Growth Hormone and Prolactin Secretion in Hypophysial Stalk-Transected Pigs as Affected by Growth Hormone and Prolactin-Releasing and Inhibiting Factors (43179)

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Abstract. **Control of growth hormone (GH) and prolactin (PRL) release was investigated in hypophysial stalk-transected (HST) and stalk-intact pigs by determining the effects of analogs of GH-releasing factors (GHRF), somatostatin (SRIF), arginine, thyrotropin-releasing hormone, α-*methyl*-ρ-tyrosine, and haloperidol. HST and control gilts were challenged with intravenous injections of human pancreatic GHRF(1–40)OH, thyrotropin-releasing hormone, and analogs of rat hypothalamic GHRF. HST animals remained acutely responsive to GHRF by releasing 2-fold greater quantities of GH than seen in controls. This occurred in spite of a 38% reduction in pituitary gland weight and a 32 and 55% decrease in GH concentration and total content. During SRIF infusion, GH remained at similar basal concentrations in HST and control gilts, but increased immediately after stopping SRIF infusion only in the controls. Releasable pituitary GH appears to accumulate during SRIF infusion. GHRF given during SRIF infusion caused a 2-fold greater release of GH than seen in animals receiving only GHRF. Arginine increased ($P < 0.05$) GH release in controls, but not in HST gilts, which suggests that it acts through the central nervous system. Basal PRL concentrations were greater ($P < 0.05$) in HST gilts than in control gilts. TRH acutely elevated circulating PRL ($P < 0.001$) in HST gilts, suggesting that it acts directly on the pituitary gland. Haloperidol, a dopamine receptor antagonist, increased circulating PRL in controls but not in HST animals. α-*Methyl*-ρ-tyrosine did not consistently increase circulating PRL, however, suggesting that it did not sufficiently alter turnover rate of the tyrosine hydroxylase pool. The results indicate that the isolated pituitary after HST remains acutely responsive to hypothalamic releasing and inhibiting factors for both GH and PRL release in the pig.** [P.S.E.B.M. 1991, Vol 196]

The hypothalamus regulates both basal and episodic secretion of growth hormone (GH) and prolactin (PRL) in pigs (1–3). Hypothalamic deafferentation revealed that there is a dependency of the ventral medial hypothalamus upon neural connections traversing the anterior and posterior aspects of the porcine hypothalamus for episodic release of GH, but not PRL release (3). It is thought that endogenous release of GH-releasing factor (GHRF) and GH release-inhibiting hormone, somatostatin (SRIF), control GH secretion, whereas PRL secretion probably is regulated by both PRL release-inhibiting factors and PRL-releasing factors (4–7). After hypophysial stalk transection (HST), normal episodic release of GH and PRL is abolished in pigs and cattle (1, 2, 8–10), and these animals have depressed growth rates (11, 12). Furthermore, an inhibitory effect of the hypothalamus is clearly indicated by consistently elevated peripheral blood concentrations of PRL seen soon after HST (1, 10). These data indicate that hypophysial stalk-transected animals of different species are useful to study comparative effects of releasing and inhibiting substances on pituitary hormone secretion.

GHRF can stimulate GH release in humans, rats,

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pigs, and cattle (5, 6, 13), whereas substances such as vasoactive intestinal peptide and dopamine effectively stimulate and inhibit PRL secretion, respectively (7). The effects of neuroendocrine modulators of GH and PRL secretion in HST pigs have not been reported and observations to date indicate comparative species differences. For example, circulating basal concentrations of GH were not reduced after HST in pigs (2) and rhesus monkeys (14), animals in which the pituitary is not under endogenous SRIF and GHRF regulation. In contrast, GH decreased after HST in cattle (15) and rats (16). In species responsive to dopamine antagonists, circulating basal concentrations of PRL increase soon after HST in pigs (1), cattle (10, 17), rhesus monkeys (18), and rats (7).

The objective of this study was to provide comparative information on the effects of HST surgery on GH and PRL release and of different analogs of GHRF, SRIF, arginine, thyrotropin-releasing hormone (TRH), α-methyl-ρ-tyrosine, and haloperidol on the release of these pituitary hormones in the pig.

Materials and Methods

Animals. Yorkshire gilts were used throughout. An indwelling catheter (Tygon Microbore tubing; i.d., 1.27 mm; o.d., 2.29 mm; Fisher Scientific Co., Pittsburgh, PA) was inserted into a jugular vein for repeated blood sample collection (19). Each gilt was fasted for 24 hr before HST or sham operation (SOC). Anesthesia was induced by intravenous injection of thiamylal sodium (0.8–1.0 g; Surital, Parke Davis, Morris Plains, NJ) and maintained by a closed circuit system of halothane (4–9%; Ayerst Laboratories, Rouses Point, NY) and O_2 (500–1000 ml/min).

A modification of the transfrontal supraorbital approach as described by Anderson *et al.* (20) was used for HST. A nylon disc (6.0 or 8.0 mm in diameter and 0.45-mm thickness) was inserted between severed ends of the stalk to prevent vascular regeneration between the hypothalamus and pituitary gland (1, 2, 8, 20). Thirteen gilts were hypophysial stalk transected (Day 0), of which one animal died within 24 hr after surgery. Five SOC gilts were subjected to the same anesthesia procedure, but stalk section procedures were not conducted. Pigs were maintained in individual pens in a recovery room at 25°C and monitored continuously. Water and food consumption usually returned to normal within 36 hr after surgery.

On Day 9, HST gilts and their respective controls were killed by an overdose of thiamylal sodium and decapitated. Studies were conducted shortly after HST, because in long-term HST gilts PRL secretion decreases and becomes similar to that of stalk-intact controls (21). Completeness of stalk section and position of the nylon disc were confirmed. Pituitary, adrenal, and thyroid glands were recovered and weighed, and cross-sections were fixed in buffered 10% neutral formalin. Sections of pituitary gland were cut transversely at 6 μm, mounted, and stained with performic acid-Alcian blue-periodic acid-Schiff-orange G according to the method of Heath (22), whereas other sections were stained with hematoxylin and eosin for histologic examination.

Gilts received bolus intravenous injections of human pancreatic (hp) GHRF (1–40)OH (23), [Nle^{27}] rat hypothalamic (rh) GHRF(1–32)NH_2, [Nle^{27}]rhGHRF(1–29)NH_2, [N-α-Me-Tyr^1, Nle^{27}]rhGHRF(1–29)NH_2 (Salk Institute, San Diego, CA), and TRH (Sigma Chemical Co., St. Louis, MO). Somatostatin-14 (SRIF; Salk Institute) and arginine (Sigma) were administered intravenously by continuous infusion pumps (Harvard model 1201; Harvard Apparatus, Millis, MA). The GHRF, TRH, SRIF, and arginine were dissolved in 0.1% acetic acid (1 μg/μl) and then diluted with a sterile buffer solution. The buffer was physiologic saline (0.8% NaCl) containing 0.01 *M* $NaH_2PO_4 \cdot H_2O$ (pH 7.0), 0.01% ascorbic acid, and 0.1% bovine serum albumin or 0.1% HST serum. The [*α-methyl-ρ*-tyrosine]methyl ester (α-MT; Sigma) was diluted in sterile buffer solution at a concentration of 5 mg/ml. The haloperidol (HAL, McN-JR-1625; NcNeil Pharmaceutical, Spring House, PA) was dissolved in warm sterile water containing 6% lactic acid and was then diluted to a concentration of 5 mg/ml. Vehicle injections consisted of sterile buffer solution. Solutions for intravenous injection and infusion were prepared the day of experimentation.

Experimental Groups. In preliminary trials in which sequential blood samples were collected at 20-min intervals from pituitary intact gilts, GH release by hp GHRH [hpGHRF(1–40)OH] occurred in a range from 0.1 to 3.3 μg/kg body wt. In the first series of studies, prepubertal gilts were hypophysial stalk-transected (n = 6, 43 ± 2.8 kg body wt, mean ± SE; Day 0) or unoperated control (n = 5, 53 ± 1.2 kg body wt). On Days 2, 3, and 4, each hypophysial stalk-transected and control gilt was given an intravenous injection of either 0, 0.1, or 0.5 μg of hpGHRF(1–40)OH/kg body wt in random order to determine acute effects on GH secretion (Fig. 1). On Day 6, they were continuously infused with SRIF at a rate of 500 μg/hr for 60 min to determine GH secretion during 180 min; blood samples were collected at 15-min intervals (Fig. 2, A and B). The effects of an intravenous injection of *α-methyl-ρ*-tyrosine (10 mg/kg body wt given on Day 7) and haloperidol (0.1 mg/kg body wt given on Day 8) on PRL and GH secretion were compared in hypophysial stalk-transected and control gilts (Fig. 5). Subsequently, a second and independent group of control gilts was infused with SRIF (500 μg/hr) for 120 min and at 60 min given an intravenous injection of 0.5 μg of hpGHRF(1–40)OH or 25 μg of TRH to compare GH

blood concentrations with those given only hpGHRF(1–40)OH (Fig. 2C).

The second series of studies used three groups of intact prepubertal gilts. Plasma GH concentrations were determined during continuous infusion of hpGHRF(1–40)OH into two groups. Animals were bled at 15-min intervals for 120 min at 22 hr between periods for 7 days (Experiment A, 39 ± 1.3 kg body wt, $n = 6$) and bled at 15-min intervals for 180 min at 21 hr between periods for 6 days (Experiment B, 42 ± 1.4 kg body wt, $n = 6$). Effects of analogs of rat hypothalamic (rh) GHRF at six dosages on GH secretion were determined with the third group of prepuberal gilts in a Latin-square design for random assignment of treatments (Fig. 3).

The third series of studies used mature gilts (111 ± 6 kg body wt) ovariectomized approximately 30 days before HST ($n = 6$) or SOC ($n = 5$). Effects of anesthesia, TRH, and arginine on PRL and GH concentrations in peripheral plasma were determined. Blood samples were collected at 15-min intervals beginning 210 min before anesthesia and continued for 105 min after anesthesia (Fig. 4). On the morning of Day 8, hypophysial stalk-transected and control gilts were given an intravenous injection of 50 μg of TRH during 60 min of sequential blood sampling, and in the afternoon they were infused with arginine (0.5 mg/kg body wt for 45 min) during 90 min of sequential blood sampling (Fig. 4). The following morning (Day 9), hypophysial stalk-transected and control gilts were subjected to the same regimen of general anesthesia, as performed on the day of cranial surgery, and bled at 15-min intervals (Fig. 4).

Radioimmunoassays. Plasma concentrations of porcine GH were measured in a double antibody homologous radioimmunoassay (24). Sensitivity of the assay was 0.1 ng/tube. Mean interassay and intraassay coefficients of variation were 9.73% (six to eight determinations per sample, three samples) and 14.7% (four samples, six assays).

PRL concentrations in plasma were quantified by double antibody homologous radioimmunoassay (23). Sensitivity of the assay was 0.34 ± 0.15 ng/ml ($n = 8$), as computed by the method of Duddleson *et al.* (25). Mean intraassay coefficient of variation was 12.11% (five to eight determinations per sample, three samples) and the interassay coefficient of variation was 9.95% (two samples, six assays).

Hemipituitaries, in 1 ml of phosphate buffer, were thawed, and an additional 1 ml of 0.1 *M* phosphate buffer (pH 8.0) was added, pH adjusted to 8.0 with 1.0 *N* sodium hydroxide, and homogenized. Homogenates were incubated for 4 hr at 4°C and then centrifuged at 100,000*g* for 45 min. Supernatants were removed and frozen for subsequent radioimmunoassay. Pituitary extracts displayed inhibition of binding parallel to the reference preparations in each radioimmunoassay.

Statistical Analysis. Experimental units in this study were the individual gilts assigned to treatment groups at random. Temporal endocrine results were analyzed by split-plot analysis of variance. The main plot effect was treatment tested against gilt within treatment (i.e., gilt-to-gilt variation) as the whole-plot error term. The individual temporal sampling times, which were repeated measures on the same animals, and resultant interaction with treatment composed the subplot part of the analysis. Student's *t* test was used for comparison between treatment groups (26, 27).

Results

Organ Weights and Pituitary Hormone Concentrations. Postmortem examination of hypophysial stalk-transected gilts revealed that the hypophysial stalk had been completely severed. The nylon disc was in the proper location and had prevented vascular regeneration of the stalk in each animal. The effects of HST on weights of adrenal, thyroid, and pituitary glands are presented in Table I. Pituitary gland weight was 32% less ($P < 0.05$) 9 days after HST as compared with control gilts. In pituitary extracts, concentration and total content of PRL were markedly decreased ($P < 0.001$) in hypophysial stalk-transected as compared with control animals. This contrasts with the greater plasma concentrations of PRL in hypophysial stalk-transected than in control animals. Likewise, pituitary GH concentration and total content were less ($P < 0.05$) in hypophysial stalk-transected gilts compared with control gilts, but basal plasma levels of GH remained similar in both groups. Histologic examination of pituitary glands from HST gilts revealed persistence of secretory cells in the same areas of the adenohypophysis as seen in the controls. Acidophils with abundant cytoplasm were dispersed in anteromedial regions of these glands. Acidophils are associated with lactotrophs, somatotrophs, and adrenocorticotrophs.

hpGHRF(1–40)OH on GH Release after HST. Plasma GH peaked within 15 min after intravenous injection of 0.5 μg of hpGHRF(1–40)OH/kg body wt both in hypophysial stalk-transected (38 ± 8 ng/ml, $P < 0.005$) and control (15 ± 3.8 ng/ml, $P < 0.025$) gilts and then decreased to basal concentrations 90 min later (Fig. 1A). GH also peaked ($P < 0.025$) within 7.5 min after injection of 0.1 μg of hpGHRF(1–40)OH/kg body wt in hypophysial stalk-transected (19 ± 5.3 ng/ml) and control (7 ± 1.9 ng/ml) gilts and returned to basal levels within 60 min (Fig. 1B). Circulating GH remained similar in diluent-treated hypophysial stalk-transected (4.5 ± 0.4 ng/ml) and control (4.1 ± 0.6 ng/ml) gilts (Fig. 1C).

Continuous Infusion of SRIF on GH Release in Hypophysial Stalk-Transected and Control Gilts. Cir-

Table I. Effect of HST on Adrenal, Thyroid, and Pituitary Glands and PRL and GH in Gilts

Item	SOC gilts		HST gilts		Ratio
	n	Weight	*n*	Weight	HST:SOC
Body (kg)	5	111 ± 4.1[a]	6	97 ± 5.1	0.874
Adrenal (g)	5	4.9 ± 0.27	6	3.4 ± 0.30[b]	0.694
Thyroid (g)	5	11.5 ± 1.75	6	11.5 ± 2.51	1.000
Pituitary (mg)	5	342 ± 10	6	213 ± 21[b]	0.623
Pituitary PRL					
Concentration (μg/mg)		2.1 ± 0.31		0.2 ± 0.05[c]	0.092
Content (μg)		711 ± 96		44 ± 14[c]	0.062
Pituitary GH					
Concentration (μg/mg)		14.2 ± 0.99		9.6 ± 3.72[b]	0.674
Content (μg)		4870 ± 390		2175 ± 937[b]	0.447

[a] Values are mean ± SE.
[b] $P < 0.05$ vs SOC.
[c] $P < 0.001$ vs SOC.

culating GH did not decrease during 60-min intravenous infusion of SRIF in hypophysial stalk-transected and control gilts (Fig. 2A); however, within 15 min after infusion of SRIF was stopped, increased GH ($P < 0.05$) was seen in controls but not in hypophysial stalk-transected animals. The intravenous injection of GHRF after 60 min of a 120-min SRIF infusion caused a greater peak ($P < 0.005$) release of GH than seen in controls without SRIF (Fig. 2, B and C). TRH given during SRIF infusion was ineffective in causing GH release.

Continuous Infusion of hpGHRF(1–40)OH on GH Release. The continuous intravenous infusion of hpGHRF(1–40)OH at a rate of 0.1 μg/kg/hr did not significantly increase GH plasma concentrations (2.3 ± 0.48 ng/ml) as compared with vehicle-infused controls (1.2 ± 0.28 ng/ml) in Experiment A. In Experiment B, plasma GH averaged 2.0 ± 0.31 ng/ml in GHRF-treated gilts and 1.8 ± 0.50 ng/ml in the vehicle-treated controls.

rhGHRF Analogs on GH Release. The effects of different analogs of rhGHRF on GH release in intact prepuberal gilts are shown in Figure 3. The intravenous injection of 0.72 μg of [Nle^{27}]rhGHRF(1–29)NH_2/kg body wt caused peak ($P < 0.001$) GH release within 7.5 min compared with the basal levels seen in the diluent-treated controls (Fig. 3A). This analog given at 0.36 μg/kg body wt also increased ($P < 0.05$) GH concentrations within 7.5 min compared with controls, but lower dosages were without effect. [N-α-Me-Tyr^1, Nle^{27}]rhGHRF(1–29)NH_2 given at a dosage of 0.36 μg/kg body wt caused peak GH release ($P < 0.05$) within 15 min compared with diluent-treated controls (Fig. 3B). Although the 0.72-μg/kg body wt dosage of this hormone increased GH release ($P < 0.05$) compared with controls, the peak was lower but GH remained elevated longer compared with gilts given 0.36 μg/kg body wt. Likewise, hormone dosages of 0.18 and 0.045 μg/kg body wt increased ($P < 0.05$) GH concentrations compared with diluent-treated controls. [Nle^{27}]rhGHRF(1–32)NH_2 at dosages ranging from 0.05 to 0.80 μg/kg body wt were ineffective in causing significant increases in plasma GH concentrations (Fig. 3C).

PRL and GH Peripheral Plasma and Pituitary Concentrations in Hypophysial Stalk-Transected and Control Gilts. PRL concentrations in peripheral plasma during the 105 min after induction of anesthesia by thiamylal sodium, endotracheal intubation and maintenance of anesthesia by halothane and oxygen increased 5-fold in hypophysial stalk-transected gilts but less than 2-fold in control gilts ($P < 0.01$, Fig. 4). Soon after induction of anesthesia, GH increased abruptly to peak levels but then declined steadily throughout anesthesia in both groups. On the morning of Day 8, basal PRL concentrations were greater ($P < 0.05$) in hypophysial stalk-transected gilts than in control gilts, whereas GH was similar in both groups. Within 5 min after intravenous injection of TRH, PRL peaked ($P < 0.001$) in hypophysial stalk-transected and control gilts and then steadily decreased. GH peaked ($P < 0.05$) within 5 min after TRH injection, with the response during 60 min being greater ($P < 0.05$) in hypophysial stalk-transected than control gilts. During the afternoon of Day 8, arginine infusion did not change the consistently greater PRL concentrations in the hypophysial stalk-transected animals compared with control animals. In contrast, GH increased ($P < 0.05$) during arginine infusion in control but not hypophysial stalk-transected gilts. On Day 9, anesthesia did not increase PRL and GH levels in these animals.

α-MT and HAL on PRL and GH Release. PRL concentrations increased ($P < 0.05$) 75 min after intravenous injection of α-MT in hypophysial stalk-transected gilts, whereas they remained unchanged in controls (Fig. 5A). Circulating GH was unaffected by α-MT in either hypophysial stalk-transected or control animals (Fig. 5B). HAL caused an abrupt increase ($P < 0.05$) of PRL in controls which remained elevated

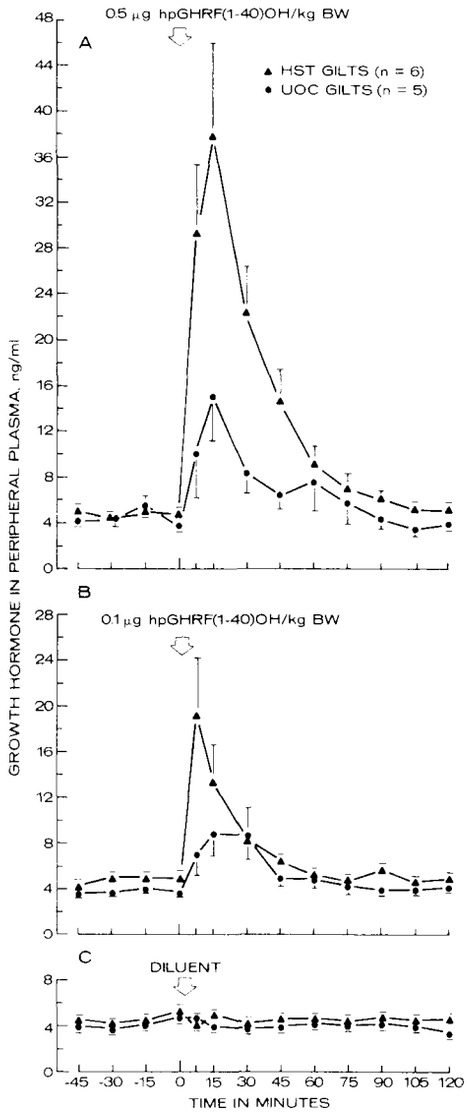

Figure 1. Effect of intravenous injection of hpGHRF(1–40)OH (A and B) or diluent (C) on GH concentrations in peripheral plasma of prepuberal hypophysial stalk-transected and unoperated control gilts. Number of gilts in each group is indicated in parentheses. Values are mean ± SE.

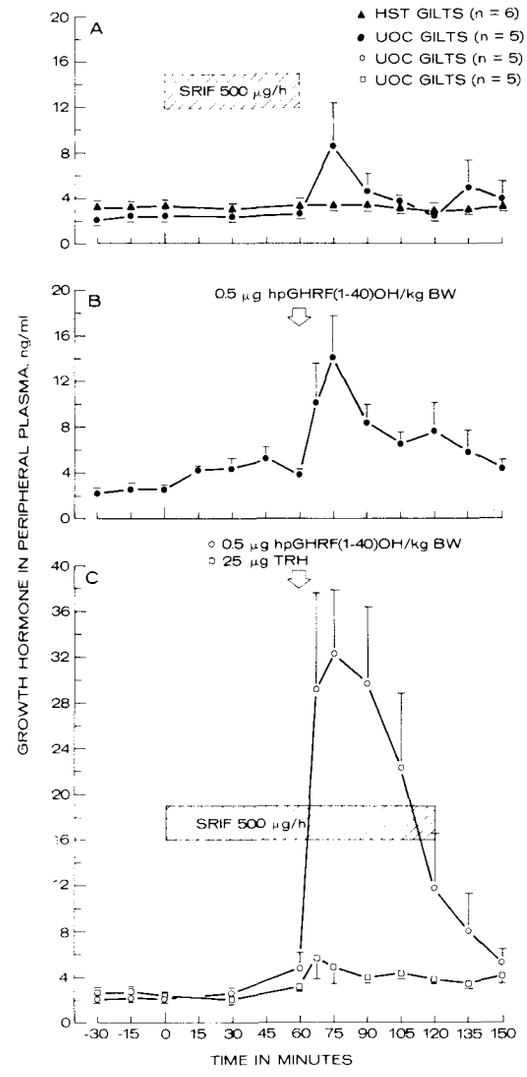

Figure 2. Effect of continuous intravenous infusion of SRIF for 60 min (A) on GH concentrations in peripheral plasma of prepuberal hypophysial stalk-transected and unoperated control gilts. (B) GH secretion in unoperated control gilts given hpGHRF(1–40)OH in the absence of SRIF. (C) GH secretion in unoperated control gilts given hpGHRF(1–40) or TRH during continuous intravenous infusion of SRIF for 120 min. Number of gilts in each group is indicated in parentheses. Values are mean ± SE.

throughout 120 min (Fig. 5C). In contrast, PRL was unchanged by HAL treatment of hypophysial stalk-transected gilts throughout the blood sampling period. GH concentrations were unaffected by HAL in hypophysial stalk-transected and control gilts (Fig. 5D).

Discussion

The results clearly indicate that hypophysial stalk-transected gilts remain acutely responsive to GHRF. A dose-dependent response to GHRF resulted in a 2-fold greater GH peak release in hypophysial stalk-transected gilts than in control gilts. Although bolus intravenous injection of GHRF caused abrupt GH release in hypophysial stalk-transected and control gilts (Fig. 1A), the continuous infusion of it was ineffective in increasing circulating GH. This greater response to GHRF occurred in hypophysial stalk-transected gilts with pituitary gland weight reduced 38%, GH concentration reduced 32%, and thus total GH content reduced 55%. Therefore, the magnified response of GH peak release within 15 min after injection of GHRF indicates accumulation of releasable pituitary GH or increased responsiveness or numbers of GHRF receptors. Comparatively, pigs are much less responsive to GHRF than cattle (e.g., 0.5 vs 0.03 μg GHRF/kg body wt) (15).

The isolated pituitary gland in hypophysial stalk-transected gilts remains viable with an adequate blood supply, as indicated by acute responsiveness to exogenous GHRF and TRH and by postmortem examina-

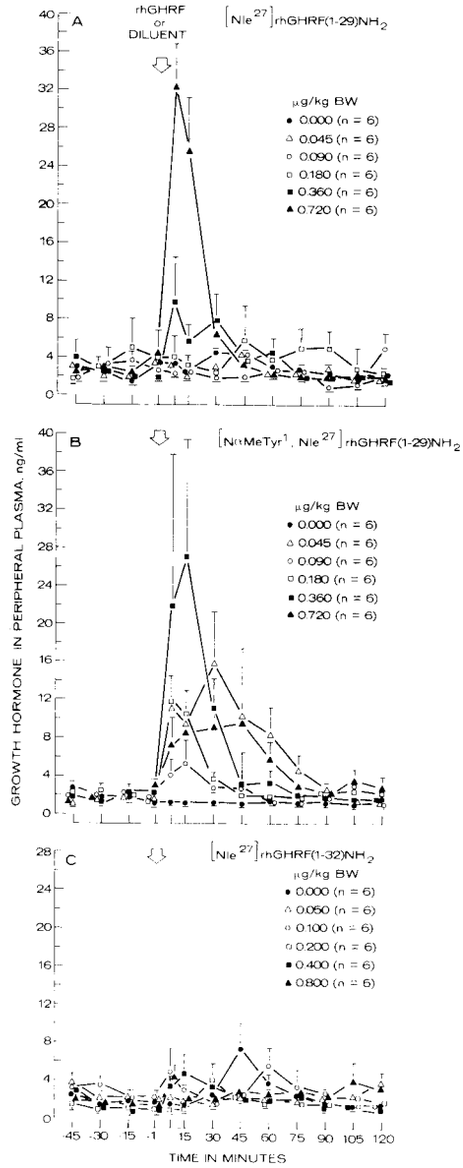

Figure 3. Effect of [Nle^{27}]rhGHRF(1–29)NH_2, [N-α-Me-Tyr^1, Nle^{27}] rhGHRF(1–29)NH_2, [Nle^{27}]rhGHRF(1–32)NH_2, and diluent on GH concentrations in peripheral plasma of prepuberal gilts. Symbols denote treatment dosages, and number of gilts in each group is indicated in parentheses. Values are mean ± SE.

tion revealing normal adenohypophysial cells. Its integrity was substantiated further by release of luteinizing hormone (LH) in response to LH-releasing hormone on Day 9. LH increased from 0.2 ± 0.1 to 7.0 ± 1.7 ng/ml at 15 min after LHRH in hypophysial stalk-transected gilts compared with an increase from 0.2 ± 0.1 to 4.5 ± 0.9 ng/ml in controls (unpublished observations). Thus, the smaller pituitary gland, which is isolated from hypothalamic input, continues to produce GH, PRL, and LH and can release large amounts of these hormones in response to specific releasing factors, such as GHRF, TRH, and LH-releasing hormone. GHRF induces GH secretion in rats that have had the *in situ* pituitary gland autotransplanted under the kidney capsule (28). In spite of similar basal GH plasma concentrations, rats with an *in situ* pituitary and bearing pituitary grafts release greater amounts of GH in response to GHRF (29).

During SRIF infusion, plasma GH remained at basal concentrations in both hypophysial stalk-transected and control gilts, but GH increased only in the controls immediately after SRIF infusion was stopped. Releasable GH likely accumulated during SRIF infusion. This idea is reinforced by the greater than 2-fold peak release of GH in controls during SRIF infusion in response to GHRF as compared with animals receiving only GHRF. Thus, the controls given SRIF and the hypophysial stalk-transected gilts responded with a similar magnitude of GH release to a given dosage of GHRF. It seems that, under different experimental conditions for hypophysial stalk-transected and control gilts, GH accumulated in the pituitary as a result of a lack of endogenous hypothalamic GHRF in hypophysial stalk-transected animals and a suppression of GHRF release by exogenous SRIF in the controls. Alternatively, the acute GH effects in control gilts at the level of GHRF receptors are also a possibility. Our observations support those of Sugihara *et al.* (30) who concluded that GHRF was necessary in intact female rats for the pituitary gland to respond with GH release immediately after infusion of SRIF. A third possibility is that because the pituitary is no longer under endogenous SRIF and GHRF regulation after HST in the pig, GH secretion is no longer affected by other hypothalamic factors such as gastrin-releasing peptide. Gastrin-releasing peptide is a potent inhibitor of GH release that blocks basal and spontaneous GH secretion as well as stimulates SRIF release (31–33).

It is clear that two analogs of rhGHRF, [Nle^{27}] rhGHRF(1–29)NH_2 and [N-α-Me-Tyr^1, Nle^{27}]rhGHRF(1–29)NH_2, were highly effective in causing an immediate peak release of GH in intact prepuberal gilts. These were not tested in hypophysial stalk-transected animals, but [Nle^{27}]rhGHRF(1–29)NH_2 stimulated GH secretion both in hypophysial stalk-transected as well as SOC calves (15).

Arginine infusion, a traditional provocative test of GH secretion, stimulated GH in controls but not in hypophysial stalk-transected pigs. The action of arginine on porcine GH secretion is through higher centers requiring intact connections of the hypothalamo-hypophysial axis, whereas TRH can act directly on the pituitary to stimulate porcine GH release. TRH-induced GH release in the pig was modest compared with GH responses to GHRF; differential responses to these releasing factors have been reported in studies with other species (13).

The consistently greater PRL secretion seen from the day of surgery throughout Day 9 in hypophysial

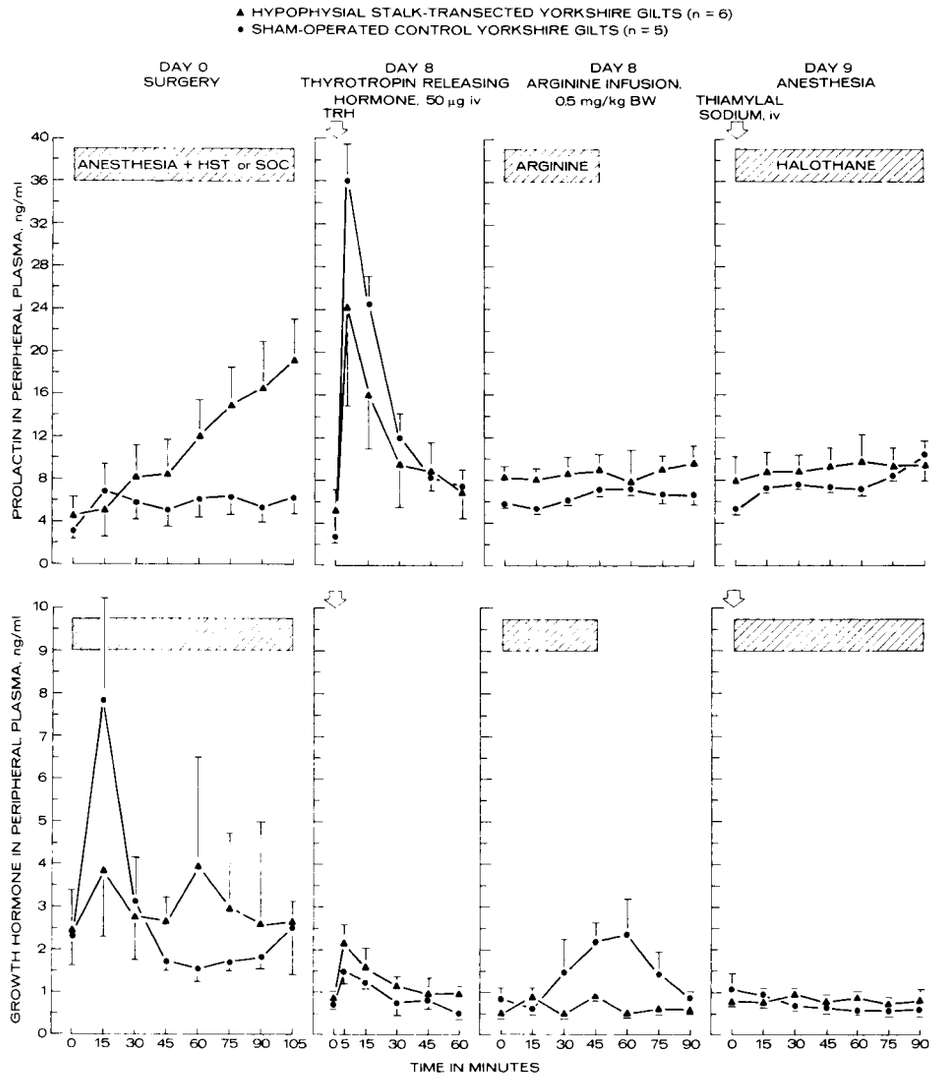

Figure 4. PRL and GH concentrations in peripheral plasma before and after HST and SOC of ovariectomized gilts. Animals were subjected to anesthesia (induction with intravenous injection of thiamylal sodium and maintained with halothane and O_2) and HST or SOC on Day 0, intravenous injection of 50 µg of TRH on the morning of Day 8, and arginine infused intravenously for 45 min on the afternoon of Day 8. On the morning of Day 9, animals were subjected to the same anesthesia regimen for 90 min and killed to confirm HST and to recover pituitary tissue. Open arrow indicates time of TRH or thiamylal sodium injection. Number of gilts is indicated in parentheses. Values are mean ± SE.

stalk-transected gilts compared with control gilts confirms previous findings (1, 34). Because pituitary PRL concentrations were less in hypophysial stalk-transected gilts than control gilts, it may be that PRL synthesis lags behind secretion. Indeed, in long-term hypophysial stalk-transected gilts PRL decreases to blood concentrations similar to those of stalk-intact animals (35). TRH caused acute PRL release in these hypophysial stalk-transected gilts 8 days after surgery, which was a response similar to that seen in hypophysial stalk-transected heifers (10). TRH acts directly on the pituitary gland to increase PRL secretion even in the presence of PRL-inhibiting factors (7). The response to TRH indicates that the porcine pituitary remains capable of secreting large quantities of PRL after HST. Other PRF of hypothalamic (i.e., oxytocin, vasoactive intestinal peptide) and posterior pituitary origin have been suggested as physiologic regulators of PRL secretion (36, 37).

α-MT inhibits catecholamine synthesis in the hypothalamus by blocking the activity of tyrosine hydroxylase (38, 39). In intact gilts, α-MT at a dose of 10 mg/kg body wt was ineffective in abruptly increasing circulating PRL, which suggests that this high dosage of α-MT cannot sufficiently decrease or alter the turnover rate of the tyrosine hydroxylase pool. A transient increase in circulating PRL occurred in hypophysial stalk-transected gilts 1 hr after α-MT injection, and this delay suggests that the action of α-MT on PRL secretion was not direct but involved intermediate responses. In contrast, the same dosage of α-MT in intact heifers caused an abrupt 16-fold increase in plasma PRL concentra-

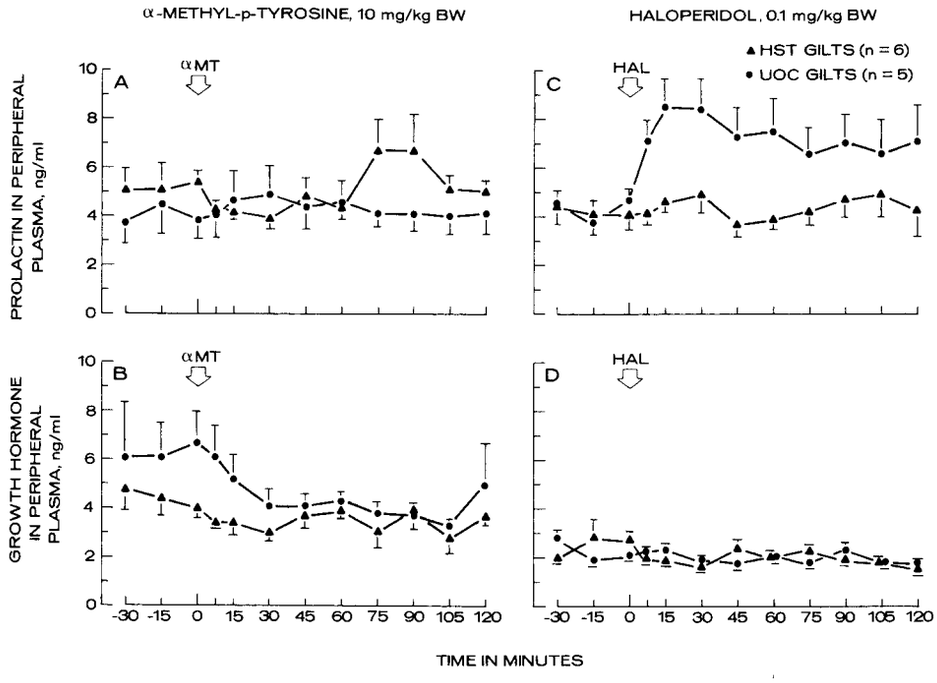

Figure 5. PRL and GH concentrations in peripheral plasma of prepuberal hypophysial stalk-transected and control gilts before and after intravenous injection of α-MT and HAL. Number of gilts is indicated in parentheses. Values are mean ± SE.

tions, which remained elevated 4 hr, but few animals responded after HST (10, 17). Other dopamine antagonists, such as chlorpromazine and reserpine, did not increase PRL blood concentrations even though they produced moderate sedation in lactating sows (40).

HAL is a neuroleptic drug that blocks dopamine receptors on lactotrophs in the anterior pituitary (38, 39). HAL caused an abrupt rise in PRL plasma concentration in controls that was maintained 2 hr whereas PRL remained unchanged in hypophysial stalk-transected gilts, a result similar to that seen in hypophysial stalk-transected monkeys (38). The marked HAL-induced PRL rise in intact gilts indicates that the pituitary lactotrophs are under significant dopamine inhibition. Since HAL was ineffective in hypophysial stalk-transected gilts, it is suggested that dopamine is not acting on the lactotrophs due to the severed stalk or the lactotrophs are unable to respond to dopamine, possibly due to down-regulation of receptors. The differences observed between the responses of hypophysial stalk-transected and intact gilts to HAL indicate that dopamine plays an important role in hypothalamic regulation of PRL secretion in this species. In intact heifers, dose-dependent changes in response to HAL administrations were found in both the rate and level of peak PRL secretion, but the response also was abolished after HST (10, 17).

The results from this study indicate that after HST the isolated pituitary remains acutely responsive to releasing and inhibiting factors in the pig. HST liberates tonic inhibition of PRL secretion, and both GHRF and SRIF can modulate GH release in this species. The results reported here for the pig indicate comparative differences in the basal regulation of GH and comparative similarities in PRL secretion to that seen in other farm and laboratory animals, but marked differences in the magnitude of hormone released in response to hypothalamic releasing and inhibiting hormones.

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