ISSN: 0976-3031

Available Online at http://www.recentscientific.com

International Journal of Recent Scientific Research
Vol. 7, Issue, 7, pp. 12511-12514, July, 2016

International Journal of Recent Scientific Research

Research Article

GHRELIN LEVEL IN BOYS WITH CONSTITUTIONAL DELAY IN GROWTH AND PUBERTY

Mohamed F. Abdul-Aziz., Yara M. Eid, *Laila M. Hendawy, Eman M. Fahmy and Nada G. Salama

Division of Endocrinology, Department of Internal Medicine, Faculty of Medicine, Ain Shams University, Cairo, Egypt

ARTICLE INFO

Article History:

Received 18th April, 2016
Received in revised form 10th May, 2016
Accepted 06th June, 2016
Published online 28th July, 2016

Key Words:

Ghrelin, growth, puberty, Constitutional delay

ABSTRACT

Constitutional delay in growth and puberty (CDGP) is a common clinical observation in childhood. Ghrelin may play a role in puberty initiation and progress. We aim to evaluate the association between ghrelin and CDGP. Case control study was conducted on 51 adolescent boys, 25 aged 14 – 18 years with CDGP (cases) and 26 healthy adolescent boys matched for age (control). Participants were subjected to history, clinical examination and investigations including CBC, ESR, Fasting blood glucose, TSH, Free T4, Growth hormone, Prolactin, FSH, LH, Testosterone, Ghrelin , Plain X-ray left wrist for bone age. Our study showed a **highly significant** difference between both groups in serum ghrelin (P=0.000), a **highly significant negative correlation** between ghrelin and FSH and serum testosterone **(P < 0.001) and significant negative** correlation between serum ghrelin and LH and growth hormone. **(P < 0.05).**Ghrelin level is associated with Constitutional delay in growth and puberty.

INTRODUCTION

Puberty is the duration of life which ends into adulthood through physiologic and psychologic changes. The onset of puberty is noticed by the appearance of secondary sexual characters, especially the appearance of breast in females and enlargement of testis in males **(**Beccuti and Ghizzoni, 2011).

Adrenarche usually starts before puberty, which causes increase of adrenal androgen between ages 6–10. It is noticed by the early appearance of pubic and axillary hair (Plant, 2001). The onset of puberty is associated with high GnRH pulse, which starts before the increase in sex hormones, LH and FSH (Plant, 2001).

Recent studies showed that ghrelin and leptin may play a role in initiation and progression of puberty. They are involved in regulation of GnRH secretion, with inhibitory effect of ghrelin and stimulatory effect of leptin (El-Eshmawy *et al*., 2010).

MATERIALS AND METHODS

Our case control study was conducted in the period from January 2013 till September 2014 in Egypt. The study included fifty one adolescent boys, twenty five adolescent boys aged 14 – 18 years old with CDGP who attended the endocrinology outpatient clinic at Ain Shams University hospitals (group 1). Twenty six healthy adolescent boys matched for age in the mid and late pubertal stage (3-5 Tanner stage) were evaluated as controls (group 2). Our study was accepted by the local ethical committee and a written consent was taken from all subjects to be included in our study.

All Participants enrolled in our study were subjected to detailed history including natal and perinatal history to exclude any brain trauma as difficulty in labour, breech delivery, birth injuries, asphyxia or congenital anomalies, pre-pubertal history to exclude cryptorchidism, history of endocrinological disorders such as diabetes mellitus or thyroid disorders, neurological manifestations such as seizures or visual disturbance, nutritional history and parental pubertal age of onset. Thorough clinical examination included blood pressure, height, weight, calculation of BMI, arm span, calculation of mid parental height and adult final height, The measurements were taken and plotted against Egyptian Growth curves for Boys aged 2-21 years (Diabetes Endocrinology Metabolism Pediatric Unit) Pubertal staging was assessed according to the method of Tanner and Whitehouse (Tanner J.M. and Whitehouse, 1976). Also left wrist radiographs were done for assessment of bone age.

Subjects with any chronic illness like diabetes mellitus, congenital heart disease, severe asthma, thalassemia major, neurological disorders (space occupying lesion), thyroid disorders, growth hormone disorders, natal and perinatal complications such as difficult normal labour, breech delivery,

*Corresponding author: *Laila M. Hendawy*
Division of Endocrinology, Department of Internal Medicine, Faculty of Medicine, Ain Shams University, Cairo, Egypt

birth injuries, asphyxia, congenital anomalies, BMI below 19 or exercise heavily were excluded. Also, subjects receiving any medications known to delay pubertal development such as prolonged use of corticosteroids, cyclophosphamide were excluded from our study.

The diagnosis of CDGP was based on the following criteria:

1. Short stature (height less than 2 SD below the mean)
2. Delayed puberty (the absence of testicular enlargement in boys at an age that is 2 to 2.5 SD later than the population mean)
3. Bone age below the 10th centile for chronological age (delayed by more than 1.5 years) Skeletal age was determined on left wrist radiographs using Greulich and Pyle method. Bone age delay was defined as a bone age that is 2 SD or more below the mean. This is approximately 20 percent below the chronological age. This translates to a difference between bone age and chronological age of approximately, 18 months between 4 and 12 years, and 24 months after age 12 (Bayley and Pinneau, 1952)
4. Absence of other causes of delayed puberty on history, examination and investigations.

Laboratory Investigations

CBC, ESR, Fasting blood glucose, Urine and stool analysis, TSH, Free T4 Growth hormone with reporting of the basal level and the highest recorded after stimulation reading, Prolactin, FSH, LH, Serum Total Testosterone, Serum Ghrelin by ELISA, Biovender, Czech republic

Statistical analysis

After collection of data, revision and tabulation; analysis was performed using SPSS statistical package version 22 (SPSS, Inc., Chicago, IL, USA).Continuous data was expressed as mean ± SD (Standard deviation). Comparison between groups done using Student T-Test (t).All quantitative data were correlated with each other using Pearson correlation coefficient(r). Non-significant test if $P > 0.05$, significant if $P < 0.05$ and highly significant if $P < 0.001$.

RESULTS

According to our study on comparing both groups, there was a **significant** difference regarding height, weight, BMI, arm span, serum ghrelin, fT4, serum testosterone and bone age **p < 0.001.**There was a **significant** difference between both groups in systolic blood pressure, diastolic blood pressure, TSH, growth hormone (basal and after stimulation), FSH and LH **p < 0.05.** There **was no significant** difference between both groups as regards to hemoglobin, platelet count, total leucocytic count, ESR and prolactin **p > 0.05 (table 1).**

On correlating serum ghrelin with different parameters, we found **a highly significant negative correlation** between ghrelin and age, nutrition, BMI, Tanner staging (P and G), TSH, free T4, FSH and serum testosterone **P < 0.001.**There was also a **significant negative** correlation between serum ghrelin and both systolic and diastolic blood pressure, growth hormone (basal and after stimulation) and serum LH **P-value <0.05**.However, there was a **non-significant** correlation between ghrelin and other parameters **table 2.**

Table 1 Comparison between group 1 and 2 regarding demographic and laboratory variables using the T-test

Characteristics	Adolescents with CDGP (n=25)		Normal control (n=26)		T	P-value
	Mean	SD	Mean	SD		
Age	15.16	±1.106	15.35	±0.977	0.636	0.527
Weight (Kg)	42.20	±9.367	62.46	±6.848	-8.790-	0.000
Height (cm)	145.12	±10.561	159.65	±6.922	-5.789-	0.000
BMI (Kg/m²)	19.92	±2.886	24.32	±1.865	-6.404-	0.000
Arm span (cm)	149.20	±11.109	162.38	±5.960	-5.252-	0.000
Systolic Bp (mmHg)	110.40	±7.348	115.58	±4.965	2.936	0.005
Diastolic Bp (mmHg)	71.40	±7.572	75.96	±6.931	2.242	0.030
Bone age (years)	13.20	±0.764	14.85	±1.047	6.434	0.008
Hemoglobin	12.56	±0.917	12.46	±0.989	0.369	0.714
Platelet count	289.56	±55.517	289.54	±64.085	0.001	0.999
Total leucocytic count	6.19	±1.531	6.52	±1.524	-0.766-	0.447
ESR	6.92	±1.579	5.88	±2.582	1.735	0.090
Fasting blood glucose (mg/dl)	85.28	±7.640	87.27	±6.960	-0.971-	0.336
TSH	2.80	±0.816	3.77	±1.394	-3.043-	0.004
Free T4	1.28	±0.458	1.73	±0.452	-3.534-	0.001
Growth Hormone (basal)	2.44	±1.850	4.04	±2.793	-2.418-	0.020
Growth Hormone (after stimulation)	11.28	±4.078	13.50	±2.534	-2.325-	0.025
Prolactin	7.56	±5.757	7.62	±3.086	-0.043-	0.966
FSH (mIU/mL)	3.96	±2.071	7.15	±2.556	-4.911-	0.032
LH (mIU/mL)	5.20	±4.848	7.46	±2.549	-2.073-	0.012
Testosterone (ng/dl)	234.80	±167.769	391.42	±82.405	-4.205-	0.000
Ghrelin (pg/dl)	82.36	±8.869	56.23	±2.643	14.140	0.000

Table 2 Correlation between ghrelin and the different variables using pearson correlation

Variable	Ghrelin	
	R	P-value
Age	0.897	0.000
SBP	-0.293-	0.020
DBP	-0.330-	0.010
Tanner staging (P)	-0.830-	0.000
Tanner staging (G)	-0.836-	0.000
BMI	-0.631-	0.000
Hb	-0.041-	0.390
Plt	0.004	0.488
TLC	0.115	0.212
ESR	0.233	0.051
Fasting blood glucose	-0.058-	0.345
TSH	-0.416-	0.001
Free T4	-0.442-	0.001
GH (basal)	-0.257-	0.036
GH (after stimulation)	-0.273-	0.027
Prolactin	0.006	0.483
LH	-0.317-	0.012
FSH	-0.533-	0.000
Testosterone	-0.439-	0.001

DISCUSSION

According to our study, ghrelin level showed a highly significant elevation in adolescent boys with CDGP compared to healthy controls. This is consistent with studies reported that children with CDGP have higher ghrelin levels than children with normal growth (Altuğ Şen *et al*., 2010).

The effect of ghrelin on the gonadotropic axis starts at the hypothalamus, ghrelin inhibited secretion of GnRH in vivo and administration of ghrelin in vitro caused decrease in GnRH frequency by hypothalamic explants **(**Lebrethon *et al*., 2004).

Also, hormonal studies demonstrated a progressive decrease in the circulating levels of ghrelin along puberty in humans. Although the functional relevance of such a decrease in the pubertal activation of the HPG axis not clearly studied, it is hypothesize that, if ghrelin conducts a dominant inhibitory

effect on puberty onset in humans, this decrease in its plasma levels, in proper metabolic conditions, may play a permissive role in pubertal onset (Kluge *et al*., 2004).

Our results showed a significant statistical difference between both groups regarding FSH level which was higher in healthy adolescent boys as well as a negative correlation between serum ghrelin and FSH. This is consistent with Other studies found that ghrelin administration can decrease FSH secretion in male humans by 25 %, although the amplitude of this inhibition is much lower than that of LH (Kluge *et al*., 2009).

Our results also showed a significant difference between both groups regarding LH being higher in healthy normal adolescents as well as a negative significant correlation between ghrelin and LH. This goes with studies stated that ghrelin is able to inhibit pulsatile LH release in prepubertal male rats and in gonadectomized male and female rats (Furuta *et al*., 2001), (Fernandez-Fernandez *et al*., 2004).

Also in the current study, there was a highly significant difference as regard serum testosterone being higher in healthy adolescent boys as well as a significant negative correlation between serum ghrelin and serum total testosterone. This is consistent with others found that ghrelin inhibits hCG and LH stimulated testosterone secretion by Leydig cells via decreasing c-AMP formation and decreasing mRNAs which encode many factors in the steroidogenesis (Tena-Sempere *et al*.,2013).

The results of the present study showed that there is a statistically significant difference between both groups as regard to growth hormone level (basal and after stimulation) which appear to be higher in normal healthy adolescents than those with CDGP. There was also a significant negative correlation between serum ghrelin and growth hormone (basal and after stimulation).This finding opposes other studies which reported that ghrelin increases GH secretion in vivo and vitro. Ghrelin causes increase in the level of intracellular Ca +2 inositol triphosphate which increases GH release (Kojima and Kangawa, 2005).

Also another study stated that ghrelin has been shown to control GH releasing hormone and somatostatin systems. However, genetic analysis showed that mutation of GHSR1a (Growth hormone secretagogue receptor variant) can lead to isolated GH deficiency (IGHD) and idiopathic short stature (ISS) and also a subgroup with CDGP in certain ethnic groups such as Brazilian, Europeans and Japanese (Seoane *et al*., 2003). These mutations lead to prominent reductions in cell-surface expression and activity of the GHS-R1a with of growth and puberty (Yin *et al*., 2014).

The results of the current study also showed that there is a highly significant difference between both groups in weight, height, BMI which appear to be higher in normal healthy adolescents than adolescents with CDGP. There was also a significant negative correlation between serum ghrelin and BMI. This matches many previous studies reporting that the BMI of the adolescent boys with CDGP were lower than that of the healthy adolescents (control group) (El-Eshmawy *et al*., 2010), (Otto *et al*., 2001).

A certain amount of body fat is needed to start and maintain puberty in mammals (El-Eshmawy *et al*., 2010), Persons with CDGP are typically underweight may be due to decreased energy intake (Wudy *et al*., 2005) or increased energy expenditure (Han *et al*., 2006).

The results of the present study showed a significant difference between both groups as regard to TSH and free T4, which were higher in healthy controls than those with constitutional delayed puberty**.** There was also a significant negative correlation between serum ghrelin and TSH and free T4.These results were in agreement with other studies found that ghrelin hormone administration to 20 healthy subjects caused initial increase of free T4 followed by significant decrease of TSH. The decrease in TSH caused by direct inhibition at the level of hypothalamic and to a smaller extent by the negative feedback of the initially increased free T4 (Kluge *et al*., 2010).

CONCLUSION

Ghrelin level is significantly higher in CGDP individuals which indicate a possible role in this subset of patients. Further prospective studies are needed to clearly demonstrate its role.

References

1. Beccuti G. and Ghizzoni L., (2011): "Normal and Abnormal Puberty." Endotext. South Dartmouth.
2. Plant TM. "Leptin, growth hormone, and the onset of primate puberty." (2001): *The Journal of Clinical Endocrinology and Metabolism*; 86 (1): 458–460
3. El-Eshmawy M., Abdel Aal I. & El Hawary A. (2010): "Association of ghrelin and leptin with reproductive hormones in constitutional delay of growth and puberty**."** Reproductive Biology and Endocrinology; 8: 153
4. Diabetes Endocrinology Metabolism Pediatric Unit Cairo University, children's Hospital: Egyptian Growth curves, boys 2-21 height growth.
5. Tanner J.M. and Whitehouse R.H. (1976): "Clinical longitudinal standards for height, weight, height velocity, weight velocity, and stages of puberty." Archives of Disease in Childhood; 51: 170-179.
6. Bayley N. and Pinneau S.R., **(1952)** "Tables for predicting adult height from skeletal age: revised for use with the Greulich-Pyle hand standards." *Journal of Pediatrics*; 40: 423.
7. Altuğ Şen T., Şimşek D.G., Darcan S., *et al*, (2010): " Ghrelin Levels in Children with Constitutional Delay of Growth and Puberty" *Journal of Clinical Research in Pediatric Endocrinology*; 2:3:117-121
8. Lebrethon M.C., Aganina A., Fournier M., *et al*, (2007): "Effects of in vivo and in vitro administration of ghrelin, leptin and neuropeptide mediators on pulsatile gonadotrophin-releasing hormone secretion from male rat hypothalamus before and after puberty" *Journal of Neuroendocrinology*;19:181-188.
9. Kluge L., *et al*, (2004):"Ghrelin levels from fetal life through early adulthood: relationship with endocrine and metabolic and anthropometric measures." *Journal of Pediatrics*; 144: 30–35.
10. Kluge M., Uhr M., Bleninger P., *et al*, (2009):"Ghrelin suppresses secretion of FSH in males." Clinical Endocrinology;70: 920–923
11. Furuta M., Funabashi T. and Kimura F., **(2001)** "Intracerebroventricular administration of ghrelin

rapidly suppresses pulsatile luteinizing hormone secretion in ovariectomized rats." Biochemistry and Biophysical Research Community;288:780–785
12. Fernandez-Fernandez R., Tena-Sempere M., Aguilar E., *et al*, (2004):"Ghrelin effects on gonadotropin secretion in male and female rats." Neuroscience Letters; 362:103-107
13. Tena-Sempere M. L., Barreiro L., González C., *et al*., (2002) "Novel expression and functional role of ghrelin in rat testis" Endocrinology; 143: 2; 717–725
14. Kojima M. and Kangawa K. (2005):" Ghrelin structure and function" Physiological Reviews; 85: 495–522
15. Seoane L.M., Lopez M., Tovar S., *et al*, (2003): "Agouti-related peptide, neuropeptide Y, and somatostatin-producing neurons are targets for ghrelin actions in the rat hypothalamus." Endocrinology; 144: 544–551
16. Yin Y., *et al*, (2014):" The Growth Hormone Secretagogue Receptor: Its Intracellular Signaling and Regulation" *International Journal of Molecular Science*;15:3;4837-4855
17. Otto B., Cuntz U., Fruehauf E., *et al*, (2001):"Weight gain decreases elevated plasma ghrelin concentrations of patients with anorexia nervosa." *European Journal of Endocrinology*;145:669-673
18. Wudy S.A., Hagemann S., Dempfle A., *et al***, (2005):** "Children with idiopathic short stature are poor eaters and have decreased body mass index.**"** Pediatrics; 116**:**e52-e57.
19. Han J.C., Balagopal P., Sweeten S., *et al*, (2006):"Evidence for hypermetabolism in boys with constitutional delay of growth and maturation." *The Journal of Clinical Endocrinology and Metabolism*;91:2081–2086
20. Kluge M., S. Riedl, M. Uhr *et al*., (2010):"Ghrelin affects the hypothalamus-pituitary-thyroid axis in humans by increasing free thyroxine and decreasing TSH in plasma," *European Journal of Endocrinology*; 162: no. 6, 1059–1065.

****

How to cite this article:

Mohamed F. Abdul-Aziz *et al*., Ghrelin Level in Boys with Constitutional Delay In Growth And Puberty. *Int J Recent Sci Res.* 7(7), pp. 12511-12514.