 ISSN 1007-9327 CN 14-1219/R WJG, 1998;4(1):92

Plasma leucine enkephalin content in patients with "Liver-blood deficiency" syndrome and clinical significance *

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Subject headings "Liver-blood deficiency" syndrome/ pathophysiology; enkephalin/blood; radioimmunoassay

INTRODUCTION

Recent studies revealed that the leucine enkephalin (L-EK) was distributed in rat brains with a higher concentration in hypothalamus and little in the pituitary gland[1], but the study on the relationship of L-EK with "liver-blood deficiency" syndrome (LBDS) was lacking. We determined the plasma L-EK levels in LBDS patients with clinical features of iron deficiency anemie (IDA) and/or chronic aplastic anemia (CAA), so as to investigate their relationship.

MATERIALS AND METHODS

Patients and controls

The LBDS was determined by the traditional Chinese medicine (TCM) and the disease entities determined by the modern medicine; an integrated method was used in selecting the objects for study. All patients came from the departments of hematology and integrated TCM and modern medicine of our hospital, most of them were inpatients, and the diagnosis of LBDS was established by two clinicians. Among the 26 patients with LBDS, 23 were iron deficiency anemia (IDA) and 3 were chronic aplastic anemia(CAA), including 7 males and 19 females; age averaged 39.6 ± 9.0 (21 - 66) years. The 30 healthy controls were employees and blood donors of our hospital, including 15 of each sex, and age averaged 32.8 ± 10.2 (21-46) years.

Diagnostic criteria

The diagnostic criteria for IDA was in conformity with the "Diagnostic criteria and curative improvement standards of clinical diseases"[2], and those of CAA accorded with the 1987 Baoji Conference revised standard[3]. The diagnostic standard of LBDS followed the certified standard of our institute[4], including: a. dizziness; b. decreased visual acuity and/or blurred vision; c. numbness of the extremities; d. face, lip and nails pale and malnourishment; e. tongue pale and pulse taut and thready or thready. Patients presenting symptoms of b or c with additional two symptoms and excluding those displaying Yinxu (deficiency of *yin*), Yangxu (deficiency of *yang*) and Qixu (deficiency of *qi*) were diagnosed as LBDS.

Determination of plasma L-EK

Radioimmunoassay was used to determine the plasma level of L-EK. Three ml fasting blood samples were collected at 6-8 a.m. in test tubes containing 50 μl proinin and 40μl EDTA, mixed and centrifuged at 3000rpm for 15min immediately to isolate the plasma and stored at a -20℃ for determination. The reagent kit was provided by the Department of Neurobiology of Second Military Medical University, Shanghai, and the test was performed according to the manual of the kit. The instrument was the FJ-2107PY immuno-automatic counter of Xi'an 262 Factory.

Statistical analysis

The results were expressed as x ± s, and the difference was examined by Student's *t* test.

RESULTS

In the 26 LBDS patients L-EK was 60.83 ng/L ± 21.44 ng/L as compared with 43.22 ng/L ± 17.99 ng/L in 30 healthy controls ($P<0.01$).

DISCUSSION

The opioid peptides, or the so-called endogenous opioid substances include enkephalin, endorphine and dynorphin, the three major categories, with algether about twenty members in this family. Enkephalins come from prepro-enkephalin A, which is composed of 267 amino acids, including six met-enkephalin and one Leu-EK molecule, which was the first isolated opioid peptide in 1975 and the most abundant opioid in the brain. In the cardiovascular system enkephalin is distributed in the atrial and ventricular conductive system and in the nerve fibers of peripheral vascular walls[5,6]. EK is a kind of neural transmitter, which plays an important role in physiologic function[6]; it affects the hypothalamus, pituitary axis functional activities significantly, including the release of luteinizing hormone (LH), the follicular stimulating hormone (FSH) and the thyroid stimulating hormone (TSH)[7]. The functions of FSH and LH on the ovary are to stimulate the production of gametes and ovarian sex hormones. In circumstances of insufficient secretion of FSH and LH, ovarian functional activities are depressed and the secretion of estrogen and progesterone is defective and ultimately results in oligomenorrhea or amenorrhea and anovulation[8]. The result of our study demonstrated a significantly increased plasma level of L-EK in patients with LBDS, and the difference from the healthy controls was very significant. It is suggested that the underlying pathophysiologic basis of oligo or amenorrhea and infertility of female patients with LBDS might be ascribed to the deficient ovarian functional activity partially induced by the obviously increased enkephalin as shown by the increased plasma level of L-EK in our study.

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***Project supported by the National Natural Science Foundation of China, No. 39170881.**
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Received 1997-08-21.