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BACTERIAL MENINGITIS: A DIAGNOSTIC APPROACH

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Over a thirteen months period, 456 patients clinically suspected of having bacterial meningitis were investigated. Cerebrospinal fluid (CSF) specimens were examined by Gram's stain, white cell (WBC) counts and culture techniques. Gram's stain and cultures of CSF (232) with a cell count of less than 10 cells/mm³ were consistently negative. However, of the CSF specimens (88) with cell counts greater than 100/mm³, 28 (6.7%) and 23 (5.2%) of the specimens were positive by cultures and Gram's stain respectively. A total of 31 isolates were identified by culture. Streptococcus pneumoniae (22.4%), Escherichia coli (16%), and Coagulase negative Staphylococci (16%) were the predominant organisms, followed by Haemophilus influenzae (6.4%) and Neisseria meningitidis (2.6%). Thus it was concluded that Gram's stain and perhaps culture of CSF with cell counts of less than 10/mm³ are of no practical diagnostic significance and may be eliminated as routine procedures. The gold standard for the diagnosis of bacterial meningitis is raised WBC cell count (>100/mm³), positive Gram's smear and/ or culture of CSF specimen.

Bacterial meningitis remains a potentially life threatening emergency with substantial morbidity and mortality world wide[1,2]. Historically newborns, infants and young children followed by elderly had the highest risk of developing bacterial meningitis. However, this profile has shifted dramatically with the widespread use of Haemopilus influenzae-b vaccine (Hib). Currently the average age of contracting meningitis is above 25 years with Streptococcus pneumoniae, Neisseria meningitidis and Haemopilus influenzae being the most common pathogens[3,4]. Despite advances in vaccine development and chemoprophylaxis, bacterial meningitis remains a major cause of death and neurological disabilities which can be prevented by early diagnosis.[5-7]

As meningitis is a serious emergency, rapid examination of cerebrospinal fluid (CSF) is considered an essential and critical step in early diagnosis and management of the patients.[4,7,8] Gram's staining, white cell count, measurement of glucose and proteins levels and CSF culture are the traditional laboratory tests used to support the clinical findings of meningeal infections[9]. Gram's stain is the most rapid, least expensive and simplest method for the presumptive diagnosis of bacterial meningitis.[9,10] However, Gram's stain is also considered the least sensitive method probably reflecting deficiencies in its technique and is also prone to errors.[11,8,12]

The present study was undertaken to evaluate the significance of Gram's stain of CSF as an aid in the rapid diagnosis of bacterial meningitis and to find out the aetiological agents in these cases of bacterial meningitis.

MATERIALS AND METHODS

Four hundred and forty six patients clinically suspected of having bacterial meningitis during the period April 2005 to April 2006, were included in this study. They were analyzed on the basis of Gram's stain, total and differential white cell count and culture of CSF. The white cell count was performed manually using a haemocytometer. The CSF was centrifuged at 2000 rpm for 10 minutes; the sediment was Gram's stained and cultured on blood agar, Chocolate agar and MacConkey's agar using standard techniques. All the isolates were identified on the basis of their colonial morphology, cultural characteristics and biochemical reactions. Gram's staining was performed using a clean glass slide previously soaked in 70% ethanol and the stained smears were carefully examined for the presence of bacteria. No attempt was made to analyze the findings on the basis of age or sex of the patients or any other factors.

RESULTS

On the basis of total white cell count (range 0 to 21000/mm³), CSF was divided into three groups as shown in Table 1. Group I included CSF samples with cell counts of 0-10/mm³, Group II samples with cell counts of 11-100/mm³ and Group III comprised of CSF samples having a cell count of more than 100/mm³.

Table 1 shows the correlation between Gram's stain, cultures & CSF cell count. Group I included 232 CSF samples, all of which were negative on Gram's stain and culture. Out of 136 CSF samples in Group II, only 1 sample was positive by Gram's stain while 3 samples were positive on culture. In Group III, a total of 28 CSF samples (with predominant polymorphonuclear cells), were positive on culture, of which 23 also yielded a positive Gram's smear.

Table 1: *Correlation of Gram's stain's, CSF culture and cell count.*

Cell count	Group I 0-10/mm³	Group II 11-100/mm³	Group III >100/mm³	Total
No. of specimens	232	136	88	456
Positive cultures	0	3 (2.2%)	28 (31.9%)	31 (6.7%)
Positive Gram's stain	0	1 (0.7)	23 (26%)	24 (5.2%)

Table 2: *Organisms isolated from CSF.*

Organisms isolated	No.	%
S. pneumoniae	7	22.4
E-coli	5	16
B haemolytic streptococci	4	12.8
Staph. aureus	3	9.6
Pseudomonas spp	2	6.4
Klebsiella spp	2	6.4
H. influenzae	2	6.4
N.meningitidis	1	3.2
Coagulase negative Staph.	5	16

S. pneumonia was the predominant agent (22.4%) followed by E-coli (16%), Coagulase negative Staphylococci (16%), B haemolytic streptococci (12.8%) and H. influenzae (6.4%). However, N. meningitidis (3.2%) was isolated from only one sample (Table 2). Cultures identified more etiological agents (31) than Gram's stained smears (24) from the clinically suspected cases of bacterial meningitis. This result (28) correlated mostly with cell counts of >100 cells/mm³ of CSF (Group III).

DISCUSSION

Laboratory investigations of CSF specimens in suspected cases of bacterial meningitis are extremely important for prompt diagnosis and management of patients. Several studies have reported Gram's staining as the most useful single test for identifying bacterial meningitis, as it revealed more positive cases than cultures.[7,10,13,14] However, in our study Gram's staining revealed the probable aetiological agent in 24 cases while 31 were positive on culture. Other workers have also reported low positivity on Gram's staining as compared to cultures.[3,11]

Very little information regarding the correlation between Gram's stain and the cellular response is available in the literature. However, relationship between normocellular CSF and cultures have been reported. In one study, 5% of the neonates with bacterial meningitis had either 0 or 1 CSF WBCs/mm³, while < 3 CSF WBCs/mm³ was seen in 10% of the neonates[1]. In another study positive cultures were reported with < than 5 WBCs/mm³ CSF count[15]. In our study, 28 cases of bacterial meningitis with positive cultures had >100 CSF WBCs/mm³ while the normo cellular CSF samples were all negative. Similar results have been reported in other studies.[8,5,1]

The data from the present study indicates that a positive Gram's stain on CSF may be strongly suggestive of bacterial meningitis but this finding is generally accompanied by an elevated cell count in the CSF. Hence, it can be concluded that Gram's stain is of little practical value in the routine evaluation of the normo-cellular CSF, but its value as a presumptive diagnostic aid in elevated CSF count can not be ignored.

The incidence of specific pathogens causing bacterial meningitis varies around the world.[3,6,7] In our study the predominant organism isolated was S. pneumoniae (22.4%) followed by E- coli (16%) and coagulase negative Staphylococci (16%). H. influenzae caused 6.2% of the total number of pyogenic meningitis, while N. meningitidis was isolated from one case only. Similar findings have been reported from other parts of the world[3,6,16], although many have reported H. influenzae and N. meningitidis as the more common pathogen.[2,17,7] This is probably due to the endemicity of the organism and to the larger number of cases included in their studies. Thus, for the diagnosis of bacterial meningitis a raised CSF cells/mm³, positive Gram's stain and / or positive CSF culture for bacterial pathogens can be considered the gold standard.

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