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Enhancement of Insulin Secretion by Human Chorionic Somatomammotropin and Related Hormones (39379)

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In response to a sustained glucose stimulus, the isolated perfused rat pancreas secretes insulin in a diphasic pattern characterized by an initial rapid secretory phase followed by a decline in the rate of secretion and then a slowly rising rate of insulin secretion (1). Hypophysectomy alters this diphasic response by producing a marked reduction in the initial secretory response and a reduction that is not so striking in the second or rising rate of insulin secretion (2). Treatment of hypophysectomized rats for a period of 2 weeks with BGH[1] completely restores the second secretory phase to normal but has little effect upon the first secretory phase (2). The amounts of BGH used in these experiments was 500 μg/day and the last injection was given the day prior to perfusion. Subsequent experiments (Bennett and Curry, unpublished data) indicated that the minimum effective dose of BGH for restoring the second secretory phase was approximately 200 μg/day. In this communication we report the comparative effectiveness of HCS and related hormones (HGH, PL-HGH, and OP) in restoring the secretory response of the perfused pancreas from hypophysectomized rats.

Experimental. The perfusion technique and medium were those previously described (1) except that the perfusion medium contained Mg^{2+} at 2.4 mequiv/liter. All animals used were male rats of the Sprague–Dawley strain weighing approximately 300 g at the time of hypophysectomy. Animals were hypophysectomized by the parapharyngeal approach and were used approximately 4 weeks after hypophysectomy. The completeness of hypophysectomy was checked by inspection of the region of the sella using a binocular microscope. All hormones were given ip at a dose of 200 μg/day for 6 days with the last, or sixth, injection being given the morning of perfusion. Animals were fasted for approximately 20–24 hr prior to having their pancreases removed for perfusion. The isolation and purification of HCS (3, 4), HGH (5), and OP (6) have been described previously. PL-HGH was prepared by the procedure of Li and Graf (7), except that the final product was purified further by exclusion chromatography on Sephadex G-100 in 0.01 *M* NH_4HCO_3 of pH 8.2. Insulin was determined by the method of Grodsky and Forsham (8). The standard glucose stimulus (300 mg/dl) was started after a 10-min equilibration period and continued for 38 min.

Results and discussion. Histological studies of the pancreases were not done, but it is significant that the rats treated with HGH showed an average body weight gain of about 5 g/day as did those treated with PL-HGH. All rats treated with HCS and OP lost small amounts of body weight.

The pertinent data from these experiments are summarized in Table I. When one compares the data from the normal control animals with the hypophysectomized control animals, it is clear that as a result of hypophysectomy, the first secretory phase is strikingly reduced ($p < 0.001$) and the second secretory phase also shows a significant reduction ($p < 0.01$). The animals pretreated with HGH do not show significantly increased insulin release during the first phase compared to the hypophysectomized controls, but the first phase remains significantly depressed below the normal controls. On the other hand, the amount of insulin released during the second secretory phase

[1] Abbreviations: BGH, bovine growth hormone; HCS, human chorionic somatomammotropin; HGH, human pituitary growth hormone; PL-HGH, plasmin-modified human pituitary growth hormone; OP, ovine prolactin.

TABLE I. *In Vitro* SECRETION OF INSULIN BY THE PANCREASES OF HYPOPHYSECTOMIZED RATS TREATED WITH HCS, HGH, PLASMIN-MODIFIED HGH, AND OVINE PROLACTIN.

	First secretory phase: Min 2-6			Second secretory phase: Min 25-40		
Experimental group[a]	Insulin release[b] (μg)	*P* vs normal control	*P* vs Hypx control	Insulin release[b] (μg)	*P* vs normal control	*P* vs Hypx control
Normal controls (4)	0.38 ± 0.03		<0.001	2.91 ± 0.19		<0.01
Hypx controls (7)	0.13 ± 0.02	<0.001		1.86 ± 0.17	<0.01	
Hypx + HGH (7)	0.23 ± 0.04	<0.05	NS	2.62 ± 0.29	NS	<0.05
Hypx PL-HGH (4)	0.32 ± 0.08	NS	NS	3.31 ± 0.44	NS	<0.02
Hypx + HCS (8)	0.26 ± 0.05	NS	<0.05	3.38 ± 0.35	NS	<0.01
Hypx + OP (4)	0.17 ± 0.03	<0.01	NS	2.41 ± 0.45	NS	NS

[a] Number of animals in parentheses.
[b] Mean ± SE.

is elevated and does not differ significantly from the amount secreted by the normal controls and is significantly higher than the amount secreted by the hypophysectomized controls.

PL-HGH produces an even more striking restoration of the ability of the pancreas to secrete insulin than does the intact HGH molecule. The total insulin released during the first phase is almost completely restored but there was large variation in the response and a small number of animals in the group. The total amount of insulin released during the second secretory phase is restored to that of the normal controls and is significantly elevated above the hypophysectomized controls.

The data from animals pretreated with HCS exhibit an increased insulin release during both the first and the second phase. In both instances the insulin release is significantly greater than that shown by the control hypophysectomized animals, and in neither case is it significantly different from that of the control normal animals. In striking contrast to the above findings is the lack of effect of OP on the first phase of insulin release. It is of interest that larger amounts of OP (5 mg/day) do produce some increased secretion in the first phase and also restore the second phase to normal (9).

The comparative effectiveness of HCS, HGH, and OP as recorded in Table I is somewhat surprising in view of their primary structures and biological properties. HCS is a potent lactogenic hormone but a weak growth-promoting agent (3, 10, 11) in comparison with HGH, whereas OP is the most potent lactogenic hormone and is practically free of growth-promoting activity in the rat (12). When the amino acid sequences of HCS (13), HGH (14), and OP (6) were compared, the homology between HGH and HCS amounted to 96% and the homology between HCS and OP was 60% (15).

The effect of HCS on insulin secretion is in accord with results reported by Malaisse *et al.* (16) who used pieces of pancreas in an *in vitro* system. We are unaware of any reports of effects of PL-HGH on insulin secretion but this material has been reported to be an active diabetogenic agent (17). It may also be noted that the growth-promoting and lactogenic activities of HGH are not altered by hydrolysis with plasmin (7).

There is good evidence that GH promotes insulin synthesis in response to a glucose stimulus (18) as measured by incorporation of radioactive leucine into insulin. On the other hand under the condition of these experiments, newly synthesized insulin is not necessarily released (19) nor does our experimental design permit such differentiations. What these experiments do demonstrate is that the ability of these four related hormones to produce bodily weight gain (presumably involving synthesis of somatic protein) cannot be correlated with their ability to promote insulin secretion in response to glucose. This ability does correlate with a high degree of homology in their amino acid sequences.

Summary. Hypophysectomized rats were treated for 6 days with 200 μg per day of either human chorionic somatomammotropin, human pituitary growth hormone, plas-

min-modified human pituitary growth hormone, or ovine prolactin. All hormone preparations except ovine prolactin enhanced the ability of the pancreases of hypophysectomized rats to secrete insulin in the isolated pancreas perfusion system.

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