Available online at www.elixirpublishers.com (Elixir International Journal)

Hormones and Signaling

Elixir Hormo. & Signal. 84 (2015) 33805-33807

Pulsatile Pattern of Growth Hormone Secretion in Females in Elele

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ARTICLE INFO

Article history: Received: 7 February 2015; Received in revised form: 8 July 2015; Accepted: 18 July 2015;

Keywords

Pulsatile pattern, Growth hormone, Secretion.

ABSTRACT

The study was carried out to determine the variations in growth hormone secretion in females between morning and evening. Blood samples were collected from 60 females at Elele, both in the morning and evening. The samples were assayed using ELISA-based Growth hormone assay to quantitate the growth hormone concentrations of these females between morning and evening. The result showed a significant increase in growth hormone secretion in the morning $(4.70\pm0.54$ mg/ml) compared with $(2.02\pm0.23$ mg/ml) obtained in the evening (P<0.05). Females less than 20 years of age (20 subjects) had the highest secretion in the morning (4.99 ± 0.98) compared with the evening (2.35 ± 0.35) while females within 20-30 years had4.58±1.04 and1.54±0.33 for morning and evening respectively. Females within 30-40 age group had 4.53 ± 0.80 and 2.18 ± 0.50 for morning and evening respectively. The result of this study suggest that Growth hormone pulse amplitude increased significantly in the morning after the onset of a night sleep (4.70 ± 0.54) and was significantly greater than the secretion rate in the evening(2.02 ± 0.23).

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Introduction

Growth hormone (GH or HGH), also known as somatropin, is a peptide hormone of about 190 amino acids synthesized and secreted by cells called somatotrophs in the anterior pituitary that stimulates growth, cell reproduction and regeneration in humans and other animals. It is a type of mitogen which is specific only to certain kinds of cells. Growth hormone is used as a prescription drug in medicine to treat children's growth disorders and adult growth hormone deficiency. In the United States, it is only available legally from pharmacies, by prescription from a doctor. In recent years in the United States, some doctors have started to prescribe growth hormone in GHdeficient older patients (but not on healthy people) to increase vitality. While legal, the efficacy and safety of this use for HGH has not been tested in a clinical trial. At this time, HGH is still considered a very complex hormone, and many of its functions are still unknown. In its role as an anabolic agent, HGH has been abused by competitors in sports since the 1960s and it has been banned by the IOC and NCAA. Traditional urine analysis could not detect doping with HGH, so the ban was unenforceable until the early 2000s when blood tests that could distinguish between natural and artificial HGH were starting to be developed. Blood tests conducted by WADA at the 2004 Olympic Games in Athens, Greece targeted primarily HGH (Powers, 2005).

Several experimental lines of evidence strongly suggest that GH is released in a pulsatile fashion with pulses occurring every 3-4 h with the greatest release occurring during sleep (Quabbe et al. 1966). The amplitude and frequency of GH pulses is dependent on age, being increased during puberty and declining with age (Finkelstein et al, 1972; Rudman et al. 1981; Martha et al. 1989). Sleep, stress, exercise, and post-prandial decline in blood glucose concentration act as stimuli to GH secretion, although it is likely that most episodes of release are spontaneous (Martin, 1976). Growth hormone secretion is sexually dimorphic. When women are compared to men, daynight differences are less prominent, interpulse baseline is higher, and the secretion pattern shows less orderliness.

Findings indicated that 24-h GH secretion varies throughout the menstrual cycle with higher pulse frequency and an enhanced day-night variation during the follicular phase of the cycle (Faria et al, 1992). Studies in women and late adolescent females have been limited either by the lack of control for the phase of the menstrual cycle, by a very small number of subjects or insufficient sampling to establish a 24-h profile. Variations of GH secretion during the menstrual cycle have been suggested, while others have found no differences between the follicular and luteal phases of the cycle. Other studies have only focused on differences of GH between the follicular and the periovulatory phases of the menstrual cycle, documenting a clear increase of GH pulses at mid-cycle (Faria et al, 1992).

Growth hormone concentrations are affected by weight and are lower in overweight and obese individuals. It is also known that GH decreases with ageing thus it is important to adjust for BMI and age and/or account for them in the selection of participants as was performed in study by Rose et al, (1991).

The aim of this research is to determine the pulsatile pattern of growth hormone secretion between morning and evening in females at Elele in Rivers State.

Methods and Materials

Test Samples

The blood samples were obtained from 60 females in Elele between the ages of 14-40 using venepuncture into plain bottles; the serum was separated, labelled and stored in a refrigerator.

Reagents Commercially prepared Growth hormone reagents were obtained from Diagnostics automation incorporated Calabasas, USA.

Determination of Growth Hormone (GH) Level

Principle: The HGH quantitative test kit is based on the principle of a solid phase enzyme-linked immunosorbent assay. The assay system utilizes a polyclonal anti-HGH antibody for solid phase (microtiter well) immobilization and a mouse monoclonal anti-ferritin antibody in the antibody –enzyme (horseradish-peroxidase) conjugate solution.



Table 1. Variations in growth hormone secretion in female							
Parameter	Morning	Evening	Т	P value			
Growth hormone	4.70±0.54ng/ml	2.02±0.23ng/ml	5.718	0.000			

Table 2. Variations in growth hormone secretion in different age group

Age group (Years)	Morning(ng/ml)	Evening(ng/ml)	Т	Р
< 20	4.99±0.98	2.35±0.35	2.956	0.008
20-30	4.58±1.04	1.54±0.33	3.355	0.003
31-40	4.53±0.80	2.18±0.50	3.667	0.002

The test sample is allowed to react simultaneously with the antibodies, resulting in HGH molecules being sandwiched between the solid phase and enzyme-linked antibodies. After 60 minute incubation at room temperature, the wells are washed with water to remove unbound labeled antibodies. The color development is stopped with the addition of 2N HCL, and the color is changed to yellow and measured spectro photomectrically at 450nm. The concentration of HGH is directly proportional to the color intensity of the test sample (Enzyme Linked Immunosorbent Assay).

Procedure`: The desired number of coated wells was secured in the holder. 50µl of the standard, specimens and controls were carefully dispensed into the appropriate well. 100µl of the Enzyme Conjugate Reagent was dispensed into each well respectively. The contents of each well was thoroughly mixed for 30 seconds, and incubated at room temperature (18-22°C) for 60 minutes. The incubation mixture was removed by flicking plate content into a waste container, and the microtiter wells were rinsed and flicked 5 times with washing buffer (IX). The wells were stroked sharply onto absorbent paper or paper towels to remove all residual water droplets. 100µl of TMB (Tetramethyl benzidine) substrate was dispensed into each well, gently mixed for 5 seconds and incubated at room temperature in the dark for 20 minutes. The reaction was stopped by adding 100µl of Stop Solution to each well and gently mixed for 30 seconds. (It is important to make sure that all the blue color changes to yellow color completely).Optical density was read at 450nm with a microtiter reader within 30 minutes.

Statistical analysis

The biochemical data were subjected to some statistical analysis as the Mean (X), standard deviation (SD), standard error of mean (SEM) and student's t-test using Statistical Package for Social Sciences (SPSS) version 17. The results were expressed in Mean \pm standard error of mean (SEM)

Result The result of the study showed that there was a significant increase (P <0.05) in female growth hormone secretion in the morning $(4.70\pm0.54$ mg/ml) compared with $(2.02\pm0.23$ mg/ml)

morning (4.70±0.54ng/ml) compared with (2.02±0.23ng/ml) obtained in the evening as shown in table 1 below. In the Table 2 below, there is significant high secretion of

growth hormone in the morning 4.99 ± 0.98 compared with 2.35 ± 0.35 mg/ml at age group < 20 years. Also at age group 20-30 years the morning GH secretory of 4.58 ± 1.04 was significant different from 1.54 ± 0.38 in the evening.

Discussion

Growth hormone is a peptide hormone that is responsible for cell growth, cell reproduction and regeneration in humans and animals. In this study, 60 female subjects had their GH determined to ascertain the difference in GH secretion between morning and evening for different age groups. Females within the age of 14-40years who are living within Elele community had their growth hormone evaluated. Plasma growth hormone was determined in the morning and evening.

The result of the study showed that growth hormone concentration in females was higher in the morning compared with the evening suggestive of high secretion of growth hormone in the morning. This is similar to report of Takahashi et al, (1968). Growth hormone (GH) is secreted in a pulsatile fashion from the anterior pituitary gland. Several experimental lines of evidence strongly suggest that GH is released in a pulsatile fashion with pulses occurring every 3-4 h with the greatest release occurring during sleep (Quabbe et al. 1966). The greatest secretion of GH occurs at night during sleep (Takahashi et al. 1968). Slow-wave sleep is frequently associated with GH release, but individual patterns of GH secretion during sleep show considerable heterogeneity (Adlard et al, 1987) and the time of sleep onset, rather than of Slow-wave sleep is more closely related to the nocturnal rise in GH secretion (Born et al, 1988). A growth hormone secretory peak occurred in the morning samples compared with the evening samples as shown in this study. Feedback mechanisms are effective on different stages of GH secretion. GH itself as well as GHRH and somatostatin can be considered to be effective on short term feedback. Hypophyseal GH, while inhibiting GHRH secretion can stimulate somatostatin secretion. GHRH has a negative feedback effect on its own stimulatory action. Low dose GHRH given to the cerebral ventricles results in the inhibition of GH release. GHRH also causes inhibition by stimulating somatostatin secretion which has the ability to suppress its own neuronal release as well as inhibiting the release of GHRH (Rosenbloom and Connor, 2007).

The result further showed that there was a significant increase in young females (<20 years) compared with those above 20 years of age whose GH secretion is significantly higher compared with those females above the age of 30 indicating that growth hormone secretion declines with age. The mean number of growth hormone secretion observed during the sampling period was significantly higher in females below the age of 20 (4.99±0.98) compared with females within 20 to 30 years of age (4.58 ± 1.04) and lesser in females within the age of 31 to 40. This is similar to Rose et al, (1991), Finkelstein et al, (1972), Rudman et al, (1981), Martha et al, (1989). A number of factors are known to affect GH secretion, such as age, sex, diet, exercise, stress, and other hormones. Young adolescents secrete GH at the rate of about 700 µg/day, while healthy adults secrete GH at the rate of about 400 µg/day. Sleep deprivation generally suppresses GH release, particularly after early adulthood (Mullinton et al, 1996).

Conclusion

In conclusion, this experiment strongly suggests that female growth hormone secretion increases in the morning compared with evening as a result of the stimulatory effect of sleep in the secretion of growth hormone.

The result also showed that secretion of growth hormone declines with age as highest secretions were obtained in young females than in older females. This may account for the relatively body growth stasis observed in young females.

Recommendation

Based on this study and observation, it is therefore recommended that individuals (adults and Children) generally should always avoid interruption and deprivation of sleep as this generally suppresses growth hormone release.

Moreover, children should never be deprived of sleep, as to ensure sufficient growth hormone release in them thus maintaining adequate growth and development.

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