Calcitonin Gene-Related Peptide (CGRP) as a GH Secretagogue in Rat and Human Pituitary Tumoral Cells

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CALCITONIN gene-related peptide (CGRP), a 37-amino-acid neuropeptide identified as a potent vasodilator [1], is widely distributed in the central nervous system. The presence of CGRP and its binding sites has been found in the pituitary [2–4], but its role in regulating pituitary function remains to be determined. In the present study, we investigated the effect of CGRP on the secretion of GH *in vitro* by using rat pituitary tumoral cells (GH₃ cells) and human pituitary adenoma cells.

Materials and Methods

GH₃ cells were purchased from the American Type Culture Collection and grown in Ham's F-10 medium supplemented with 15% horse serum, 2.5% fetal calf serum, penicillin G (50 U/ml), and streptomycin (50 µg/ml) in a humidified atmosphere of 95% air and 5% CO₂, as previously described [5]. Human somatotroph adenoma tissues were obtained by transsphenoidal surgery. The adenoma cells were dispersed with 0.25% trypsin, plated onto poly-L-lysine-coated microplates (about 25,000 cells/well), and cultured for 48 h in the same medium as GH₃ cells. The tumoral cells were incubated with various concentrations of CGRP dissolved in Ham's F-10 medium containing

0.1% bovine serum albumin for 4 h. The medium was collected and stored at –20 °C until assayed. The concentrations of rat and human GH were measured by specific radioimmunoassay provided by NIDDK and with an immunoradiometric assay kit provided by Daiichi RI Lab, respectively. The results were expressed as the mean ± SEM.

Results

The addition of CGRP (1 pM–100 nM) evoked GH secretion from GH₃ cells in a bell-shaped manner. One hundred pM of CGRP caused the maximum increase in GH secretion ($172 \pm 14\%$ of control, $P < 0.01$ vs. control: Fig. 1). In a case of human somatotroph adenoma (Case 1), GH secretion was stimulated by CGRP (1–100 nM) in a dose-related manner. In another case (Case 2), both 1 and 100 nM of CGRP evoked GH secretion with the same potency (Fig. 2).

Discussion

In the present study, we have shown that CGRP significantly stimulated GH secretion from rat and human pituitary tumoral cells *in vitro*. Previous studies with normal rat pituitary cells have shown that CGRP acted on GH secretion in a biphasic manner, namely 100 pM of CGRP inhibited and 100 nM of CGRP stimulated GH release [6]. These discrepancies may be explained by the different CGRP receptor subtypes and/or different intracellular mechanisms of CGRP action between

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Fig. 1. Effects of various doses of CGRP on GH secretion from rat pituitary cell line, GH₃ cells. Means $\pm$ SEM are shown. * P <0.01 vs. control.

normal and tumoral pituitary cells. It is noteworthy that the effects of CGRP on GH secretion from GH₃ cells were bell-shaped, suggesting that a different mechanism operated when a larger dose of CGRP was used. CGRP present in gonadotrophs of rat and human pituitary gland [7] may be a new member of the GH secretagogues acting in a paracrine manner.

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Fig. 2. Effects of CGRP on GH secretion from somatotroph adenoma cells of two acromegalic patients. Means $\pm$ SEM are shown. * P <0.01, ** P <0.05 vs. B (control).

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