Meningitis-associated low molecular mass proteins in cerebrospinal fluid

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Recent investigations show that a great deal of effort is being put to develop a specific test which could be effectively used to diagnose meningitis and which could also be helpful in differential diagnosis of infectious diseases of the central nervous system [1-3]. In this context several constituent proteins of cerebrospinal fluid (CSF) have been evaluated to establish their possible correlation with bacterial meningitis [1, 4, 5]. We previously evaluated the use of ultra-thin-layer SDS/PAGE as a rapid method to analyse CSF proteins, as qualitative changes occurring in these might be of some value in the diagnosis of bacterial meningitis [6]. In this communication we report the presence of two low molecular mass proteins which appear to be specific for bacterial meningitis.

CSF samples from 112 patients with bacterial meningitis were examined by ultra-thin-layer SDS/PAGE using the procedure described elsewhere [6]. The total protein concentration in these CSF samples was between 21 and 3500 mg/dl (mean 232 mg/dl). This indicates that total protein concentration in this disease is inconsistently raised, thus making it a highly unreliable parameter for diagnostic purposes. When these CSF samples were analysed by SDS/PAGE (Fig. 1a) they showed a consistent pattern of protein bands despite the fact that their total protein concentrations were not comparable in many cases. In addition to the protein bands present at M, 66000, 55000 and 25000 which seemed to be the common feature of CSF samples, the most noticeable finding was the presence of two low molecular mass protein species at approximate M, 12000 and 15000. In 91 out of 112 samples of bacterial meningitis these protein bands were clearly visible. The same combination of these two bands was not detected in the CSF sample of viral meningitis and other disorders of the nervous system.

To further investigate whether these proteins exist as monomeric proteins or are subunits of each other, the same samples were also subjected to PAGE in the absence of detergent and reducing agent. Fig. 1(b) shows the electrophoretic profiles of CSF proteins from individuals with bacterial meningitis. On this system, those low molecular mass protein bands were not present. Instead, a single band at approximate M, 30000 was present which could possibly be a dimer of the two low molecular mass proteins described earlier on SDS-gels. Attempts are now being made to establish the physicochemical characteristics of these proteins.

Our results show that the specificity of these low molecular mass proteins was 81%, which could possibly place these proteins which appear to be specific for bacterial meningitis.

Abbreviation used: CSF, cerebrospinal fluid.

Isolation of low molecular mass proteins from renal stones

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Abbreviation used: THM, Tamm-Horsfall mucoproteins.

Incidentes of renal stone formation are fairly common in areas of hot climatic conditions like the Asian subcontinent and the Middle East. Current theories suggest that some inhibitors and promoters of stone formation present in urine may play a key role in the prevention and formation of stones [1, 2]. A disturbance in the ratio between promoters and inhibitors might therefore be the cause of stone formation in...
In order to investigate the possible involvement of proteins in stone formation we decided to analyse renal stones for the presence of such proteins which might be actively participating in this process. Identification of such proteins as essential constituents of renal stones would have an important bearing in determining a more specific promoter of renal stone formation. For this study we selected ten renal stones of various sizes and five gall stones. The outer surface of each stone was washed once with 0.1 M HNO₃ and once with 2% (w/v) SDS. These washings were carried out to make all stones free of any contaminating mucus. After breaking the stone into two halves, the nidus was removed and crushed into fine powder. For the extraction of proteins, powdered nidus was mixed with a ratio of 1:4 (w/v) in 0.065 M-Tris–HCl buffer containing 2% (w/v) SDS, 5% (v/v) β-mercaptoethanol and 10% (v/v) glycerol. This mixture was then heated to 100°C for 10 min. After centrifugation, supernatant was removed and analysed by thin-layer SDS/PAGE [7]. Fig. 1 shows the electrophoretic profiles of proteins extracted from renal stones. Renal stone proteins extracted by this procedure were shown to be composed of three protein species as detected by SDS/PAGE. It appears that these proteins exist as very tightly attached components to other constituents of stone matrix, since these were totally unextractable by any other method using non-denaturing conditions. When β-mercaptoethanol and SDS were excluded from the extraction buffer no protein band was detected on SDS/polyacrylamide gels. These proteins also seemed to be specifically associated with renal stones as gall stone sample extracts did not show the presence of such proteins when analysed by the same procedure.

The presence of these low molecular mass proteins in the core of renal stones and their firm attachment with other matrix substances strongly suggests that these proteins might have some influence on stone formation. Further studies are underway to fully characterize these proteins and to carry out quantitative measurements of these in urine and serum of stone formers.

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Evaluation of creatine kinase in the cerebrospinal fluid of patients with various neurological diseases

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Abbreviations used: CK, creatine kinase; CSF, cerebrospinal fluid.

The evaluation of creatine kinase (CK) in the cerebrospinal fluid (CSF) is of potential interest for determining the extent of injury in cerebrovascular accident and hypoxic–ischaemic brain damage [1, 2]. While assessing the qualitative or quantitative changes in CSF most investigators have used techniques like immunoassays and/or agarose-gel electrophoresis. Although CK-BB is the predominant species present in CSF, other isozymes and subforms of iso-

Fig. 1. SDS/PAGE of renal stone proteins

Samples were isolated and prepared according to conditions described in the text.


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