NOTE

Luteinizing Hormone-Releasing Hormone in Cerebrospinal Fluid of Women

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Synopsis

The concentration of luteinizing hormone-releasing hormone (LH-RH) in both cerebrospinal fluid (CSF) and plasma in 36 women with various conditions were determined, and the correlation of LH-RH concentrations between plasma and CSF was studied. Paired samples of plasma and CSF were obtained just before operation and were extracted with methanol for LH-RH radioimmunoassay. LH and FSH were also measured by radioimmunoassay. The mean plasma LH-RH levels were 3.79 pg/ml in the follicular phase, 4.15 pg/ml in the luteal phase, 12.73 pg/ml in post-menopausal women and 2.31 pg/ml in pregnant women at term. No immunoreactive LH-RH in the CSF was found in 29 out of 36 subjects, and the remaining 7 subjects showed very low levels (less than 0.95 pg/ml). The LH-RH levels in the CSF of post-menopausal women were not higher than those in the remaining 3 groups, although their plasma levels of LH-RH were significantly ($p < 0.001$) elevated. The mean plasma levels of LH and FSH in post-menopausal women were significantly higher than those in normal cyclic women and in pregnant women. The present results suggest that little or no LH-RH is present in the CSF, and it is not clear that there is any correlation between the LH-RH level of plasma and those of CSF.

Several investigators (Allen *et al.*, 1974; Assies *et al.*, 1978; Jordan *et al.*, 1976) have demonstrated the presence of pituitary hormones, *i.e.*, luteinizing hormone (LH), follicle stimulating hormone (FSH), adrenocorticotrophic hormone, growth hormone and prolactin in human cerebrospinal fluid (CSF). It was reported that CSF in 26 human adults (Gunn *et al.*, 1974) and from 5 human subjects (Mortimer *et al.*, 1976) contained no detectable LH-releasing hormone (LH-RH), although rat CSF contained LH-RH (Morris *et al.*, 1975; Cramer and Barraclough, 1975) and thyrotrophin releasing hormone (Knigge and Joseph, 1974). The aim of the present study is to measure the concentrations of LH-RH in both CSF and plasma in women

with various conditions and to examine the relation between the concentrations of LH-RH in both biological fluids.

Materials and Methods

Thirty-six women aged between 21 and 50 years voluntarily participated in this study; fourteen were in the follicular phase and 9 in the luteal phase of the normal menstrual cycle, 6 were post-menopausal and 7 were pregnant. Paired samples of plasma and CSF were obtained just before gynecological or obstetric operations under the premedication with pentazocine and hydroxyzine hydrochloride. The levels of LH-RH, LH and FSH in plasma and the level of LH-RH in CSF were determined. LH and FSH in CSF were not measured because the volume of CSF that was sufficient to measure could not be obtained. LH and FSH were measured by double antibody radioimmunoassay as reported previously using 2nd IRP-HMG as the standard (Aono *et al.*, 1976). LH

in the plasma of pregnant women was determined by specific LH radioimmunoassay using the LH- β -subunit which was not affected by 500 IU/ml of hCG in the samples (Miyake *et al.*, 1976). Antiserum to synthetic LH-RH conjugated with human serum albumin at the N-terminal was raised in rabbits, and used at a final dilution of 1:200,000 in the assay. Synthetic LH-RH was used as a standard, and it was iodinated by a modification of the chloramine-T method using 5 μ g of LH-RH, 1 mCi of I^{125} , 50 μ g of chloramine T and 125 μ g of sodium metabisulfite. The labelled hormone was separated from inorganic iodine by the passage through a column of carboxymethylcellulose. The specific activity of iodinated LH-RH was 50–100 μ Ci/ μ g. The lower limit of sensitivity of the LH-RH assay was 0.6 pg/ml, and there was no cross-reaction with any other hypothalamic, pituitary or placental hormones under the physiologic concentrations. The antiserum to LH-RH seem to be specific to C-terminal of LH-RH for the rates of cross-reactivity with LH-RH³⁻⁶ and LH-RH³⁻⁸ were less than 0.01% although those with LH-RH³⁻¹⁰ and LH-RH⁴⁻¹⁰ were about 100%. One ml of the plasma and one of ml CSF were extracted with 3 volumes of methanol and centrifuged, and the supernatant was evaporated at 40°C under a stream of air. The residue was dissolved in 0.6 ml of 0.01 M phosphate buffered saline containing 1% serum albumin immediately before assay. The present LH-RH radioimmunoassay gives a recovery of $85.6 \pm 3.2\%$ in the range of 5 to 100 pg/ml.

Results and Discussion

The LH-RH levels found in the plasma and CSF and the LH and FSH levels in the plasma are shown in Fig. 1. The mean $\pm$ SE plasma LH-RH levels were 3.75 ± 0.40 pg/ml in the follicular phase, 4.15 ± 0.52 pg/ml in the luteal phase, 12.73 ± 1.42 pg/ml in post-menopausal women and 2.81 ± 0.63 pg/ml in pregnant women at term. The plasma LH-RH levels of post-menopausal women were significantly ($p < 0.001$) higher than those in the remaining three groups. No immunoreactive LH-RH in the CSF was found in 29 out of 36 subjects, and the LH-RH in the remaining 7 subjects were less than 0.95 pg/ml. The LH-RH levels in the CSF of post-menopausal women were not higher than those in other groups. The mean plasma levels of LH and FSH in post-meno-

Fig. 1. LH-RH levels in the plasma and CSF, and LH and FSH levels in the plasma of 36 women. Numbers in parentheses represent numbers of subjects. The hatched area indicates the minimum level detectable by our LH-RH radioimmunoassay. The bars show the median values. Results were analyzed by the unpaired Student's *t* test. The difference between the plasma level in post-menopausal women and in the other three groups was statistically significant ($p < 0.001$).

pausal women were 57.8 ± 15.8 mIU/ml and 69.3 ± 8.9 mIU/ml, respectively, and these levels were significantly ($p < 0.001$) higher than those in women in the follicular and luteal phase which ranged 5 and 10 mIU/ml. The levels of LH and FSH in pregnant women were less than 0.5 mIU/ml, and these levels were significantly ($p < 0.001$) lower than those in normal cyclic women.

The present results suggest that it is not clear that there is any correlation between the LH-RH level of plasma and that of CSF, because little or no LH-RH is present

in the CSF, although the LH-RH level in the plasma is mostly parallel to levels of plasma gonadotrophins. The present findings in women at various condition are consistent with the observation of Mortimer *et al.* (1976) who found no detectable LH-RH in CSF from 5 human subjects, and that of Coppins *et al.* (1977) who found no immunoreactive LH-RH in ovine CSF from the third ventricle. However, the present results contradict the previous reports that CSF in rats contained relatively high quantities of immunoreactive LH-RH (Morris *et al.*, 1975; Cramer & Barraclough, 1975). This discrepancy might possibly be due to the anatomical differences in hypothalamo-CSF system.

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