around the discs were measured and interpreted as sensitive, intermediate or resistant as per the guideline set by Bauer, *et al* (Bauer *et al.*, 1966).

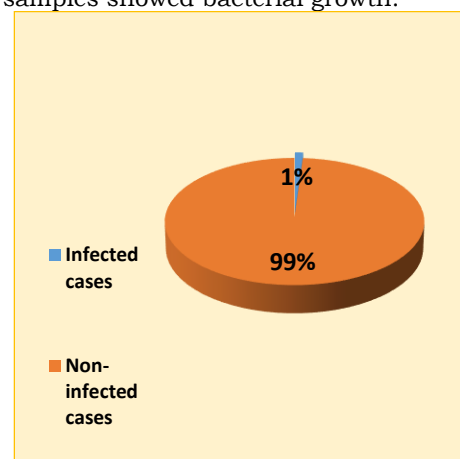
**Statistical analysis:** - Analysis was done by the standard deviation function is STDEV, and the test uses is T- test.

## Results

Three thousand and one hundred eighty four (3184) cases were examined using culture in bacteriology department, Paediatric hospital, Benghazi.

### 4.1 Cases distribution according to bacterial growth:

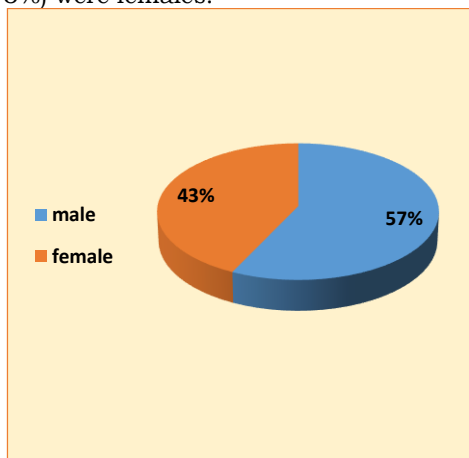
A total 3184 CSF samples, 3149 (99%) CSF samples had no bacterial growth, while 35 (1%) CSF samples showed bacterial growth.



**Fig. 1:** Cases distribution according to bacterial growth

**4.2 Distribution of affected cases according to gender:**

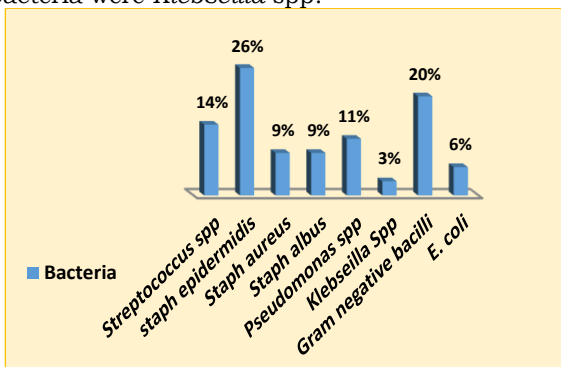
Of 35 of affected cases, 20 (57%) were males and 15 (43%) were females.



**Fig. 2:** Distribution of affected cases according to gender

**4.3 Organisms isolate from CSF cultures of patients of bacterial meningitis in paediatric hospital Benghazi during 2016-2017.**

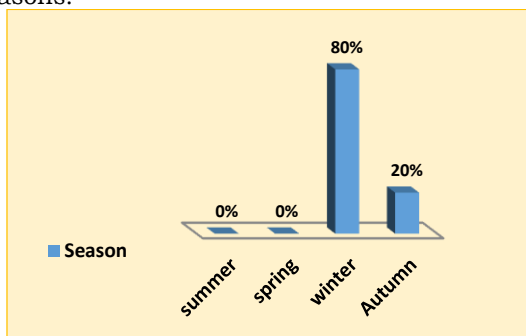
*Staph epidermidis* 9 (26%) was more bacteria isolated than patient samples, followed by Gram negative bacteria 7 (21%). While the least isolated bacteria were *Klebsiella* spp.



**Fig. 3:** Organisms isolate from CSF cultures of patients of bacterial meningitis in paediatric hospital Benghazi during 2016-2017.

**4.4 Seasonal variations of the isolated bacterial meningitis from CSF at Paediatric hospital from January 2016 to December 2017**

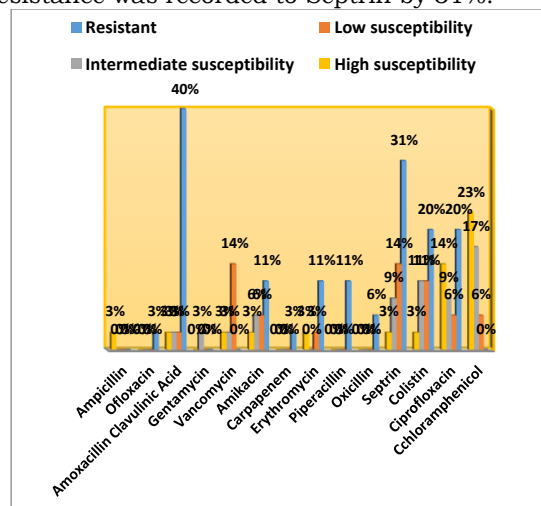
Most of the cases were recorded in 28 (80%) winter, followed by 7 (20%) autumn. There are no recorded cases in the spring and summer seasons.



**Fig. 4:** Seasonal variations of the isolated bacterial meningitis from CSF at Paediatric hospital from January 2016 to December 2017

**4.5 Susceptibility patterns of different bacteria isolated from CSF at paediatric hospital from January 2016 to December 2017:**

The bacteria have recorded high sensitivity to chloramphenicol by 23%, followed by Ciprofloxacin 14%, while the highest bacterial resistance was recorded to Septrin by 31%.



**Fig. 5:** Susceptibility patterns of different bacteria isolated from CSF at paediatric hospital from January 2016 to December 2017.

**Discussion**

Acute bacterial meningitis is still considered one of the most dangerous infectious diseases in children. Three thousand and one hundred eighty four suspected meningitis cases were examined using culture in microbiology department. A total 3184 CSF samples, 3149 (99%) CSF samples had no bacterial growth, while 35 CSF samples showed bacterial growth (1%). Bacterial isolation and identification were attempted for 35 specimens. The number of females infected with meningitis was more than males 20 (57%) than females 15 (43%).

The most bacterial species identified was *staph epidermidis* (26%) and Gram negative bacteria (21%). This result was similar to the results of other studies were found *Staph Epidermidis* was the most isolated bacteria. [9, 18] While another study found that the most pathogenic organism was Gram negative bacteria as etiological agents of bacterial meningitis.[17] contrary to other studies that found *Neisseria meningitidis* and *Streptococcus pneumonia* were the most isolated pathogen.[10, 11,13, 14] The present study reported that, Chloramphenicol and Ciprofloxacin were the most antagonists that affect the types of bacteria significantly, the result of our study was similar to a study in which ciprofloxacin had a high effect on *Klebsiella* spp. [16]

**Conclusion**

Early and correct diagnosis and suitable treatment of bacterial meningitis in children remains a major challenge, as reflected by the continued high morbidity and case-fatality rates of the disease worldwide. Right use of antibiotics has proved helpful in the avoidance of neurologic

consequence in children with bacterial meningitis. We should focus on avoidable features of vaccines, to reduce the disease problem.

#### Acknowledgment

The corresponding author would like to thank the entire microbiologist staffs who have assisted in document in Microbiology department, Pediatric hospital, Benghazi..

#### References

- [1]- Agier, L., Broutin, H., Bertherat, E., Djingarey, M. H., Lingani, C., Perea, W., S. Hugonnet., (2013). Timely detection of bacterial meningitis epidemics at district level: a study in three countries of the African Meningitis Belt. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, 107(1):30–36.
- [2]- Farag, H. F. M., Abdel-Fattah, M. M., Youssri, A. M., (2005). Epidemiological, clinical and prognostic profile of acute bacterial meningitis among children in Alexandria, Egypt. *Indian Journal of Medical Microbiology*, 23(2):95–101.
- [3]- Afifi, S., Wasfy, M. O., Azab, M. A., Youssef, F. G., Pimentel, G., Graham, T. W., Mansour, H., Elsayed, N., Earhart, K., Hajjeh, R., Mahoney, F., (2007). Laboratory-based surveillance of patients with bacterial meningitis in Egypt (1998–2004) *European Journal of Clinical Microbiology and Infectious Diseases*, 26(5):331–340.
- [4]- Adriani K. S., Van de, D., Brouwer, M. C., Spanjaard, L, Gans, J., (2007). Community-acquired recurrent bacterial meningitis in adults. *Clinical Infectious Diseases*, 45(5):e46–e51.
- [5]- Nigrovic, L. E., Malley, R., Macias, C. G *et al.*, (2008). Effect of antibiotic pretreatment on cerebrospinal fluid profiles of children with bacterial meningitis. *Pediatrics*, 122(4):726–730.
- [6]- Allan, R., Tunkel, B. J., Hartman, S. L., Kaplan, B. A., Kaufman, K. L., Roos, W., Michael, S., Richard, J. W., (2004). *Clinical Infectious Diseases*, Volume 39, Issue 9, 1 November, Pages 1267–1284, <https://doi.org/10.1086/425368>
- [7]- Roos, K. L., (2003). Cerebrospinal fluid. In: Roos KL, editor. *Principles of Neurologic Infectious Diseases*. chapter 1. McGraw-Hill; pp. 1–12.
- [8]- Gerdes, L. U., Jørgensen, P. E., Nexø, E., Wang, P., (1998). C-reactive protein and bacterial meningitis: a meta-analysis. *Scandinavian Journal of Clinical & Laboratory Investigation*, 58(5):383–394.
- [9]- Jiang H *et al.* Prevalence and antibiotic resistance profiles of cerebrospinal fluid pathogens in children with acute bacterial meningitis in Yunnan province, China, 2012–2015. *PLoS One*. 2017 Jun 29;12(6):e0180161.
- [10]- Karou, D *et al.*, (2012). Epidemiology and antibiotic resistance of bacterial meningitis in Dapaong, northern Togo. *Asian Pacific Journal of Tropical Medicine* Volume 5, Issue 11, November, Pages 848–852.
- [11]- Tegene, B., Gebreselassie, S., Fikrie, N., (2015). Bacterial Meningitis: a five-year retrospective study among patients who had attended at University of Gondar Teaching Hospital, Northwest Ethiopia. *Biomedical research and therapy*, 2(5):270–278 issn2198-4093.
- [12]- Aseffa, A., and Yohannes, G., (1996). Antibiotic sensitivity pattern of prevalent bacterial pathogens in Gondar, Ethiopia. *East African medical journal* 73, 67–71.
- [13]- Akpede, O., Abiodun, P., Sykes, M., and Salami, C., (1994). Childhood bacterial meningitis beyond the neonatal period in southern Nigeria: changes in organisms/antibiotic susceptibility. *East African medical journal* 71, 14–20.
- [14]- Bhat, B. V., Verma, I. C., Puri, R. K., Srinivasan, S., Nalini, P., (1991) A profile of pyogenic meningitis in children. *J Indian Med Assoc*, Aug;89(8):224–7.
- [15]- Bauer, A. W., Kirby M W. M., Sherris. J. C., Turck, M., (1996). Antibiotic susceptibility testing by 310 a standardized single disk method. *Tech. Bull. Regist. Med. Technol*, Vol. 36 (3): 49– 311 52.
- [16]- Mengistu, A., Gaeseb, J., and Sagwa, E *et al.*, (2013). Antimicrobial sensitivity patterns of cerebrospinal fluid (CSF) isolates in Namibia: implications for empirical antibiotic treatment of meningitis. *J Pharm Policy Pract*. 2013; 6: 4. Published online 2013 Jun 13. doi: 10.1186/2052-3211-6-4
- [17]- Corman, C. A *et al.*, (1962). Bacterial meningitis. II. Infections caused by certain Gram negative bacteria enteric organisms. *Proceedings of the Staff Meetings of the Mayo Clinic*. 37:703–5
- [18]- Tunkel, A. R., Hartman, B. K. S *et al.*, (2004). Practice guidelines for the management of bacterial meningitis. *Clin Infect Dis*, 39:1267 – 84