The Effect of Pentobarbital Sodium on Growth Hormone Release in Shiba Goats

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Abstract The effect of intravenous (iv) injection of pentobarbital sodium (PB) on plasma concentration of growth hormone (GH) in the Japanese Shiba goats was studied. An acute significant increase in plasma GH was observed in the goats after a single PB injection of 19 mg/kg BW. Plasma GH concentration remained unchanged after injection of vehicle or 10 mg/kg BW PB in the goats. The present results suggested that anesthetic dose of PB induced a release of GH in the Shiba goats.

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Growth hormone (GH) secretion is mainly regulated by two hypothalamic factors, growth hormone-releasing hormone and somatostatin (SRIF). Recently, it has been suggested that the other factors such as neurotransmitters may be involved in regulation of GH⁵⁾.

Pentobarbital sodium (PB) stimulates GH release in rats^{3,7,9,12)}. However, it is possible that there are species differences in this phenomenon since the effect of PB on GH release has not been observed in dogs¹⁰⁾. Although it has been suggested that hypothalamic factors may play a major role in this phenomenon¹¹⁾, the mechanism remains to be clarified. Moreover, the effect of PB on GH secretion in ruminant animals has never been reported. In the present study, the effect of PB on GH secretion was

investigated in order to clarify the central nervous control of GH secretion in Japanese Shiba goats.

Materials and Methods

Five female Shiba goats weighing 12.8 ± 1.7 kg in BW were used in the following experiments : two doses of pentobarbital sodium (PB, Somnopentyl, Pitman-Moore, Mundelein, IL, 10 mg/kg BW or 19 mg/kg BW) or 10% ethanol in physiological saline (vehicle of Somnopentyl, 0.6 ml/kg BW as control) were administered intravenously by a single injection. Since 19 mg/kg BW of PB had induced a significant increase in GH concentration in Shiba goats during brain surgery (unpublished data in our laboratory), we determined to use the same dose and its half dose in this experiment.

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Before injections of PB or saline, animals were normal and standing. However, they fell and began to sleep just after injections of both doses of PB.

Blood samples were taken from animals through an indwelling catheter previously inserted into the external jugular vein by venipuncture. Blood was collected at 10-min intervals for 1.5 h before and for 2.5 h after the drug injection. The blood samples were taken into heparinized tubes, and plasma was immediately separated by centrifugation (2,500 r.p.m., 5 min). The plasma was stored at -20°C until GH assay.

GH concentrations were determined in $100\mu\text{l}$ aliquots of plasma by a double-antibody radioimmunoassay⁸. The GH standard preparation used and hormone for iodination were bGH-B1. The parallelism between caprine plasma GH and bGH was described previously⁶. All samples were assayed in a single assay. The least detectable value of assay was 0.3 ng/ml . The intra-assay coefficients of variation in this assay was 4.0%.

Data were represented as mean $\pm$ SE. Statistical significance of differences were determined by Student's *t*-test or Cochran-Cox test after analysis of uniformity of variance by Bartlett's test¹⁴.

Results and Discussion

Injections of a vehicle placebo failed to induce any significant change in plasma GH levels in female Shiba goats as shown in Fig. 1-A. When the 10 mg/kg BW of PB was injected, plasma GH level tended to increase but not significantly 20 to 40 min after the injection compared with the preinjection values (Fig. 1-B). However, injection of 19 mg/kg BW of PB induced a significant five-fold increase in GH concentration ($P < 0.05$) 30 min after the injection compared with the preinjection values (Fig. 1-C). The elevated GH level declined within 30 min after the peak and returned to the preinjection level 2 h after the injection.

Both doses of PB injection induced deep anesthesia. The duration of deep sleep were 8-50

Fig. 1. The mean plasma GH level after iv injection of vehicle (A) or 10 mg/kg BW of pentobarbital sodium (B) and 19 mg/kg BW of PB (C). Arrows indicate time of injection. The mean GH concentration of 10 fractions before the injection was taken as a preinjection level. An asterisk indicates the statistical significance; * $P < 0.05$ as compared with the preinjection level. Each value represents the mean $\pm$ SE.

and 25–120 min after the administration of 10 and 19 mg/kg BW of PB, respectively.

The present study clearly demonstrated that anesthetic dose of PB induced a remarkable GH release in goats. Unlike that of the dog¹⁰⁾, our result that PB induced GH release in Shiba goats was consistent with those reported in rats^{3, 7, 9, 12)}. In rats, since the GH releasing effect of PB was blocked by lesion of the hypothalamic ventromedial nucleus or isolation of mediobasal hypothalamus¹¹⁾, and since pentobarbital failed to stimulate GH release from dispersed and cultured rat anterior pituitary cells *in vitro*²⁾, it has been suggested that the drug exerted an effect via the central nervous system. It has been reported that PB decreased SRIF release in rat hypophyseal portal blood⁴⁾ or brain¹³⁾. Furthermore, SRIF blocked PB-stimulated GH release in rats¹⁾. However, there was also a possibility that GH-releasing hormone might be involved in PB-induced GH stimulation in rats because PB-induced GH release was blocked by the destruction of the arcuate nucleus. In conclusion, the present results suggested that anesthetic dose of PB induced a significant GH release in goats.

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References

- 1) Brazeau P, Rivier J, Vale W, Guillemin R. Inhi-

- bition of growth hormone secretion in the rat by synthetic somatostatin. *Endocrinology*, 94 : 184–187. 1974.
- 2) Brown M, Vale W. Growth hormone release in the rat : effects of somatostatin and thyrotropin-releasing factor. *Endocrinology*, 97 : 1151–1156. 1975.
- 3) Chihara K, Arimura A, Chihara M, Schally AV. Effect of intraventricular administration of anti-somatostatin gamma-globulin on the lethal dose-50 of strychnine and pentobarbital in rats. *Endocrinology*, 103 : 912–916. 1978.
- 4) Chihara K, Arimura A, Schally AV. Immunoreactive somatostatin in rat hypophyseal portal blood : effects of anesthetics. *Endocrinology*, 104 : 1434–1441. 1979.
- 5) Devesa J, Lima L, Tresguerres JAF. Neuroendocrine control of growth hormone secretion in humans. *Trends Endocrinol. Metab.*, 3 : 175–183. 1992.
- 6) Hashizume T, Kanematsu S. Effects of cholecystokinin octapeptide on the release of growth hormone in perfused pituitary and hypothalamus of the goat. *Anim. Sci. Technol. (Jpn.)*, 62 : 343–350. 1991.
- 7) Howard N, Martin JM. A stimulatory test for growth hormone release in the rat. *Endocrinology*, 88 : 497–499. 1971.
- 8) Johke T. Effect of TRH on circulating growth hormone, prolactin and triiodothyronine levels in the bovine. *Endocrinol. Japon*, 25 : 19–26. 1978.
- 9) Kokka N, Garcia JF, George R, Elliott HW. Growth hormone and ACTH secretion : evidence for an inverse relationship in rats. *Endocrinology*, 90 : 735–743. 1972.
- 10) Lovinger RD, Connors MH, Kaplan SL, Ganong WF, Grumbach MM. Effect of L-dihydroxyphenylalanine (L-dopa), anesthesia and surgical stress on the secretion of growth hormone in the dog. *Endocrinology*, 95 : 1317–1321. 1974.
- 11) Martin JB. Studies on the mechanism of pentobarbital-induced GH release in the rat. *Neuroendocrinology*, 13 : 339–350. 1974.
- 12) Settembrini BP, Gallardo MGP, Tramezzani JH. Pituitary graft inhibits the release of prolactin and growth hormone induced by sodium pentobarbital. *Med. Sci. Res.*, 16 : 765–766. 1988.
- 13) Takahashi H, Shintani Y, Okauchi T, Ishikawa M, Bando H, Azekawa T, Morita Y, Saito S. Measurement of somatostatin release in rat brain by microdialysis. *J. Neurosci. Methods*, 52 : 33–38. 1994.
- 14) Wakabayashi K. Arrangement of experimental data. 44–57, 90–133. Baihukan. Tokyo. 1984.