RESEARCH ARTICLE

FREQUENCY OF BACTERIAL, VIRAL AND FUNGAL MENINGITIS OCCURRENCE IN GADAP TOWN KARACHI, PAKISTAN

Khurram Jah Siddiqui*, Shazia Azhar, Syed Manzoor Iqbal Chishty, Syed Aley Hasan Zaidi
Department of Pathology and Molecular Medicine, Baqai Medical University, Karachi, Pakistan.

ABSTRACT
In the present study the frequency of bacterial (including tuberculous meningitis), viral and fungal meningitis occurrence in Gadap Town, Karachi, Pakistan has been determined. It is a population-based descriptive study which was carried out in Fatima Laboratory Hospital, Baqai Medical University, Karachi from November 2013 to November 2014. A total of 100 cases of meningitis were reported in the hospital during the study period. The physical, chemical and microscopic examinations of the samples were performed following the criteria of the Centers for Disease Control and Prevention (CDC). In biochemical analysis, glucose was estimated by hexokinase method whereas protein was estimated by calorie method of molybdate. The results of the analysis indicated that the incidence of bacterial meningitis was highest (65%) in the studied population, in which 30% cases were diagnosed with tuberculous meningitis and 35% with other bacterial infections. In the remaining cases, the fungal meningitis (20%) was second most common followed by viral meningitis (15%).

Keywords: CSF, meningitis, biochemical analysis.

1. INTRODUCTION
The term meningitis refers to the inflammation and swelling of the membranes that cover the brain and spinal cord1. Meningitis is caused by bacteria, fungi, viruses and parasites. Major sign and symptoms of meningitis include with cold hands and patients may feel agitated, some patients can become very sleepy and it may be difficult to wake them up. They may appear confused and unresponsive and may also develop a blotchy red rash that does not fade when you roll a glass over it. Other symptoms may also include severe, vomiting, high temperature, stiff neck, sensitivity to light and rapid breathing1.

There are four types of meningitis that includes bacterial, fungal, viral and parasitic2. Bacterial meningitis is very serious and should be treated as a medical emergency. It is most commonly caused by Streptococcus pneumoniae, Neisseria meningitidis and Haemophilus influenza. If the bacterial infection is left untreated, it can cause severe and infect the blood (septicemia). Bacterial meningitis is most common in children under five years of age particularly in babies under the age of one year3. Tuberculous meningitis is also a type of bacterial meningitis. It is a contagious airborne disease that typically affects the lungs. Tuberculosis (TB) is caused by a bacterium called Mycobacterium tuberculosis. If TB infection is not treated promptly, the bacteria can invade the bloodstream. It can infect other organs and tissues in the body4. A very complex and integrated series of events involving host cytokines, chemokines, photolytic enzymes, and oxidants appears to be responsible for meningitis-induced brain dysfunction5. This has resulted in the search for adjunctive therapies for bacterial meningitis, including corticosteroids6. The pathogenesis of bacterial meningitis involves colonization of the nasopharynx by the pathogens, microbial invasion into the intravascular space and survival within the bloodstream7. In addition, the microbial entry mechanisms into central nervous system (CNS), survival within the subarachnoid space, and host and/or environmental factors also contribute to the susceptibility of disease invasion. Such factors are reviewed extensively elsewhere8. M. tuberculosis bacilli initially cause a primary
infection, which commonly results in an acute illness. Acute illnesses with pneumonia, pleural effusion and marked mediastinal or hilar lymph node enlargement (may compress bronchi in children) have also been observed. Small pleural effusions are predominantly lymphocytic, typically contain few organisms, and clear within a few weeks. This sequence may be more common among young children and recently infected or re-infected immunosuppressed patients. Extra pulmonary TB at any site can sometimes manifest without evidence of lung involvement. About 95% of primary infections are asymptomatic and followed by a latent (dormant) phase. M. tuberculosis bacilli must be ingested by alveolar macrophages as a result of immune system activation. TB bacilli that are not killed by the macrophages actually replicate inside them, ultimately killing the host macrophage (with the help of CD8 lymphocytes); inflammatory cells are attracted to the area, causing a focal pneumonitis that coalesces into the characteristic tubercles that can be seen histologically. Infection is usually not transmissible in the primary stage and is never contagious in the latent stage. Foci of bacilli in the lung or other sites resolve into epithelioid cell granulomas, which may have caseous and necrotic centers. Infectious foci may leave fibro nodular scars in the apices of one or both lungs (Simon foci, which usually result from hematogenous seeding from another site of infection) or small areas of consolidation (Ghon foci). A Ghon focus with lymph node involvement is a Ghon complex, which if calcified is called a Ranke complex.

Meningitis can be difficult to diagnose because it often comes on suddenly and can be easily confused with as many of the symptoms are the same. Do not wait for the purple rash to appear because not everyone gets a rash. Where meningitis is suspected, treatment usually begins before the diagnosis has been confirmed. This is because some of the tests can take several hours to complete and it could be dangerous to delay treatment for that amount of time. On the contrary, viral meningitis is the most common and less serious type of meningitis. It is difficult to estimate the number of cases of viral meningitis because symptoms are often so mild that they are mistaken for flu.

For the investigation of various type of meningitis sample like cerebrospinal fluid (CSF) can be collected by lumbar puncture (spinal tap). The fluid will be sent to a laboratory for analysis to confirm the condition. It is apparent that the host immune response is incapable of controlling infection within the CNS.

2. MATERIALS AND METHODS
2.1. Study Design and Sample Collection
This study was carried out on the patients (aged 10-40 years) at Fatima Hospital Pathological Laboratory, Baqai Medical University from November 2013 to November 2014. A total of 100 samples were selected for this study. Signs and symptoms such as fever, headache and skin rash were also included as selection criteria of this study. Physical, chemical and microscopic examination of CSF was done according to the criteria of the Centers for Disease Control and Prevention (CDC). Samples were collected in vacuum tubes containing citrate and dextrose. The physical examination of the fluid samples was performed by observing the appearance, color and specific gravity.

2.2. Glucose and Protein Analysis in CSF
CSF glucose was estimated by Hexokinase method. This method is totally enzymatic utilizing both hexokinase and glucose-6-phosphate dehydrogenase enzymes. CSF protein was estimated by quantitative method in which pyrogallol red complexes with the protein in an acid environment containing molybdate ions. The resulting blue colored complex absorbs maximally at 600 nm and is directly proportional to the protein concentration of the sample.

2.3. Microscopic Examination
Microscopic examination of CSF was also performed. Red blood cells (RBC) and white blood cells (WBC) counts were done by Turk’s fluid in
Neubauer’s counting chamber. Identification of *M. tuberculosis* was made through acid-fast staining using Ziehl-Neelsen stain whereas Gram staining was carried out for the identification of other bacteria. Leishman’s stain was used for the staining of blood smears for the identification of type of WBC.

2.4. Statistical Analysis
All parameters were statistically analyzed using SPSS version 10. All values are expressed as the standard error of the mean (±SEM).

3. RESULTS AND DISCUSSION
Total 100 cases of meningitis were reported during November 2013 to November 2014. Out of those reported cases, the highest incidences of meningitis were found to be caused by bacteria (65%) followed by fungi (20%) and viruses (15%). In bacterial meningitis, 30% of the cases were found to be of tuberculous meningitis while the remaining 35% of other bacterial origin.

In physical examination of CSF, exudative CSF was found in case of bacterial meningitis whereas transude nature of CSF was observed in case of viral and fungal meningitis. The statistical analysis was applied on the biochemical parameters of CSF glucose and protein and is shown in Table 1. It was observed that CSF glucose was higher only in case of viral meningitis as compared to bacterial and fungal meningitis (Fig. 1). However, in case of CSF protein, it was observed that the protein level was low in viral meningitis as compared to others (Fig. 1). The microscopic examination of the samples revealed a high total leucocyte count (TLC) in bacterial meningitis as compared to fungal and viral meningitis (Table 2).

Meningitis is swelling of the membranes around the spinal cord and brain. It can be caused by microorganisms or by an injury. More often it is due to bacterial, fungal or viral infection. Bacterial meningitis is a serious illness and can be life threatening. It is most often caused by *Streptococcus pneumoniae, Neisseria meningitidis, and Haemophilus influenzae*, all of which are contagious.

The pathogenesis and pathophysiology of bacterial meningitis involves a complex interplay between virulence factors of the pathogens and the host immune response. Apart from these bacteria, *M. tuberculosis* also causes meningitis. CNS TB includes three clinical categories that include (1) Tuberculous meningitis, (2) Intracranial tuberculoma, and (3) Spinal tuberculous arachnoiditis. Children are especially vulnerable to bacterial meningitis.

Meningococcal bacteria cannot survive outside the body for long, so prolonged close contact with an infected person and contaminated food may increase the risk of transmission. Some of the individuals have meningitis causing bacteria in their throats or noses. Meningitis may cause fever, feeling of illness, headache, and skin rash. The chance of occurrence of a subependymal tubercle, with progression and rupture into the subarachnoid space, is the critical event in the development of tuberculous meningitis. Meningitis develops most commonly as a complication of post primary infection in infants and young children or in some cases from the chronic reactivation bacillemia in older adults with immune deficiency caused by aging, alcoholism, malnutrition, malignancy, human immunodeficiency virus (HIV) infection, or drugs (e.g., tumor necrosis factor [TNF]-alpha inhibitors).

Fungal meningitis is usually caused by Cryptococcus species and is not contagious. Parasitic meningitis is extremely rare but can be life threatening. It is caused by an amoeba called *Naegleria fowleri*. This parasite enters the body through the nose usually via contaminated lakes and rivers. Viral meningitis is the most common type but it is not usually life-threatening. The enteroviruses that cause meningitis can spread through direct contact with saliva, nasal mucus, or feces. They easily spread through coughing and sneezing. This is a concern in daycare centers, schools, and colleges. Arboviruses that also cause meningitis can be transmitted through insects like mosquitoes and ticks. Around 1,265 cases of meningitis were reported in England and Wales in 2009 and 2010.
Table 1. Statistical analysis of biochemical parameters of CSF.

<table>
<thead>
<tr>
<th>Biochemical analysis</th>
<th>Bacterial</th>
<th>Fungal</th>
<th>Viral</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Tuberculous</td>
<td>Other</td>
<td></td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>24.2 ± 8.92</td>
<td>24.2 ± 8.8</td>
<td>24.2 ± 8.2</td>
</tr>
<tr>
<td>Protein (mg/dl)</td>
<td>256.8 ± 134.2</td>
<td>256.8 ± 134.2</td>
<td>255.2 ± 138.1</td>
</tr>
</tbody>
</table>

Table 2. Total leucocyte count (TLC) in different types of meningitis.

<table>
<thead>
<tr>
<th>Meningitis</th>
<th>TLC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacterial</td>
<td></td>
</tr>
<tr>
<td>Tuberculous</td>
<td>20-25 / HPF (lymphocytes)</td>
</tr>
<tr>
<td>Other</td>
<td>25-30 / HPF (neutrophils)</td>
</tr>
<tr>
<td>Fungal</td>
<td>10-15 / HPF (neutrophils + lymphocytes)</td>
</tr>
<tr>
<td>Viral</td>
<td>10-15 / HPF (neutrophils + lymphocytes)</td>
</tr>
</tbody>
</table>

Fig. 1. Glucose and protein levels of CSF in meningitis samples.
4. CONCLUSION
Incidences of bacterial meningitis were higher in the studied population as compared to fungal and viral meningitis. CSF glucose was found higher only in viral meningitis as compared with the bacterial and fungal meningitis. CSF protein was low in viral meningitis as compared to bacterial and fungal meningitis.

REFERENCES


