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Cerebrospinal fluid lysozyme in the diagnosis of tuberculous meningitis

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Pre-treatment lysozyme levels in cerebrospinal fluid (CSF) were estimated in 37 patients with tuberculous meningitis (TBM), 16 with non-tuberculous meningitis (non-TBM) and 13 with non-inflammatory conditions of the central nervous system (controls) in an attempt to assess the role of CSF lysozyme in the diagnosis of TBM. Lysozyme content in the CSF was found to be elevated in all patients with bacteriologically confirmed TBM and in a large proportion of patients in whom the disease was clinically diagnosed but bacteriologically not confirmed. The enzyme was not detected in all but one of the control subjects and in most (69%) of the non-TBM patients.

Tuberculous meningitis (TBM) is a serious form of extra-pulmonary tuberculosis associated with high mortality and morbidity in children and adults. The diagnosis is mainly based on clinical and biochemical parameters since bacteriological confirmation is not always possible. Hence, there is a need to develop tests with high degree of specificity and sensitivity to aid in the diagnosis of tuberculous meningitis in its early stages.

Serum lysozyme has been found to be elevated in, conditions characterised by epitheloid cell granulomas, especially sacroidosis¹, tuberculosis² and leprosy³. In the present study, an attempt has been made to elucidate the role of cerebrospinal fluid (CSF) lysozyme levels in the diagnosis of TBM by comparing the enzyme levels in patients clinically diagnosed as tuberculous meningitis (with and

without bacteriological evidence) with those in patients of non-tuberculous meningitis (non-TBM) and control subjects with non-inflammatory condition of the nervous system.

Material & Methods

The lysoplate assay method as described by Osserman and Lawlor⁴ was followed to estimate the lysozyme levels. The zone of lysis was found to be linearly related to the log concentration of lysozyme standard. Batch variation (lysoplates prepared in different batches) and day-to-day variation were studied and the overall coefficient of variation of the estimate with 16 observations was less than 5 per cent.

The diagnosis of TBM was based on clinical criteria and biochemical characteristics and cellular profile of CSF. The

patients in the control group had no clinical evidence of meningitis and were admitted to the hospital for investigation of non-inflammatory conditions of the central nervous system. The CSF samples were obtained before the start of treatment from children below 15 yr of age with IBM and non-TBM. All the specimens were subjected to bacteriological examination for evidence of tuberculosis.

A total of 66 CSF samples (37 TBM, 16 non-TBM, 13 controls) were studied. All the CSF samples were stored at -20°C till they were assayed. The CSF samples and standards were coded and randomly allocated to the lysoplate wells.

Results & Discussion

The TBM patients were divided into culture-positive and culture-negative group based on CSF culture positivity for *Mycobacterium tuberculosis*. The results of lysozyme estimation in TBM and non-TBM patients and controls are shown in the Fig. Lysozyme could be detected in all the 11 patients of TBM with CSF cultures positive for *M. tuberculosis*. Of the 11, 9 showed lysozyme levels >2.7 and the values for the remaining two were 2.1 and 1.8. Among the 26 TBM patients with CSF cultures negative for *M. tuberculosis*, only 4 did not have detectable amounts of lysozyme in CSF. Of the remaining 22 samples, 9 exhibited values < 2 , 5 showed values between 2 and 2.99 and 8 had values ≥ 3 . The mean lysozyme concentration was found to be higher in culture-positive than in culture-negative TBM group. In the non-TBM group only two specimens had values > 1.8 out of 5 positive for lysozyme. Among the controls, the single

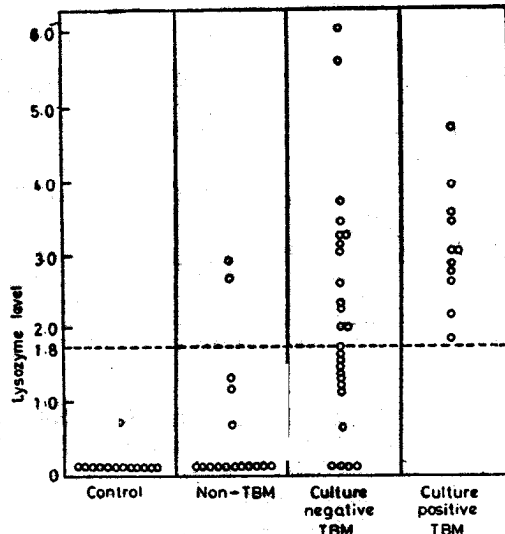


Fig. Lysozyme levels [\log_e , $\mu\text{g/ml}$]-1 in CSF in the different groups

Group	No.	Mean \pm SD
Control	13	0.05 \pm 0.19
Non-TBM	16	0.56 \pm 0.99
Culture -ve TBM	26	2.05 \pm 1.59
Culture +ve TBM	11	3.07 \pm 0.78

positive result had a value of 0.7. The mean lysozyme levels in culture-positive and culture-negative TBM groups were significantly higher than in the non-TBM and control groups ($P < 0.01$).

From this it is evident that the lysozyme levels were found to be elevated in all the bacteriologically confirmed patients (culture-positive) of TBM, the lowest estimate among this group being 1.8. Among the culture-negative TBM group, approximately half the samples showed values of 1.8 or more. The enzyme was not detected in most of the non-TBM patients and in 12 out of 13 control subjects. In view of these findings, it seems possible that a lysozyme concentration

of 1.8 or above in CSF specimens may prove as valuable aid in the diagnosis of TBM. By applying this criterion only 13 out of 26 patients could be confirmed as TBM among culture negative TBM group. However, from the observations, if a level of 1.0 is considered as the diagnostic criterion, 21 patients from culture-negative TBM group would have been classified as definite TBM cases thereby increasing the sensitivity of the assay. Though two patients from the non-TBM group would be classified as TBM the specificity of the assay is not much altered.

The preliminary observations are encouraging and it may be possible to arrive at a definite criterion (lysozyme level) for diagnosis of TBM by assaying a sufficiently large number of TBM patients with and without bacteriological confirmation.

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