CORRELATION BETWEEN THE CONCENTRATION OF $\beta$-TRACE PROTEIN AND THE NUMBER OF SPERMATOZOA IN HUMAN SEMEN

J. E. OLSSON

Departments of Neurology and Forensic Medicine, University Hospital, S-221 85 Lund, Sweden

(Received 17th June 1974)

Beta-trace protein ($\beta$-TP) is predominantly associated with the central nervous system (CNS) and was first discovered in concentrated cerebrospinal fluid (CSF) by Clausen (1961). The protein has a low molecular weight of 31,000 and is present in high concentration in the CSF (0.8 to 3.9 mg/100 ml), constituting about 7% of the total CSF protein concentration (Link, 1967). By contrast, the serum concentration of the protein is low (0.26 to 0.60 mg/100 ml) and $\beta$-TP constitutes only 1/20,000 of the total serum protein content (Olsson, Link & Nosslin, 1973). The protein is also found in urine (Hochwald & Thorbecke, 1962), in which the daily excretion is below 9.5 mg (Ericsson, Link & Zettervall, 1969), as well as in various organ extracts, e.g. brain, kidney, pancreas and genital organs (Laterre, Heremans & Carbonara, 1964; Penny & Osserman, 1971). The highest amounts of $\beta$-TP are found in white matter (Olsson & Link, 1973) and in the epididymis (Olsson & Nord, 1973). In the former, $\beta$-TP is mostly found in the glial cells and in brain tumours of glial cell origin (Olsson, Blomstrand & Haglid, 1974).

An increased concentration of $\beta$-TP is found in the CSF after strokes involving brain damage (Link & Olsson, 1972) and in severely disabled patients with multiple sclerosis (J. E. Olsson, H. Link and R. Müller, unpublished observations), and its excretion in urine is increased in patients with brain tumours, cerebrovascular diseases and renal failure (Ericsson et al., 1969). Recently, another clinical application of $\beta$-TP, as a marker of fetal neural-tube defects in samples of amniotic fluid, has been reported (Macri, Weiss, Joshi & Evans, 1974).

Interest in the curious anatomical distribution of $\beta$-TP, especially in the CNS and the genital organs and its high concentration in the CSF, initiated the present study of the protein in normal and pathological human seminal fluid.

Samples of semen were obtained from 309 men, who were examined because of barren marriages or as a link in a paternity examination. The numbers of spermatozoa were counted and the patients were allocated to four groups according to the number of cells. In addition, the motility and the morphology of the cells were examined using a light microscope. The different groups of patients are shown in Table 1. In all samples, the concentration of $\beta$-TP was determined by single radial immunodiffusion on agar as described by Mancini,
Carbonara & Heremans (1965) and the total protein content was assessed by the method described by Lowry, Rosebrough, Farr & Randall (1951). Semen samples were obtained from eight men whose ejaculates showed 'normal' sperm morphology, motility and concentration. The samples were centrifuged three times at 9000 g for 15 min on each occasion. The resulting supernatant was free from spermatozoa when examined using a light microscope. The total protein content and the concentration of β-TP were determined in the original sample, the pellet and the supernatant. A specific antiserum against β-TP was prepared by repeated immunization of rabbits with pure β-TP obtained from human CSF (Link, 1967). The

antiserum was absorbed with human serum. An immunological identity between human CSF and semen was found when testing these fluids against antiserum against human β-TP by the double diffusion technique described by Ouchterlony (1948).

The concentration of β-TP and the total protein concentration in semen samples containing different numbers of spermatozoa are given in Table 1. Comparisons between the mean numbers were performed with Student's t test. A strong correlation was found between the concentration of β-TP in human seminal fluid and the number of sperm cells. The motility and the morphology of the cells, however, did not appear to correlate with the content of β-TP.

The concentration of β-TP decreased when the number of spermatozoa decreased and the values differed significantly (P<0·001) between all groups. The total protein content was constant in all patients who had spermatozoa in the semen, but was somewhat lower (P<0·05) in the azoospermic patients. In the centrifuged samples of 'normal' semen, the concentration of β-TP and the total protein content remained fairly constant in the original sample and in the supernatant of the centrifuged sample, whereas the concentration of β-TP was about 5 times lower and the total protein content about 2·5 times lower in the pellet.

Table 1. Number of spermatozoa, concentration of β-trace protein and total protein content in human semen in different groups of patients

<table>
<thead>
<tr>
<th>No. of sperm. (× 10⁶/ml)</th>
<th>β-trace protein (mg/100 ml)</th>
<th>Total protein (g/100 ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>40 to 250 (N = 140)</td>
<td>4·9±0·296 (0·6 to 18·0)</td>
<td>5·1±0·196 (3·2 to 9·9)</td>
</tr>
<tr>
<td>10 to 40 (N = 89)</td>
<td>3·3±0·354 (0·3 to 21·6)</td>
<td>5·0±0·110 (2·5 to 7·8)</td>
</tr>
<tr>
<td>≤ 10 (N = 61)</td>
<td>1·7±0·209 (0·0 to 8·9)</td>
<td>5·1±0·160 (1·8 to 7·6)</td>
</tr>
<tr>
<td>Azoospermia (N = 19)</td>
<td>0·8±0·175 (0·0 to 3·3)</td>
<td>4·4±0·232 (3·3 to 6·6)</td>
</tr>
</tbody>
</table>

Values are given with mean ± S.E.M. and range in parentheses. N = number of patients.
Beta-trace protein in human semen

In some of the samples, the content of fructose was determined. There was no significant correlation between the concentrations of fructose and those of β-TP.

The present findings of a mean concentration of about 5 mg β-TP/100 ml in normal human semen indicate that the protein constitutes about 1/1000 of the total protein concentration in semen. Thus, the relative concentration of β-TP in human semen is lower than that in CSF but higher than that in serum. In patients with azoospermia, the mean relative concentration of β-TP in semen is only about 2/10,000 of the total protein content.

The connection between β-TP and the number of spermatozoa in human semen will be further investigated.

REFERENCES


