Cytological and Biochemical Profile of Cerebrospinal Fluid from Meningitis Patients

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BACKGROUND: The term Meningitis is used to describe an inflammatory infection of the membranes surrounding the brain and spinal cord, which occurs as either a primary disease or secondarily to disease in some other part of the body. The diagnosis is primarily confirmed by analyzing cerebrospinal fluid (CSF). Early diagnosis of the cause may be based on the cytological and biochemical parameters. Our objective was to determine the cytological and biochemical profile of CSF from meningitis patients.

METHODS: In this cross sectional study, a total of 356 CSF specimens were collected from patients suspected of meningitis and processed microscopically and microbiologically by standard microbiological methods in Emergency Lab of Tribhuvan University Teaching Hospital (TUTH) Kathmandu, Nepal, over a period of six months, from March to August 2014 to determine cytological and biochemical parameters.

RESULTS: Out of the 13 confirmed bacterial meningitis cases from 356 processed CSF samples, the mean value of total leukocytes count (TLC) was found to be 337.3 cells/mm^3 with predominant neutrophils (73.8%). The mean value of glucose and protein was 28.8 mg/dL and 89.4 mg/dL respectively in the cases of bacterial meningitis. Among the three confirmed cases of fungal meningitis, the mean value of TLC was 11.7 cells/mm^3 with lymphocytic predominance. In fungal meningitis glucose level was found to be normal (45.0 mg/dL) with slight increase in protein (48.7 mg/dL).

CONCLUSIONS: Bacterial meningitis is generally characterized by increased TLC with predominance of neutrophils, decreased glucose and increased protein levels whereas fungal meningitis displays decreased TLC predominance of lymphocytes, normal or reduced glucose and slight increased protein level.

Key words: Bacterial meningitis, biochemical and cytological parameters, fungal meningitis.

Introduction

The CSF is a clear bodily fluid that occupies the space between the arachnoid mater (meninges) and the pia mater. It is formed in the choroid plexus by both filtration and active transport. It protects the brain from the sudden change in pressure, it maintains stable chemical environment and removes wastes products of cerebral metabolism [1].

CSF evaluation is the single most important aspect of the laboratory diagnosis of meningitis. Analysis of the CSF abnormalities produced by bacterial, mycobacterial, and fungal infections may greatly facilitate diagnosis and direct initial therapy. Basic studies of CSF that should be performed in all patients with meningitis include measurement of pressure, cell count and white cell differential; determination of glucose and protein levels; Gram's stain; and culture [2].

The inflammation by various pathogens induces anatomical and physiological changes in the meninges which are responsible for characteristic changes in the values of CSF from patients with meningitis. The loss of integrity of cerebral capillaries and thus, the loss of integrity of the blood-brain barrier results in leakage of protein into the CSF and increased migration of Polymorphonuclear (PMN) leukocytes into the CSF [3].

Normal CSF contains 0-5 lecukocytes/mm^3, mainly lymphocytes, though in neonates cell count is up to 30/mm^3 [4]. WBC count of >500/mm^{3}with a preponderance of neutrophils is characteristic of a bacterial meningitis, and a WBC count of >100/mm^{3}with a preponderance of monocytes is characteristic of a viral meningitis a considerable pattern overlap is often found [5]. CSF glucose levels are used to distinguish bacterial meningitis (where it is usually decreased, usually <40 mg/dl) from aseptic meningitis (where the glucose levels are usually unaltered) [5]. Decreased CSF glucose results from changes in the physiological functioning of the choroid

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epithelium as well as from consumption by bacterial pathogens and leukocytes [6]. Chemically meningitis can be differentiated from bacterial meningitis by CSF glucose levels (<10 mg/dL) and CSF WBC values (>7500 cells/mm^3) [7].

Proteins are largely excluded from the CSF by the blood - CSF barrier. Protein gaining access to the CSF primarily reaches the CSF by transport within pinocytotic vesicles traversing capillary endothelial cells [1]. Protein level greater than 200 mg/dL, is highly significant for bacterial meningitis indicating disruption of the blood-brain or the blood-CSF barrier [8].

This study aims to look at the changes in the cytological and biochemical value of CSF for the diagnosis of meningitis.

Methods

Across sectional study was carried out at the Emergency laboratory in Tribhuvan University Teaching Hospital (TUTH) from March to August 2014. Total 356 CSF samples were collected from patients clinically suspected of meningitis. The samples were processed microscopically and microbiologically to determine cytological and biochemical and microbiological parameters for diagnosing meningitis. The TLC were counted by Neubauer counting chamber method and differential leukocytes count (DLC) was by Wright's staining. The level of protein and glucose was determined by using Biochemistry Automatic Analyzer (Erba XL-200).

The specimens were cultured on Chocolate agar (CA), Blood agar (BA), MacConkey agar (MA), Mannitol salt agar (MSA), Nutrient agar (NA) and Sabouraud Dextrose agar (SDA). For bacterial isolates, BA and CA plates were incubated in candle jar (5-10% CO_2) at 37°C for overnight. MA plates were incubated at 37°C in incubator for overnight. SDA plates were used for culture of fungal isolates and incubated at 37°C for 2-3 days.

SPSS version 20 was used to analyze quantitative data.

Results

Among the total 356 processed CSF samples, there was a slight male dominance in the sex ratio (1.3:1) with males contributing 56.5% and females 43.5% (Table 1). The mean age of the patients was 27.8 years.

Among the total processed specimen (N=356), only 16(4.5%) cases were known to have laboratory confirmed cases of meningitis with the help of CSF culture results.

CYTOLOGICAL PROFILE

The maximum number (N=10, 13.5%) of isolates were isolated from the CSF samples having TLC >100 cells/mm^3 (Table 2).

And no isolate was from samples having normal cell count range (Table 2).

The mean value of TLC was found to be more (337.3 cells/mm^3) in bacterial meningitis compared to fungal meningitis (11.7 cells/mm^3). Neutrophils were predominant in bacterial meningitis whereas lymphocytes were predominant in fungal meningitis (Table 3).

BIOCHEMICAL PROFILE

The isolation rate of pathogens was highest (13 out of 72, 18.1%) in the CSF samples having glucose level <40 mg/dL. Similarly highest number of pathogens (13 out of 150, 8.7%) were isolated from the CSF samples having protein level >45 mg/dL.

Table 1. Age and Sexwise Distribution of Suspected Cases of Meningitis

Age Groups (Years)	Sex				Total	
	Male		Female			
	N	%	N	%	N	%
<1	21	5.9	16	4.5	37	10.4
1 - 14	56	15.7	45	12.6	101	28.4
14 - 30	47	13.2	37	10.4	84	23.6
30 - 60	49	13.8	42	11.8	91	25.6
> 60	28	7.9	15	4.2	43	12.1
Total	201	56.5	155	43.5	356	100

Table 2. TLC of Total CSF Specimen and Culture Positive Isolates

TLC (cells/mm^3)	Total CSF Samples (N=356)		Culture Positive Isolates (N=16)	
	Frequency (N)	Percentage (%)	Frequency (N)	Isolation Rate (%)
(Normal) 0 - 5	153	43.0	0	0
5 - 100	129	36.2	6	4.7
> 100	74	20.8	10	13.5

Table 3. Cytological Parameters in Different Types of Meningitis

Types of Meningitis/Cytological Parameters	Range	Mean	Standard Deviation
Bacterial Meningitis (N=13)			
TLC (cells/mm^3)	10 - 2000	337.3	523.8
DLC (%)			
Lymphocytes	5 - 46	26.2	12.9
Neutrophils	54 - 95	73.8	12.9
Fungal Meningitis (N=3)			
TLC (cells/mm^3)	10 - 15	11.7	2.9
DLC (%)			
Lymphocytes	100	100	0.0
Neutrophils	0	0	0.0

Table 4. Biochemical Parameters in Different Types of Meningitis

Types of Meningitis/ Biochemical Parameters	Range	Mean	Standard Deviation
Bacterial Meningitis (N=13)			
Glucose (mg/dL)	1.8-68.4	28.8	20.0
Protein (mg/dL)	46.9 - 175.6	89.4	42.6
Fungal Meningitis (N=3)			
Glucose (mg/dL)	30.6 - 72	45.0	23.4
Protein (mg/dL)	39.8 - 57.3	48.7	8.8

The mean value of glucose level (28.8 mg/dL) was found to be reduced than normal range and the protein level (89.4 mg/dL) was found to be above normal in bacterial meningitis cases whereas among the fungal meningitis, the mean value of glucose level (45 mg/dL) was found to be in normal range and protein level (48.7mg/dL) was slightly increased than the normal level (Table 4).

Discussion

Out of 356 CSF samples processed, only 16(4.5%) samples showed growth on CSF culture. This finding of culture positive result is in agreement with several similar studied conducted in Nepal. A study conducted in Manipal Teaching Hospital Nepal from 2000 to 2005 demonstrated 4.58% growth on CSF culture [9]. A similar study conducted from 2004 to 2008 showed 4.4% growth on CSF culture [10].

Among the 13 bacterial meningitis cases confirmed by culture results, it was observed that TLC was greater than normal range and found in the range of 10-2000 cells/mm^3 with predominant neutrophils (73.8%) in all cases, protein level were greater than normal value (89.4 mg/dL) and glucose contents were lower than normal range (28.8 mg/dL). Markedly decreased CSF glucose with markedly increased total protein, high WBC count with 89% Neutrophils, and the presence of a large number of PMN leukocytes and bacteria in the Gram-stained smear of the CSF sediment are the most striking laboratory results in bacterial meningitis [11].

Similarly, a study also mentioned that the examination of the CSF of a patient with acute bacterial meningitis characteristically reveals a cloudy fluid, consisting of an increased white blood cell count and predominance of PMN leukocytes, a low glucose concentration in relation to serum value, a raised concentration of

protein, and positive Gram stained smear and culture for the causative microorganism [12]. During the bacterial infection, due to microbial physiology, the protein is released and thus the level of protein is increased in CSF. The change in protein level than normal can be used to get the idea to distinguish viral from bacterial meningitis, as in bacterial infection, the protein level is usually raised in case of viral infection, the level of protein remains almost normal. Thus this finding in this study goes well with the established medical knowledge.

Among the fungal meningitis, mean value of protein level was slightly increased and glucose was within normal range. These findings are in accordance with other researcher's [13] finding which also displayed typical changes with elevated lymphocytes, elevated proteins (0.82 g/L), and decreased glucose levels (1.3 mmol/L).

Conclusion

Bacterial meningitis is generally characterized by increased TLC with predominance of neutrophils, decreased glucose and increased protein levels whereas fungal meningitis displays decreased TLC predominance of lymphocytes, normal or reduced glucose and slight increased protein level.

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