

## Occurrence of Pneumococcal Meningitis in Iraq

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### Abstract:

This study focuses on the relative frequency of *Streptococcus pneumoniae* in children with culture-proven bacterial meningitis. The epidemiology has been investigated with clinical and laboratory findings in The Central Public Health Laboratory, Iraq from April to September, 2010. From (138) samples, (55) cases (39.85 %) were diagnosed by physician as bacterial meningitis, (23) cases (16.66 %) as viral meningitis, (18) cases (13.04 %) as different medical cases (e.g febrile convulsion, meningoencephalitis, epilepsy ...etc) and (42) cases (30.43 %) as unidentified medical cases. For bacterial meningitis *S.pneumoniae* was isolated from (10) cases (18.18 %) only. The median age for bacterial meningitis was (130.75) month and the predominance was in the age group (1-20) months which appear as (42) cases (76.36 %). Fever was the frequent presenting symptoms in (49) cases (89.09 %), followed vomiting in (34) cases (61.81 %), then irritability and seizures in (29, 25) cases (52.72, 45.45) % respectively. In bacterial meningitis, the mean for glucose and protein levels was (44 and 65) (mg/dl) respectively.

**Key words:** bacterial meningitis, *Streptococcus pneumoniae*, cerebrospinal fluid.

### Introduction:

Meningitis is inflammation of the protective membranes covering the brain and spinal cord, known collectively as the meninges [1]. The inflammation may be caused by infection with viruses, bacteria, or other microorganisms [2]. The classic triad of diagnostic signs consists of nuchal rigidity, sudden high fever, and altered mental status [3]. In infants up to 6 months of age, bulging of the fontanelle may be present [4]. Seizures may occur for various reasons; in children, seizures are common in the early stages of meningitis [5]. The types of bacteria that cause bacterial meningitis vary by age group. In premature babies and newborns up to three months old, common causes are group B Streptococci and those that normally inhabit the digestive tract

such as *Escherichia coli* (carrying K1 antigen). Older children are more commonly affected by *Neisseria meningitidis*, *Streptococcus pneumoniae* and those under five by *Haemophilus influenzae* type B [1]. The most important test in identifying meningitis is analysis of the cerebrospinal fluid (CSF) through lumbar puncture [6]. The CSF sample is examined for presence and types of white blood cells, red blood cells, protein content and glucose level. Gram staining of the sample may demonstrate bacteria in bacterial meningitis [5]. Various more specialized tests may be used to distinguish between various types of meningitis. A latex agglutination test may be positive in meningitis caused by *Streptococcus pneumoniae* [5].

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## Materials and Methods :

### 1- Collection of patients information's:

All recorded information of meningitis cases have been reported to The Central Public Health Laboratory (from different hospitals which included : Al-Kadhymia Pediatric Hospital , Central Child Hospital, Children Welfare Teaching hospital, Elwiyah Pediatric Hospital, Basrah General Hospital) from April to September, 2010 have been examined for basic demographic information including : age , sex, symptoms (which included : fever, headache, neck rigidity , vomiting , bulging fontanelle , seizures , altered mental status and irritability) , CSF informations (CSF appearance , glucose level , protein level ,count of :white blood cells, red blood cells , polymorphonuclear leukocytes, lymphocytes) , CSF culture and discharge diagnosis).

**2- Collection of CSF :** CSF should be inoculated directly onto both a chocolate agar plate and a blood agar plate. Samples of CSF were collected and immediately transported to the microbiology laboratory, where they should be examined within one hour from the time of collection . If transportation of CSF samples was delayed , the samples were inoculated aseptically into a Trans-Isolate (T-I) medium with a syringe and held overnight at, or close to, (35) °C [7]. One ml of CSF was inoculated into the T-I medium, which has either been pre-warmed in the incubator (37) °C or kept at room temperature (25) °C. The remaining CSF in the syringe was held at room temperature before Gram staining. After inoculation, T-I bottles have been incubated at (35) °C overnight [7].

**3- Diagnosis of bacterial meningitis:** Documentation of the diagnosis by physician was when the CSF examination shows neutrophilic

pleocytosis with raised CSF protein (< 200 mg/dl) and lower CSF sugar (< 40 mg/dl) [9]. A case was diagnosed by physician as viral meningitis when CSF analysis showed lymphocytic pleocytosis , slightly raised CSF protein , normal or slightly reduced CSF sugar. In the presence of negative Gram stain , CSF culture and Latex Agglutination Test with above clinical and laboratory criteria , these cases were considered as viral meningitis [8].

### 4- Primary Culture, Subculture, and Presumptive Identification :

Once the CSF has arrived at the microbiology laboratory, it was centrifuged for (20) minutes at (2000) rpm. The supernatant was drew off , one or two drops of sediment were used to prepare the Gram stain and one drop were used to streak the primary culture media . The agar plates were incubated in a candle-jar. When T-I medium was used for transport, after 24 hours of incubation, with a sterile needle and syringe, it was transferred (100) µl of the liquid portion of T-I onto both a chocolate agar plate and a blood agar plate, and streaked for isolation, then incubated at 35 °C in a candle jar for up to 48 hours [7].

### 5- Identification of *S. pneumoniae* :

**A. Susceptibility to Optochin :** Each isolate was cultured on (5 %) human blood agar plates, then optochin discs were placed on the agar and incubated overnight at (37°C) in a candle jar. Inhibition zones of (14) mm or greater indicate optochin sensitivity [9].

**B. Bile Solubility Test :** Each isolate was prepared as a suspension of bacterial cells from fresh growth on agar plate which were suspended in one ml of saline to make heavy suspension (McFarland 2.0 or greater). The suspension was divided in half in two glass tubes, and (0.5) ml of saline was added to one tube and (0.5) ml of (10 %) deoxycholate was added to the

other. The tubes were incubated up to (30) minutes at (35°C). Lysis of the cells in the tube with deoxycholate indicates a positive test [9].

**C. Latex Agglutination Test :** Each isolate was mixed with one drop of test latex into a separate circle on a reaction card by using separate mixing sticks to gently mix the latex reagent with bacteria and spread mixture over as much of the circle as possible , then it was rocked the card slowly and observed the agglutination within few minutes which considered as positive result.

**D. Microscopic examination of colonies :** Gram stain was done [10].

**E. Biochemical identification with API 20 Strep. :** It was applied according to the manufacturer’s instructions (bioMerieux, France).

**Results and Discussion :**

(138) CSF samples were collected during the period of the study , (82) samples (59.42 %) from males and (56) samples (40.57 %) from females From (138) samples ,(55) cases (39.85 %) were diagnosed by physician as bacterial meningitis, (23) cases (16.66 %) as viral meningitis , (18) cases (13.04 %) as different medical cases (e.g febrile convulsion, meningoencephalitis, epilepsy ...etc) and (42) cases (30.43%) as unidentified medical cases. For bacterial meningitis *S.pneumoniae* was isolated from (10) cases (18.18 %) only.Pneumococci are a major cause of childhood mortality world wide, causing an estimated one million

deaths among children aged < 5 years [11]. The burden of pneumococcal disease is largely under investigation in developing countries [8]. T-I is a biphasic medium that is useful for the primary culture of meningococci and other etiological agents of bacterial meningitis from CSF and blood samples[7] Optochin sensitivity is the most important identification criteria for pneumococcus as showed in the detailed study of [12] who found that typical pneumococci (as defined by optochin sensitivity and bile solubility) , whether capsulated or not , form genetically homogenous group. Males constituted (57.0, 58.5) % of all patients in the studies of [13] and [8] respectively, and this was so close to the males percentage (59.42) % of this study. The three most common etiologic agents are *Haemophilus influenzae* type b ,*S.pneumoniae* and *Neisseria meningitides* , which account for (90 %) reported cases of acute bacterial meningitis in infants and children more than four weeks of age [14]. For bacterial meningitis *S.pneumoniae* was isolated from (10) cases (18.18 %) , this so close to [15] and [13] who found this bacteria caused bacterial meningitis in (15 ,16.7) % respectively of cases. In bacterial meningitis the median age was (130.75) months within range (1-144) month as shown in table (1), the predominance was in age group (1-20) months which appear as (42) (76.36 %).

**Table (1) Cases distribution according to age for bacterial meningitis**

| Age Class (Month) | 1-20    | 21-40  | 41-60  | 61-80 | 81-100 | 101-120 | 121-140 | 141-160 |
|-------------------|---------|--------|--------|-------|--------|---------|---------|---------|
| No.               | 42      | 4      | 5      | 0     | 1      | 1       | 0       | 2       |
| (%)               | (76.36) | (7.27) | (9.09) | (0)   | (1.81) | (1.81)  | (0)     | (3.63)  |

Clinical presentation results for bacterial meningitis showed the frequency of presenting symptoms of

meningitis as shown in table (2) . In bacterial meningitis fever was the frequent presenting symptom which

occurred in (49) cases (89.09 %), followed by vomiting in (34) cases (61.81 %) and irritability and seizures in (29, 25) cases (52.72, 45.45) %. The diagnosis of meningitis in children relies on clinical suspicious and correct interpretation of available laboratory tests including CSF culture. Alternative laboratory methods such as antigen detection and nucleic acid amplification should be integrated into routine laboratory procedures to improve the detection of

microorganisms in CSF [16]. Many children had clinical and / or CSF features that suggested bacterial meningitis; however, due to various contributing factors such as use of antibiotics prior to lumbar puncture, parents' refusal to perform lumbar puncture in some children, and laboratory insufficiencies to isolate microorganisms, the etiology could not be established [17].

**Table (2) Cases demographic data**

| Demographic data | Fever         | Headache     | Neck rigidity | Vomiting      | Bulging fontanelle | Seizures      | Altered mental status | Irritability  |
|------------------|---------------|--------------|---------------|---------------|--------------------|---------------|-----------------------|---------------|
| Case No. (%)     | 49<br>(89.09) | 6<br>(10.90) | 11<br>(20)    | 34<br>(61.81) | 9<br>(16.36)       | 25<br>(45.45) | 21<br>(38.18)         | 29<br>(52.72) |

Fever is the most common chief symptom in infants. The causes of fever in these infants vary from mild viral infections (which are usually self-limiting) to serious bacterial infections (such as urinary tract infections, bacteremia, and bacterial meningitis) which are progressive and lead to permanent neurological sequelae or death without proper antibiotic treatment [18]. The clinical presentation of the present study resembled to that found by [19] who found fever and vomiting by (100 and 61) % respectively. The Biochemical results are shown in tables (3) and (4). In bacterial meningitis the mean for glucose and protein levels were (44 and 75) (mg/dl) within ranges (0.4-106) and (0.16-230) (mg/dl) respectively. Primary care physician frequently perform lumbar puncture, because CSF is an invaluable diagnostic window to the central nervous system [20]. Biochemical results by [19] showed that the mean of glucose was (42) (mg/dl) which was

so close to what was found in the present study (44) (mg/dl).

**Table (3) Cases distribution of glucose level for bacterial meningitis**

| Glucose level (mg/dl) | 0-17      | 18-35       | 36-53        | 54-71       | 72-89     | 90-107     |
|-----------------------|-----------|-------------|--------------|-------------|-----------|------------|
| No. (%)               | 8<br>(20) | 7<br>(17.5) | 11<br>(27.5) | 7<br>(17.5) | 6<br>(15) | 1<br>(2.5) |

**Table (4) Cases distribution of protein level for bacterial meningitis**

| Protein level (mg/dl) | 0-37          | 38-75        | 76-113      | 114-151  | 152-189  | 190-227     | 228-265     |
|-----------------------|---------------|--------------|-------------|----------|----------|-------------|-------------|
| No. (%)               | 10<br>(43.47) | 8<br>(34.78) | 2<br>(8.69) | 0<br>(0) | 0<br>(0) | 2<br>(8.69) | 1<br>(4.34) |

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### حدوث التهاب السحايا البكتيري في العراق

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#### الخلاصه :

هذه الدراسة تركز على بكتريا *Streptococcus pneumoniae* في الاطفال الذين اظهروا نتائج زرعيه ايجابية لحالات التهاب السحايا البكتيري. تم التحري عن الوبائيه ، النتائج المخبريه والطبيه في مختبر الصحه المركزي – العراق للفترة من نيسان الى ايلول 2010. تم جمع ( 138 ) عينه كان منها (55) حاله ( 39,85 %) مشخصه من قبل الطبيب المعالج كحاله التهاب سحايا بكتيري ، ( 23 ) حاله ( 16,66 %) كحاله التهاب سحايا فايروسي ، ( 18 ) حاله ( 13,04 %) كحالات طبيه مختلفه (مثل febrile convulsion ، ningoencephalitis ، الصرع ،... الخ) و ( 42 ) حاله ( 30,43 %) كحالات غير مشخصه طبييا. عزلت بكتريا *Streptococcus pneumoniae* من ( 10 ) حالات فقط ( 18,18 %) من حالات التهاب السحايا البكتيري . كان متوسط اعمار المرضى لحالات التهاب السحايا البكتيري ( 130,75 ) شهر و كانت اعلى نسبه في الفئه العمريه ( 1-20 ) شهر والتي ظهرت بعدد ( 42 ) حاله ( 76,36 %). الحمى كانت من اكثر الاعراض تكرارا في ( 49 ) حاله ( 89,09 % ) متبوعه بالغثيان في ( 34 ) حاله ( 61,81 % ) ومن ثم الانفعاليه والصرع في ( 29 ، 25 ) حاله ( 52,72 ) و ( 45,45 % ) على التوالي. كان متوسط الكلوكوز والبروتين لحالات التهاب السحايا البكتيري ( 44 و 65 ) (ملغم/ديسليتر) على التوالي.