INTRODUCTION

Anorexia nervosa (AN) is a serious eating disorder characterized by self-starvation and extreme weight loss, resulting in a body mass index (BMI) of less than 17.5 kg/m$^2$ (American Psychiatric Association [APA], 2000; Dartmouth College Health Services [DCHS], 1999; Eating Disorders Awareness and Prevention [EDAP], Inc., 2000; Stoving et al., 1999). The psychological disturbances behind the disorder include a refusal to maintain an appropriate weight for the height, age, body type, and activity level of the individual. Patients also suffer from an unreasonable fear of being fat, an obsession with body weight and image, and a constant preoccupation with food and weight (APA, 2000; DCHS, 1999; EDAP, 2000). These features lead to self-denial of nourishment, compulsive exercise, low self-esteem, and withdrawal from friends and family (APA, 2000; DCHS, 1999; EDAP, 2000). Other health consequences develop as well, such as amenorrhea in women, digestive problems, hair loss, sensitivity to cold, slow heart rate, low blood pressure, decreased bone density, muscle loss, and dehydration (DCHS, 1999; EDAP, 2000).

Approximately 90-95% of the people suffering from AN are female (EDAP, 2000). In the United States, 1-2% of girls and women are afflicted with the disease (EDAP, 2000). AN is the most common psychological disorder in adolescent girls and has one of the top death rates of any mental disorder (Stoving et al., 1999; EDAP, 2000).

Several studies have been performed on the endocrine responses in AN. A deeply investigated area is the elevated basal and pulsatile secretory levels of growth hormone (GH) in individuals with AN. The release of GH from the pituitary is mediated by the secretion of growth hormone releasing hormone (GHRH) and somatostatin from the hypothalamus: GHRH...
stimulates secretion while somatostatin inhibits secretion. Acetylcholine, a neurotransmitter, indirectly stimulates GHRH secretion by inhibiting somatostatin release (Ghigo et al., 1994). A continual subject of investigation has been whether the increased basal and pulsatile secretion of GH is due to increased GHRH discharges or reduced inhibition by somatostatin. E. Ghigo et al. (1994) performed a study on the effects of the somatostatin inhibitor, the amino acid arginine, on GH levels in patients with AN. Arginine was found to enhance GH secretion, suggesting that somatostatinergic inhibition plays a role in the augmented GH levels seen in AN and is probably responsible for the increase in basal secretion of GH (Ghigo et al., 1994). M. Scacchi et al. (1997) demonstrated that the high nocturnal secretion of GH found in AN is partially caused by an increase in GH pulse. High GH pulse frequency indicates that AN triggers an elevation in GHRH discharges (Scacchi et al., 1997).

However, there is no clear evidence about the role of irregular GH secretion which is not detectable through pulsatility measurements in AN. In the current study, Stoving et al. investigated spontaneous GH secretion in individuals with AN using two novel techniques: deconvolution analysis and approximate entropy (ApEN). Deconvolution analysis permits evaluation of GH secretion minus the impact of hormone kinetics and ApEN allows measurement of irregular GH concentration patterns (Stoving et al., 1999).

**SUBJECTS AND METHODS**

The subjects in this study consisted of 8 women with AN and 11 healthy women as normal controls. The subjects were all in their mid-20s, had no significant weight changes during the week before the study, and were not taking any medication. Three days prior to the study, all subjects were admitted to the hospital and put on a standardized diet. The patients were required to stay in bed for at least 1 hour after meals and no exercise was allowed except for minimal walking. The controls were determined to be healthy by a physical examination and all had a regular menstrual cycle. Laboratory tests revealed normal serum levels of luteinizing hormone (LH), follicle-stimulating hormone (FSH), testosterone (total T3), fasting blood glucose, and insulin.

Blood was sampled continuously from the subjects for 24 hours and vials were replaced every 20 minutes. Serum levels of GH, insulin-like growth factor I (IGF-I), IGF binding protein 1 (IGFBP-1), IGFBP-3, insulin, glucose, leptin, LH, FSH, estradiol (E2), total T3, cortisol, and urinary cortisol were determined with various immunofluorometric assays and radioimmunoassays. Deconvolution analyses and ApEN were calculated via complex statistical methods.

24 hour serum GH concentration profiles were determined for the patients with AN and the normal controls (Fig. 1). Mean serum GH concentrations were obtained by plotting serum GH concentration versus time, and the GH secretion rate and secretory parameters were obtained by deconvolution analysis.

**SUMMARY OF FINDINGS**

In AN, there was an increased GH secretion frequency, decreased interpulse intervals and increased burst mass (concentration of GH per burst). The basal GH secretion rate was increased 20-fold, whereas the pulsatile secretion rate was increased 4-fold. The GH half-life was the same in AN and the normal controls. Thus, the elevated serum GH concentrations in the women with AN were a result of increased secretion rather than delayed degradation of GH. A negative correlation existed between BMI and the 24 hour basal and pulsatile secretion rates, burst frequency, and burst mass. ApEN analysis revealed that AN patients had a much more irregular GH secretory pattern than normal controls. The diurnal pattern of serum GH concentrations in AN showed a greater amplitude and average value (mesor) compared to women without AN, but a similar clock time at which the maximum value occurred (acrophase).

AN patients had elevated 24 hour mean serum cortisol and urinary cortisol. These values were negatively correlated to BMI, and positively correlated to basal and pulsatile GH secretion and burst frequency. The cortisol secretory pattern in AN was similar to the normal controls. Analysis of the diurnal pattern showed a comparable amplitude and acrophase but an increased mesor.

Significant decreases in IGF-I, IGFBP-3,
leptin, LH, FSH, E₂, total T₃, and insulin levels were seen in the women with AN. IGFBP-1 was elevated and no difference was detected in fasting glucose concentrations. Leptin was negatively correlated only to the pulsatile GH secretion rate.

**DISCUSSION**

The researchers clearly took many factors into consideration when selecting subjects and constructing the study. On average, the AN patients were younger than the normal controls but the slight difference in ages is nonsignificant. The effects of medication and diet were controlled for by choosing subjects that had not been on medication for at least 6 months prior to the study, and by offering a standardized diet to the subjects before and during the study. The researchers understood the psychology behind AN since they eliminated the opportunity to purge by keeping the subjects in bed after each meal, and they prevented any compulsive exercise by allowing only minimal walking.

Despite these efforts to control the study conditions between the AN patients and normal controls, the standardized diet may have been a source of inaccuracy in the study. The diet that was offered to the subjects contained 7000-8000 kJ/day, which is the average diet of a normal person without AN. Since one of the characteristics of AN is self-starvation, the daily diet of an individual with AN contains much fewer kilojoules than the diet of a normal person. Studies of fasting patients have shown elevated GH secretion levels (Stoving et al., 1999). Therefore, one would expect that the inverse is true: an increased intake of kilojoules for 4 consecutive days would result in lower GH basal and pulsatile secretion and less irregular secretion in the AN patients, compared to the values that would be obtained if the patients followed their usual diet.

The 20-fold increase in the basal GH secretion rate and the 4-fold increase in the pulsatile secretion rate reveal that the 10-fold elevation in 24 hour serum GH concentrations in AN is primarily due to enhanced basal secretion. These results support findings from earlier studies on the cause of high GH concentrations in AN. Elevated basal GH levels point toward a decrease in somatostatinergic inhibition and increased pulsatile GH secretion indicate a higher frequency of hypothalamic GHRH discharges.

The negative correlation between basal and pulsatile GH secretion rates and BMI show that both basal and pulsatile GH levels are significantly influenced by weight. Therefore, the extreme weight loss in AN is a major contributing factor to the amplification of basal and pulsatile GH secretion. Previously, J. Argente et al. demonstrated that AN patients attained a normal GH secretory pattern after gaining 10% or more of their initial weight (Argente et al., 1997). In the current study, 4 of the women with AN who gained weight to a BMI of 17.3 ±

![Figure 1. 24 hour serum GH concentration profiles of the patients with anorexia nervosa and the normal controls. Reproduced with permission from the Journal of Clinical Endocrinology & Metabolism, 84, p. 2058.](image)
0.9 kg/m² after 3 months were restudied. The basal and pulsatile GH secretion rates moved towards the range of the normal controls but unfortunately, the sample size was too small for the data to be statistically significant. Further studies of recovering AN patients should be conducted to examine the changes in GH secretion as a consequence of weight gain.

A prior study of women with a normal BMI and amenorrhea due to hypothalamic dysfunction revealed a slight increase in basal GH secretion and a decrease in pulsatile secretion, despite an increase in pulse frequency (Stoving et al., 1997). Based on these findings, Stoving et al. hypothesized that 2 mechanisms influence the elevated pulsatile GH secretion in AN: the extreme reduction in weight resulting in enhanced burst mass, and the low estradiol levels causing an augmented pulse frequency. ApEN data showed that AN patients have a much greater irregular GH secretory pattern than the normal controls. It would be interesting to observe the alterations in GH concentrations and secretory behavior in obese subjects participating in a standardized weight loss program. Such a study may disclose information on the relationship between BMI and GH concentrations that would be diagnostically and prognostically valuable in the treatment of AN and obesity. Another worthy study would be the examination of changes in GH pulsatile secretion as a consequence of estrogen administration to individuals with AN to see if increasing estrogen in individuals with AN would lead to higher GH concentrations.

Thus far, most studies of the endocrine abnormalities resulting from AN have been on female subjects. Estrogen is one of the key sex hormones in females and testosterone is one of the key sex hormones in males. This parallel would lead one to expect that testosterone has an influence on GH secretion in men with AN as well. Future studies should be performed on men suffering from AN and men with a normal BMI but low testosterone levels to observe the alterations in GH concentrations and pulse frequency.

Low serum IGF-I and IGFBP-3 levels reveal a disruption in the GH-IGF-I axis in patients with AN. Normally, GH stimulates the production of IGF-I. Despite elevated levels of GH in AN, serum IGF-I concentrations are low, thus reducing IGFBP-3 levels since IGFBP-3 is the binding protein for IGF-I (Griffin & Ojeda, Eds., 2000). IGF-I usually inhibits the release of GH so enhanced GH secretion in AN may be due to the lack of negative feedback regulation to the hypothalamus and pituitary from IGF-I. The effects of the administration of IGF-I on GH concentrations in AN individuals would be an interesting study to locate the disruption in the regulation of the GH-IGF-I axis.

The elevated 24 hour mean serum and urinary cortisol in AN, but normal diurnal secretory pattern, indicate that AN may cause disturbances in the cortisol releasing hormone (CRH)-adrenocorticotropin hormone (ACTH) axis. A positive correlation exists between cortisol concentrations, and 24 hour GH basal and pulsatile secretion and burst mass. One would expect cortisol and GH levels to be negatively correlated since cortisol is involved in the direct effects of GH but the impact of high cortisol on the inhibition of GH seems to be altered in AN. Studies on the relationship between cortisol and GH in obese subjects may shed some light on this matter.

Leptin levels are low in AN. In addition, leptin is negatively correlated with pulsatile, but not basal, GH secretion. This may be evidence that leptin contributes to GHRH release, supporting the idea that BMI influences GH secretion since leptin is produced in adipose tissue the quantity of which vary with BMI. Further studies should be conducted on patients with defective leptin or leptin receptors to investigate the effects on GHRH and GH secretion.

**CONCLUSION**

Deconvolution analysis and approximate entropy indicate that the elevated basal and pulsatile GH secretion in AN is due to enhanced hypothalamic GHRH discharges and lowered inhibition by somatostatin. High GH pulse frequency and short interpulse intervals demonstrate the increase in GHRH release. Augmented basal GH levels reveal the reduced somatostatinergic inhibition. The high serum GH but low IGF-I concentrations suggest a disruption in the GH-IGF-I axis in AN. Additional studies on recovering AN patients, GH secretory behavior in
obese subjects, men with AN, administration of IGF-I to individuals with AN, and leptin may shed some light on the altered relationship between GH and IGF-I in AN.

REFERENCES


ABOUT THE AUTHOR

Ying Li is a Genetics, Cell, and Developmental Biology major. She serves as Events Chair of 2001 Class Council, an Eating Disorders Peer Advisor, a vocalist in the music ministry at Aquinas House, and an active member of her sorority. Ying has avidly pursued medicine by interning at a hospital in Hong Kong, volunteering and shadowing at DHMC, and interning at an orthopaedic clinic in New York. Next year, Ying will be attending medical school. She's not entirely sure where she's going for medical school yet but she can guarantee that it will be somewhere warmer than Hanover!