ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN – POLONIA
VOL. LIX, N 2, 108 SECTIO D 2004

Department of Developmental Paediatrics, Skubiszewski Medical University of Lublin
Department of Obstetrics and Gynecology, Regional Hospital in Rzeszów

BEATA KULIK-RECHBERGER, OLIMPIA JANISZEWSKA

Insulin-like growth factor 1, its binding protein 3, and sex hormones in girls during puberty

Puberty is a time of very intensive hormonal and somatic changes, which is characterized by two main occurrences – an increase of the velocity of linear growth and development of secondary sexual characteristics. The main hormone that stimulates somatic growth is growth hormone (GH). Growth hormone acts directly on growing tissues and stimulates local production of insulin growth factor 1 (IGF-1) in the epiphyseal plate to induce bone growth (6, 12). Insulin growth factor is synthesized locally in fibroblasts, myoblasts, chondroblasts, osteoblasts and in other cells such as cells of central nerves system, digestive system or kidney where it acts in paracrine/autocrine manner. Circulating IGF-1 is synthesized in the liver and its concentration in serum is regulated by GH, insulin and nutrition. IGF-1 circulates in the blood as a free form and in complex with a binding protein. About 90% of IGF-1 is bounded with binding protein 3 (IGFBP-3). It plays a role of a store contributing to prolongation of the half-life of growth factor (10, 11). Apart from regulation of proliferation and/or differentiation of cartilage cells, IGF-1 stimulates osteoblasts to synthesis of collagen type I, which increases bone mass (5). Experimental evidence suggests that IGF-1 may also play an important role in the normal development of mammary tissue (14) as well as in augmentation of FSH activity in granuloma cells in ovary, which increases the production of estrogens (10).

It is well established that GH and IGF-1 are crucial for increased longitudinal bone growth during puberty, but for normal bone growth androgens and estrogens are also needed. Both estradiol and testosterone influence GH secretion in the pituitary and stimulate synthesis of IGF-1 independently on GH. (4). Some clinical observation indicates that sex steroids regulate longitudinal bone growth in a non GH-dependent manner. Estradiol and testosterone cause increments of bone mass and skeletal maturation (2, 3). Stimulatory effect of sex steroids on longitudinal bone growth occurs probably through regulation of binding protein amount that is important for bioactivity of IGF-1. With regard to information presented above we examine relationships between IGF-1, estradiol and testosterone as well as between IGFBP-3 and estradiol serum concentration in girls during puberty taking into consideration gonadotropins level and nutritional status.

MATERIAL AND METHODS

In this study 86 girls aged 9.8–14.7 were examined. Girls were divided into three groups according to the pubertal stage (using Tanner scale). The first group (n = 23) was composed of girls in prepubertal stage, the second group (n = 35) of girls in the second and third stages and the third group (n = 28) of girls in the fourth and fifth stages (Table 1). Body height, weight and thicknesses of skin folds at the triceps site, subscapular site and on abdomen were measured. Body mass index (BMI) was calculated.

Blood samples were collected and serum concentration of IGFBP-3, estradiol, FSH and LH were assessed. Additionally in 51 girls serum concentration of IGF-1, and testosterone were estimated (Table 2). This group was composed of 17 girls in prepubertal stage, 20 girls in the second and third pubertal stages and 14 girls in the fourth and fifth pubertal stages. All biochemical parameters were assessed using RIA method. Differences between groups were determined using U Mann-Whitney test. Correlations between parameters were given by the Pearson correlation coefficients.

RESULTS

The lowest mean serum IGFBP-3 concentration (4107.55 ± 735.37 ng/ml) was observed in girls in prepubertal stage (group I) (Table 1). Concentration of this protein was growing with pubertal development but in girls in the second and third stages (group II) was not different from the concentration in group I. The difference was observed, however, between binding proteins concentration in group II and III (respectively 4421.14 ± 135.50 ng/ml and 5122.60 ± 1436.68 ng/ml; $p < 0.03$). Similarly, as in the case of binding protein, the lowest serum concentration of IGF-1 (206.08 ± 73.78ng/ml) was observed in girls in the first group (Table 2). This value was statistically lower than in the second group (343.40 ± 164.09 ng/ml; $p < 0.01$) but the latter was lower than in girls in the third group (706.16 ± 325.22 ng/ml; $p < 0.001$). As expected, serum concentration of FSH, LH, estradiol and testosterone were growing with pubertal development as well (Table 1 and 2). Serum concentrations of estradiol, testosterone, FSH and LH were considerably higher in the second group than in the first group. In the third group serum concentrations of estradiol and testosterone were higher than in the second group but serum concentrations of FSH and LH were as high as in girls in the second group.

On analyzing relationships between investigated biochemical parameters it turned out that there were significant positive correlations between IGF-1 and testosterone serum concentration ($r = 0.44$; $p < 0.0001$), IGF-1 and estradiol serum concentrations ($r = 0.3$; $p < 0.01$) as well as between IGF-1 concentration and BMI ($r = 0.31$; $p < 0.03$) and IGF-1 and the sum of three skin folds thicknesses ($r = 0.29$; $p < 0.04$). It has been also proved that there are positive correlations between IGFBP-3 and estradiol serum concentration ($r = 0.47$; $p < 0.0001$) as well as between IGFBP-3 and age ($r = 0.38$; $p < 0.0001$), body height ($r = 0.34$; $p < 0.001$) and body weight ($r = 0.36$; $p < 0.001$) in girls during puberty.

Table 1. Mean age, anthropometrical feature and serum concentration of binding protein 3 (IGFBP-3), follicle stimulating hormone (FSH), luteinizing hormone (LH) and estradiol (E_2) in girls during puberty

	Group I (n=23)	Group II (n=35)	Group III (n=28)	I vs II	II vs III
Age (years)	10.1 ± 1.1	12.2 ± 1.4	13.7 ± 0.5	p < 0.001	p < 0.001
Body height (cm)	137.4 ± 5.5	151.1 ± 6.5	162.3 ± 5.6	p < 0.001	p < 0.001
Body weight (kg)	29.9 ± 4.1	39.9 ± 5.3	51.5 ± 8.2	p < 0.001	p < 0.001
BMI (kg/m^2)	15.8 ± 1.8	17.5 ± 1.7	19.6 ± 2.7	p < 0.001	p < 0.001
Sum of skin-folds (mm)	29.5 ± 10.2	32.1 ± 8.4	39.3 ± 11.1	ns	p < 0.006
FSH (IU/L)	2.18 ± 0.99	3.65 ± 1.36	3.51 ± 1.18	p < 0.001	ns
LH (IU/L)	1.79 ± 0.89	3.28 ± 2.02	4.04 ± 1.19	p < 0.002	ns
E_2 (pmol/L)	26.12 ± 34.60	103.92 ± 90.95	239.65 ± 142.3	p < 0.001	p < 0.001
IGFBP-3 (ng/ml)	4107.55 ± 735.37	4421.14 ± 135.50	5122.60 ± 1436.68	ns	0.03

Group I – girls in prepubertal stage; Group II – girls in the second and third pubertal stages; Group III – girls in the fourth and fifth pubertal stages; ns – not statistically different

Table 2. Mean age, anthropometrical feature and serum concentration of insulin-like growth factor 1 (IGF-1), estradiol (E_2) and testosterone (T) in girls during puberty

	Group I (n=17)	Group II (n=20)	Group III (n=14)	I vs II	II vs III
Age (years)	11.3 ± 0.8	11.9 ± 0.8	13.8 ± 0.8	$p < 0.05$	$p < 0.001$
Body height (cm)	139.5 ± 4.7	152.4 ± 5.0	161.0 ± 3.0	$p < 0.001$	$p < 0.001$
Body weight (kg)	31.2 ± 4.0	39.3 ± 5.1	52.0 ± 7.2	$p < 0.001$	$p < 0.001$
BMI (kg/m^2)	16.1 ± 1.8	16.9 ± 1.6	20.0 ± 2.7	ns	$p < 0.001$
Sum of skin-folds (mm)	29.9 ± 10.6	32.4 ± 8.1	39.8 ± 11.4	ns	$p < 0.006$
E_2 (pmol/L)	19.56 ± 28.60	77.56 ± 84.18	159.59 ± 113.07	$p < 0.006$	$p < 0.001$
T (nmol/L)	0.61 ± 0.31	0.91 ± 0.31	2.18 ± 0.96	$p < 0.003$	$p < 0.001$
IGF-1 (ng/ml)	206.08 ± 73.78	343.40 ± 164.09	706.16 ± 325.22	$p < 0.01$	$p < 0.001$

Group I – girls in prepubertal stage; Group II – girls in the second and third pubertal stages; Group III – girls in the fourth and fifth pubertal stages; ns – not statistically different

DISCUSSION

Insulin growth factor type 1 is necessary for normal growth of fetus and child in postnatal life. It takes part in the formation of the bones – it stimulates replication and differentiation of bones cells and bone matrix apposition by augmentation of synthesis of collagen type I (the only collagen in bones) and blocking degradation of bones collagen, probably by reducing expression of collagenase in osteoblasts (8).

The insulin-like growth factor activity depends on its production, blood concentration and receptors that have shown to be present in different organs and cells of the body. In serum, IGF-1 is bound to binding proteins. They act as a storage of protein that protect IGF-1 from degradation and regulate its bioactivity. Binding proteins are synthesized in the liver but also locally in many cells of the body like in bones when they take part in their growth (11).

As it is commonly known, puberty is a time of dramatic alteration of the overall rate of body growth. After the onset of puberty the velocity of linear growth rapidly increases. We call this phenomenon "pubertal growth spurt". The pubertal growth spurt occurs at the second and third stage of pubertal development. From our own observation we know that serum concentration of IGF-1 in girls in the first stage of pubertal development (peripubertal period) is lower than in the second and third pubertal stage but in the fourth and fifth stages are higher than in earlier stages. On the basis of this result, we conclude that serum IGF-1 concentration increases at the time of acceleration of growth. After the growth spurt, when girls achieve menarche, growth velocity decreases but IGF-1 concentration is still high. The same results have been obtained by other authors (3, 9). From their observations we know that IGF-1 levels can remain elevated for a few years after menarche, despite the fact that growth rate is decreasing at this time. For example Juul (9) noted that the maximum increase of serum IGF-1 concentration is present in girls in the third and fourth pubertal stages and after that its concentration decreases. Blumsohn (3) observed a higher concentration of IGF in girls before menarche than after menarche, but the oldest girl was 15.5 years old. Toublanç (15) noted that the level of this factor is elevated for a few years after a peak of growth spurt. In our study the age of the oldest girl does not exceed fifteen. Why the level of IGF-1 remain high during decrease of velocity

of growth is not clear. The evaluation of IGF-1 concentration during puberty is complicated by the fact that IGF-1 circulate in blood binding to protein, mainly IGFBP-3 and IGFBP-1. Almost 85–95% of IGF-1 is connected to IGFBP-3. Estimating the serum concentration of this protein we conclude that it was growing with pubertal development and correlates with age, body height and weight. The increase of its concentration is not steady regular. In girls in the fourth and fifth pubertal stages concentration of growth factor was higher than in girls in earlier pubertal stages but in girls in the second and third pubertal stages was not different from the concentration in girls in prepubertal stage. These results can be explained by the fact that binding protein acts as a store for IGF-1 and regulates its activity. A marked increase of IGFBP-3 serum concentration during pubertal growth spurt can probably cause the decrease of growth velocity.

The majority of data indicates that apart from GH/IGF-1 systems, pubertal growth and development are regulated by the sex steroids as well (13). As it was expected, serum concentrations of gonadotropins (FSH and LH), estradiol and testosterone were growing with pubertal development. There were positive correlations between IGF-1 and estradiol as well as between estradiol and IGFBP-3 serum concentrations. Apart from the increase of estradiol concentration, the increase of testosterone was also observed in the investigated girls. It is commonly known that testosterone takes part in growth by regulation of growth hormone releasing hormone (GHRH) and somatotropin release inhibiting hormone (SRIH) synthesis as well as by direct stimulation of somatotropic cells (1). On the basis of this information relationships between serum concentration of IGF-1 and testosterone were investigated. We have found positive correlations between these two hormones. It can suggest the influence of testosterone on GH/IGF-1 axis.

A number of investigators demonstrate that IGF-1 is down-regulated during fasting, suggesting that it plays a role in regulating metabolism (7). In our observation we have found positive correlations between IGF-1 serum concentration and body mass index as well as between the concentration of IGF-1 and the ticknesses of skin folds.

CONCLUSION

These results allow to conclude that there are relationships between serum concentrations of sex hormones, IGF-1, IGFBP-3 and anthropometrical parameters in girls during puberty.

REFERENCES

1. Argente J. et al.: Sex steroid and developmental regulation of somatostatin and growth hormone-releasing hormone gene expression. In: Human Growth: Basic and Clinical Aspects. M. Hernandez & J. Argente eds., 295, 1992.
2. Attie K. M. et al.: The pubertal growth spurt in eight patients with true precocious puberty and growth hormone deficiency: evidence for a direct role of sex steroids. J. Clin. Endocrinol. Metab., 71, 975, 1990.
3. Blumsohn A. et al.: Biochemical markers of bone turnover in girls during puberty. Clin. Endocrinol., 40, 663, 1994.
4. Bourguinon J. P.: Linear growth as a function of age at onset of puberty and sex steroid dosage. Therapeutic implication. Endocr. Rev., 9, 467, 1988.
5. Canalis E.: Insulin-like growth factors and osteoporosis. Bone, 21, 215, 1997.
6. Fichna P. et al.: Wydzielanie i działanie hormonu wzrostu. Ped. Prakt., 2, 113, 1994.

7. Grinspoon S. K. et al.: Effects of rhIGF-1 administration on bone turnover during short--term fasting. J. Clin. Invest., 96, 900, 1995.
8. Hock J. M. et al.: Insulin-like growth factor I /IGF-1/ has independent effects on bone matrix formation and cell replication. Endocrinology, 112, 254, 1988.
9. Juul A. et al.: Serum levels of insulin-like growth factor (IGF)-binding protein-3 (IGFBP-3) in healthy infants, children and adolescents: the relationship to IGF-I, IGF-II, IGFBP-1, IGFBP-2, age, sex body mass index and pubertal maturation. J. Clin. Endocrinol. Metab., 80, 2534, 1995.
10. Khamsi F. et al.: Recent discoveries in physiology of insulin-like growth factor-1 and its interaction with gonadotropins in folliculogenesis. Endocrine, 16, 151, 2001.
11. Niedźwiedzka A.: Insulinopodobny czynnik wzrostowy 1 (somatomedyna C) i jego białka wiążące 1 i 3 u dzieci, ze szczególnym uwzględnieniem cukrzycy. Endokrynol. Diabetol. Chor. Przemiany Materii Wieku Rozw., 6, 51, 2000.
12. Ohlsson C. et al.: Endocrine regulation of longitudinal bone growth. Acta Paediatr., Suppl. 391, 33, 1993.
13. Pescovitz O. H.: The endocrinology of the pubertal growth spurt. Acta Paediatr. Scand., Suppl. 367, 119, 1990.
14. Ruan W. et al.: Intact and aminoterminally shortened forms of insulin-like growth factor I induce mammary gland differentiation and development. Proc. Natl. Acad. Sci., USA, 89, 10872, 1992.
15. Toublanc J. E.: Modifications of growth hormone secretion during female puberty. Ann. N.Y. Acad. Sci., 816, 60, 1997.

SUMMARY

The aim of the study was to estimate relationships between serum concentration of insulin-like growth factor 1 (IGF-1), its binding protein (IGFBP-3), estradiol and testosterone in girls during puberty, taking into consideration gonadotropins level and nutritional status. Eighty-six girls aged 9.8-14.7 were examined. The girls were divided into three groups according to the pubertal stage. Body height, weight and thicknesses of skin folds were measured. Biochemical parameters were assessed using RIA method. It has been found that all investigated parameters are growing with pubertal development. There are significant positive correlations between IGF-1 and testosterone, IGF-1 and estradiol serum concentrations as well as between IGF-1 serum concentration, BMI and sum of skin folds thicknesses. It has been also proved that there are positive correlations between IGFBP-3 and estradiol serum concentration as well as between IGFBP-3 and age, body height and body weight in girls during puberty.

Insulinopodobny czynnik wzrostu 1, jego białko wiążące 3 oraz hormony płciowe u dziewcząt w okresie pokwitania

Celem pracy było ustalenie zależności pomiędzy stężeniem insulinopodobnego czynnika wzrostu typu 1, jego białka wiążącego 3 (IGFBP-3), estradiolu i testosteronu w surowicy dziewcząt w okresie pokwitania, z uwzględnieniem stężenia gonadotropin w surowicy i stanu odżywienia. W badaniach wzięło udział 86 dziewcząt w wieku 9,8 –14,7 lat. Dziewczęta podzielono na grupy w zależności od stopnia dojrzałości płciowej. Mierzono wysokość, masę ciała oraz grubość fałdów skórno-tłuszczowych. Parametry biochemiczne oznaczano przy użyciu metody RIA. Wszystkie oceniane parametry wzrastały w miarę osiągania dojrzałości płciowej. Stwierdzono dodatnie korelacje pomiędzy stężeniem IGF-1 i testosteronu, IGF-1 i estradiolu w surowicy, a także pomiędzy stężeniem IGF-1, BMI i sumą grubości fałdów skórno--tłuszczowych. Wykazano również dodatnie korelacje pomiędzy stężeniem IGFBP-3 i estradiolu w surowicy oraz stężeniem IGFBP-3 i wiekiem, wysokością i masą ciała dziewcząt w okresie pokwitania.