LHRH Increases Plasma 7B2 Concentration in Normal Human Subjects

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Abstract

We studied the response of plasma 7B2 to LHRH and ovine corticotropin releasing hormone (o-CRH) in healthy young subjects. The plasma 7B2 concentration significantly increased from 78.3 ± 7.5 (mean ± SEM) to 102.0 ± 6.0 ng/L (142.7 ± 12.7% of the basal value; P < 0.01) following iv administration of LHRH in seven young subjects. On the other hand, no increase in plasma 7B2 was found after iv administration of o-CRH in six young subjects. These results, together with our previous report of no increase in plasma 7B2 after administration of TRH and GHRH in young subjects, suggest that pituitary 7B2 may be present in gonadotrophs and be released only by LHRH in physiological conditions.

A novel pituitary protein, ‘7B2’, was isolated from porcine and human pituitary gland (Hsi et al., 1982, Seidah et al., 1983). Recently, 7B2 cDNA was cloned and its base sequences were determined from a pituitary cDNA library of black-adapted South-Africa clawed toads, Xenopus laevis, using a differential hybridization technique (Martens, 1988). 7B2 consists of 185 amino acid residues and its molecular weight is about 20,800 (Martens, 1988). Using a specific RIA, 7B2-immunoreactivity (IR) has been found to be present not only in the pituitary gland which contained the largest amount among several tissues examined but also in the various regions of the central nervous system, thyroid gland, and adrenal medulla in the rat (Iguchi et al., 1984, Iguchi et al., 1985). In the anterior pituitary gland, it has been demonstrated that the majority of 7B2-IR is localized immunohistochemically in the gonadotrophs of rats (Leduc et al., 1987, Marcinkiewicz et al., 1987) and man (Steel et al., 1988). In addition, Deng et al. (1986) showed that 7B2 is released from cultured rat anterior cells by LHRH. These data led us to examine whether LHRH could increase plasma 7B2 in healthy young subjects.

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Subjects and Methods

All twelve young subjects (9 men and 3 women; 20–29 yrs) were tested at bed rest after an overnight fast. LHRH (100 µg; Tanabe Pharmaceutical Co. Ltd., Osaka, Japan) and o-CRH (100 µg; Peptide Institute, Minoh, Japan) were given iv to 7 subjects (5 men and 2 women) and 6 subjects (4 men and 2 women), respectively. Informed consent was obtained from each subject before the study. Synthetic o-CRH-(1–41)NH₂ was sterilized by filtration through a 0.22-µm filter (Millipore Corp., Tokyo, Japan) before administration. Blood samples were collected at 15 min before and 0, 15, 30, 45, 60, 90, and 120 min after iv injection of LHRH and o-CRH and collected in an ice-cold glass tube containing EDTA-2Na. The basal level was determined as the average for the plasma at -15 and 0 min. The plasma sample separated by centrifugation was kept at -20°C until assay. The RIA for 7B2 was performed as described previously (Iguchi et al., 1984, Natori et al., 1988). A synthetic fragment, 23–39 amino acids of authentic 7B2, was used as a tracer after iodination and a standard. The 7B2-antiserum used in the present study was a gift from Dr. S. R. Bloom (Hammersmith Hospital, London). Plasma LH and FSH levels were assayed in duplicate with RIA kits (Eiken Immunochemical Laboratory, Tokyo, Japan). Plasma ACTH concentrations were determined in duplicate by the modified method of Nicholson et al. (1984) using [¹²⁵I] ACTH (New England Nuclear Research Products, Boston, MA, USA) and an antibody obtained from IgG Corp. (Nashville, TN, USA). The statistical analysis between basal levels and peak levels of plasma 7B2, LH, FSH and ACTH after administration of LHRH and o-CRH, was performed by Student's t-test.

Results

Changes in plasma 7B2 concentrations

Table 1. Changes in plasma 7B2 concentrations after iv administration of LHRH in each subject

<table>
<thead>
<tr>
<th>Subject/Time (min)</th>
<th>−15</th>
<th>0</th>
<th>15</th>
<th>30</th>
<th>45</th>
<th>60</th>
<th>90</th>
<th>120</th>
<th>150</th>
</tr>
</thead>
<tbody>
<tr>
<td>24 yr-old/M</td>
<td>56.0</td>
<td>53.8</td>
<td>73.1</td>
<td>87.9</td>
<td>79.7</td>
<td>64.7</td>
<td>65.1</td>
<td>56.7</td>
<td>59.0</td>
</tr>
<tr>
<td>25 yr-old/M</td>
<td>58.9</td>
<td>56.6</td>
<td>94.4</td>
<td>105.3</td>
<td>53.9</td>
<td>58.0</td>
<td>56.6</td>
<td>58.0</td>
<td>45.6</td>
</tr>
<tr>
<td>26 yr-old/M</td>
<td>98.0</td>
<td>131.3</td>
<td>119.3</td>
<td>96.3</td>
<td>143.6</td>
<td>117.4</td>
<td>115.9</td>
<td>111.0</td>
<td>103.0</td>
</tr>
<tr>
<td>26 yr-old/M</td>
<td>74.3</td>
<td>55.4</td>
<td>111.5</td>
<td>85.9</td>
<td>77.6</td>
<td>77.5</td>
<td>62.7</td>
<td>94.4</td>
<td>56.7</td>
</tr>
<tr>
<td>29 yr-old/M</td>
<td>66.9</td>
<td>66.7</td>
<td>94.7</td>
<td>100.8</td>
<td>94.7</td>
<td>88.9</td>
<td>78.5</td>
<td>94.8</td>
<td>71.2</td>
</tr>
<tr>
<td>20 yr-old/F</td>
<td>69.3</td>
<td>63.4</td>
<td>85.6</td>
<td>103.6</td>
<td>91.1</td>
<td>74.2</td>
<td>74.0</td>
<td>95.7</td>
<td>68.8</td>
</tr>
<tr>
<td>20 yr-old/F</td>
<td>124.7</td>
<td>120.3</td>
<td>137.8</td>
<td>133.9</td>
<td>123.5</td>
<td>106.0</td>
<td>94.7</td>
<td>112.0</td>
<td>108.8</td>
</tr>
</tbody>
</table>

Mean ± SEM 78.3 ± 7.5 102.3 ± 8.3 102.0 ± 6.0 94.9 ± 11.4 83.8 ± 8.2 78.2 ± 7.8 88.9 ± 8.6 73.3 ± 9.0

The unit for each value is ng/L.

M; male subject, F; female subject.

Table 2. Changes in plasma 7B2 concentrations after iv administration of o-CRH in each subject

<table>
<thead>
<tr>
<th>Subject/Time (min)</th>
<th>−15</th>
<th>0</th>
<th>15</th>
<th>30</th>
<th>60</th>
<th>90</th>
<th>120</th>
</tr>
</thead>
<tbody>
<tr>
<td>24 yr-old/M</td>
<td>83.4</td>
<td>55.3</td>
<td>62.3</td>
<td>71.8</td>
<td>81.9</td>
<td>73.5</td>
<td>66.6</td>
</tr>
<tr>
<td>24 yr-old/M</td>
<td>47.3</td>
<td>53.2</td>
<td>56.0</td>
<td>58.4</td>
<td>59.6</td>
<td>54.0</td>
<td>54.3</td>
</tr>
<tr>
<td>26 yr-old/M</td>
<td>75.9</td>
<td>83.9</td>
<td>75.1</td>
<td>79.4</td>
<td>87.9</td>
<td>88.7</td>
<td>76.2</td>
</tr>
<tr>
<td>27 yr-old/M</td>
<td>102.4</td>
<td>90.9</td>
<td>100.9</td>
<td>88.8</td>
<td>87.9</td>
<td>89.4</td>
<td>79.2</td>
</tr>
<tr>
<td>20 yr-old/F</td>
<td>91.4</td>
<td>89.3</td>
<td>88.0</td>
<td>80.6</td>
<td>78.8</td>
<td>104.4</td>
<td>84.4</td>
</tr>
<tr>
<td>20 yr-old/F</td>
<td>78.3</td>
<td>102.9</td>
<td>85.3</td>
<td>87.7</td>
<td>101.1</td>
<td>105.0</td>
<td>107.7</td>
</tr>
</tbody>
</table>

Mean ± SEM 79.6 ± 7.1 77.9 ± 6.9 77.8 ± 4.6 82.9 ± 5.6 85.3 ± 8.0 78.1 ± 7.3

The unit for each value is ng/L.

M; male subject, F; female subject.
in each subject after the administrations of LHRH and o-CRH were shown in Table 1 and in Table 2, respectively. The mean plasma 7B2 concentration increased from 78.3 ± 7.5 (mean ± SEM) to 102.0 ± 6.0 ng/L after an administration of LHRH; however, the plasma 7B2 concentrations were almost unchanged in the o-CRH test.

Fig. 1 showed that mean plasma 7B2 level rose significantly over the basal level which is designated as 100% after administration of LHRH in 7 young healthy subjects, though the rate of increase was relatively lower than in plasma LH and FSH in response to LHRH. The plasma 7B2 level did not change significantly after the administration of o-CRH in spite of the marked response of plasma ACTH to o-CRH in 6 subjects (Fig. 2).

Discussion

In the present study, we found that the plasma 7B2 concentration increased significantly after iv administration of LHRH in young healthy subjects. But no increase in 7B2 was observed after iv administration of CRH. Previously, we reported that plasma 7B2 did not increase significantly after iv administration of GHRH or TRH in young subjects (Natori et al., 1988). This suggests that LHRH may be a hypothalamic hormone responsible for 7B2
release from the anterior pituitary gland among the known endogenous hypothalamic releasing hormones, indicating a close physiological relationship between 7B2 and gonadotropins which was also proposed by several other investigators. With immunohistochemistry. Leduc et al. (1987), Marcinkiewicz et al. (1987) and Steel et al. (1988) have demonstrated that the majority of cells immunostained by 7B2 antibody were defined as gonadotrophs in the anterior pituitary gland of the rat and man. In the culture experiment with rat anterior pituitary cells, Deng et al. (1986) found LHRH-induced release of 7B2 into the culture media. However, the percent increase in 7B2 release due to LHRH was much less than that in LH or FSH release due to LHRH, and consistent with our in vivo data. Further studies will be required to resolve this issue.

In conclusion, we noted LHRH could increase the plasma concentration of 7B2 in young subjects, suggesting the involvement of 7B2 in the pituitary-gonadal axis.

Acknowledgements

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References


Marcinkiewicz, M., S. Benjannet, N. G. Seidah, M. Cantin and M. Chrétien (1987). The


