

Editorial: When the Light Turns Blue

When the light is green you g(r)o(w).
When the light is red you stop.
But what do you do
When the light turns blue
With orange and lavender spots?¹

My son, Jonathan, has recently had his thirteenth birthday and instantly became a certified teenager. We have yet to witness the horrors of adolescent behavior. On the contrary, out of the goodness of his heart, Jonathan hired me as an unpaid research assistant to perform an important scientific experiment. Every couple of weeks he takes me downstairs and there he stands "at attention" by the old door. My task consists of leveling a book on his head and marking his height. To my astonishment, every notch gets scratched about half a centimeter above the earlier one. The urge to publish this fascinating case report is tempered by the sober realization that the same thing is happening to every teenager around the world. Jonathan has hit the growth spurt. His GH secretion is in full swing, pouring out at high amplitude pulses and increasing his insulin-like growth factor I (IGF-I) into an acromegalic range. However, thousands of children who lack the ability to secrete GH may never reach normal height.

A couple of years down the road things will quiet down, Jonathan's growth will stop, and his GH will start its inexorable decline to the low levels of his "fortysomething" unpaid research assistant. But, in contrast to Jonathan's short-lived state of functional, pubertal gigantism, about 40 people out of every million develop GH-producing pituitary tumors and have persistent and unrestricted growth.

What is the intricate wiring of this fine-tuned machinery that allows the rest of us to rev up our growth rate for a couple of years at the green light of adolescence and then bring it down to a screeching halt as soon as the red light of adulthood turns on?

As far as we know, some unidentified signals from unknown parts of the brain come to the hypothalamus and regulate the activities of GH-releasing hormone (GHRH) and somatostatin (SRIF) neurons. GHRH exerts mitogenic effect upon pituitary somatotrophs and also increases GH synthesis through an intracellular cascade involving G_s protein, adenylyl cyclase, protein kinase A, and transcription factors CREB and Pit-1. Abnormalities in this cascade may cause either somatotroph hypoplasia or a formation of somatotropinoma. Recently, a family of GH-releasing peptides or their nonpeptidic analogs exerting powerful action on GH release has been synthesized. The identification of specific

hypothalamic and pituitary binding sites for these GH secretagogues (1) indicates the existence of a novel endogenous GH-releasing hormone. Both GHRH and SRIF are secreted in pulses. The possibility that these secretory pulses are reciprocal, 180 degrees out of phase, has been suggested by indirect pharmacological approaches (2) but could not be confirmed by direct measurements in the pituitary-portal circulation in sheep (3). Whether endogenous GH secretagogue (GHS) is also secreted in pulses is unknown, but it is likely to be so. The importance of GH pulsatility in the regulation of growth (4) has been demonstrated. The hypothalamic interplay between GHRH, SRIF, and (likely) endogenous GHS as well as the mechanisms of the negative feedback by GH and IGF-I are not well understood. Hidden somewhere within this complex network are the "on" and "off" switches for both the generation of the Morse code of GH pulsatility and for the long-term augmentation of GH secretion during puberty and its subsequent extinction toward old age.

In this issue, four manuscripts address different aspects of GH physiology. Morpurgo *et al.* (5) extended their earlier studies aimed at elucidating the mechanisms of somatotroph differentiation in chickens. They have shown that the addition of serum from 16-day-old embryos (when somatotrophs are well-defined and functionally competent) to the pituitary cultures obtained from 12-day embryos (when these cells are normally dormant) stimulated somatotroph development. Using steroid hormone receptor antagonists and antisera, they have shown that the circulating differentiating agent was corticosterone. The effect of corticosterone appears to be postmitotic because it continued to work in the presence of a mitotic inhibitor and because its effect was not accompanied by [³H]-thymidine accumulation. Moreover, the effect of corticosterone on either somatotroph differentiation or the ability to synthesize and release GH did not require GHRH. We have grown so accustomed to viewing GHRH and transcription factors CREB and Pit-1 as crucial for somatotroph development, that the involvement of corticosterone comes as a revelation. Perhaps classical endocrinology, as study of circulating hormones, still holds some surprises. One can almost predict the future questions. What is the interplay of corticosterone with transcription factors? A suggestion that glucocorticoids may interact with Pit-1 in lactotrophs has already been made (6). Would inhibitors of steroidogenesis (such as o'p'DDD) cause dwarfism in chickens? Is the effect of corticosterone species-specific? The authors suggest that the same process may be operative in rats, a model that is biologically closer to humans than the heraldic bird of Colonel Sanders. Is this effect limited to a narrow time window during embryogenesis? High does of glucocorticoids powerfully release GH in humans, but in contrast to all other secretagogues, there is a 2- to 3-h delay of action (7). Do they reawaken some dormant somatotrophs? No matter what the answers may be, the somatotroph-differentiating effect of

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¹ With apologies to Shel Silverstein.

glucocorticoids must only be modulating and secondary in importance to the GHRH-cAMP-CREB-Pit-1 cascade. Indeed, isolated GHRH deficiency or Pit-1 mutations do not affect normal corticotroph development and glucocorticoid production. Nevertheless, pituitary somatotrophs in these conditions remain atrophic and nonfunctional. Moreover, selective neonatal GHRH deprivation, although transient in nature, leaves somatotrophs permanently handicapped and unable to respond to GHRH during adulthood (8).

The importance of GHRH in somatotroph growth and function is nicely demonstrated by the paper by Kovacs *et al.* (9). They looked at the effects of a novel GHRH receptor antagonist, MZ-5-156, in transgenic mice expressing the human GHRH gene. In these animals, the entire body becomes a huge arcuate nucleus producing industrial doses of GHRH. As a result, plasma GHRH rises from undetectable up to almost 1 $\mu\text{g}/\text{ml}$, somatotrophs undergo hyperplasia that subsequently transforms into an adenoma, and GH increases many fold. The GHRH antagonist at doses of 50–200 μg decreased GH in these animals both acutely and chronically. After 3 days of treatment, an almost 40% inhibition of plasma IGF-I was observed. These results tie very well with the recent data showing that another GHRH antagonist (*N*-Ac-Tyr¹, *D*-Arg²) GHRH-(1–29) NH₂ suppressed plasma GH in a patient with acromegaly due to ectopic GHRH secretion (10). However, the present antagonist, MZ-5-156, is much more potent and appears to be longer acting, providing a more convenient tool to probe GHRH physiology. The animals used in this study were 3 months old. At that time, their pituitaries show purely hyperplastic changes. Will this antagonist be equally successful in 12- to 24-month-old mice whose hyperplasia has already converted into an adenoma? This would be a nice way to probe whether adenomatous transformation leads to functional autonomy. Human somatotroph adenomas, as opposed to other pituitary tumors or normal pituitaries, contain both GHRH messenger RNA (mRNA) and GHRH (11). Does endogenous GHRH (autocrine, paracrine, or hypothalamic) play a role in the pathogenesis of acromegaly? GHRH antagonists provide a novel and powerful tool with which to settle this old question.

In the third paper, Pellegrini *et al.* (12) studied the mechanisms of the hypothalamic feedback of GH upon GHRH and SRIF in two distinctly different models of rat dwarfism. The DW animals have a primary pituitary defect and their hypothalamic GHRH is high, whereas Tgr rats, expressing hGH transgene in the hypothalamus, have low hypothalamic GHRH. In both models, implantation of the tumor-derived, GH-producing GC cells increased plasma GH levels and stimulated somatic growth. High circulating GH levels suppressed high GHRH in the DW rats but were without effect in the Tgr model. The low SRIF levels in the DW rats was increased by GC implants, but the already high SRIF in the Tgr animals was not. These outcomes were fully expected. Similar data were obtained in these models using chronic GH administration. The advantage of the CG-implanted model lies in its ability to provide constantly high GH levels, but physiological GH is never constant. In rats, plasma GH rises from undetectable interpulse levels to 200–300 ng/ml over several minutes and then goes down within an hour or two reflecting the half-life of the hormone. I would be more

interested to know how the acute GH discharge terminates its own release. For this reason, IV boluses of GH to DW rats may be more revealing. Zeitler *et al.* (13) have already shown that the hypothalamic content of GHRH and SRIF mRNAs fluctuates acutely, concordant to plasma GH pulsatility. However, neither hypothalamic mRNA nor peptide contents tell us much about the actual secretion of GHRH and SRIF into the pituitary-portal circulation. It is this process that ultimately determines the initiation and the extinction of GH pulses. The models described by the authors will unlikely be very useful in understanding the neuroendocrine pathophysiology of dwarfism and especially, acromegaly. Subcutaneous implantation of GH producing cells is a far cry from spontaneous development of pituitary tumors, just as feeding high doses of L-thyroxine does not tell us much about the pathogenesis of Graves' disease. I still put my money on Kovacs and her GHRH antagonist.

The fourth paper is an important addition to the evolving knowledge about GH-secretagogue, a still unidentified third regulator of GH secretion. The existence of an endogenous GHS was postulated by the biological effects of GH-releasing peptides and by the demonstration of the presence of their specific receptor in the pituitary and the hypothalamus (1). Bennett *et al.* (14) strengthen the case by showing that GHS receptors (GHS-R) in the hypothalamus can be regulated by circulating GH in a physiologically meaningful fashion: they are high in dw/dw rats and decline after GH administration. The location of the GHS-R in the arcuate nucleus corresponded nicely to the location of the GHRH neurons, suggesting interplay between the two hormones. Indeed, it has already been shown that administration of the GHRP-6 releases GHRH and GHRH antagonist and GHRH antiserum block GH responses to GHRP-6. The study by Bennett *et al.* (14), like all good studies, raises a whole new series of questions. If the infusion of GHRP-6, as they have shown, does not down-regulate its own receptors, but at the same time releases GHRH, what accounts for homologous desensitization of GH to bolus GHRP-6 but heightened response to GHRH (15)? If high circulating GH suppresses GHS-R, why do all patients with acromegaly exhibit major GH discharges to GHRP but only some of them respond to GHRH (16)? Clearly, hypothalamic GHS-R content is not the only determinant of the GH sensitivity to GHS. What is the potential role of the GHS receptors found by the authors in the hippocampus? Are they involved in the mediation of the known sleep and eating-promoting effects of GHS?

High GH secretion, eating and sleeping . . . Come to think of it, that's an adolescent in a nutshell.

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