INTRODUCTION

Growth hormone (GH) is one of several hormones produced by the pituitary gland. When the child's pituitary is producing GH in inadequate amounts or not at all, the child is diagnosed as "growth hormone deficient" (GHD). Sometimes it occurs by itself, whereas other times it accompanies other pituitary hormone deficiencies. The type of underlying pathophysiology differs in childhood-onset compared with adult-onset GHD. In childhood, the commonest etiology is isolated idiopathic GHD ***(Saz-Parkinson et al., 2013)***.

Children with GHD usually present with short stature and a low growth velocity for age and pubertal stage. Those children present with clinical characteristics that vary based on the underlying etiology and severity of the insufficiency. A "typical" clinical picture of the GHD child includes severe proportional short stature, height velocity abnormal for age, delayed bone age and delayed puberty. In addition to the growth sequelae, there are also metabolic consequences of pediatric GHD. These include a tendency toward (or the presence of) hypoglycemia in infancy and impaired lipid mobilization, protein synthesis, bone mineralization, and anabolism in childhood. These later consequences persist irrespective of the status of epiphyseal growth ***(Root and Diamond, 2002)***.

Vitamin D is critical for calcium (Ca) homeostasis and for mineralization of the skeleton, especially during periods of rapid growth, namely infantile and pubertal growth periods. Without vitamin D, only 10-15% of dietary Ca and about 60% of phosphorus is absorbed. The active form, 1,25-dihydroxy vitamin D (1,25-$(OH)_2$ D3), markedly increases the efficiency of intestinal Ca and phosphorus absorption ***(Soliman et al., 2008)***. Serum levels below 30 ng/ml are associated with a significant decrease in intestinal Ca absorption. In children, adolescents, and adults; this is associated with increased PTH and deceased IGF-I secretion. Serum levels of 25-OH-D are directly related to bone mineral density with a maximum density achieved when the 25-OH-D level reached 40 ng/ml or more ***(Bischoff-Ferrari et al., 2006)***.

There is a bidirectional link between insulin growth factor-1 IGF-I and vitamin D, which is played out in different forms at the systemic (circulating) and the local (growth plate) level. Both GH and IGF-I significantly increase renal 1a-hydroxylase expression and serum 1,25 (OH)2 D3 concentrations ***(Ameri et al., 2013)***.

Growth hormone deficiency and vitamin D deficiency are two issues of increasing concern and the relation and the association between them are yet to be understood ***(Ciresi et al., 2014)***.

AIM OF THE WORK

To assess:

1- Serum 25 (OH)D in prepubertal children with isolated growth hormone deficiency.

2- Its relation to various auxological parameters.

GROWTH HORMONE DEFICIENCY

Growth

Growth is a complex biological process, product of the interaction between multiple endogenous factors (genetic, hormonal, metabolic, receptivity of target tissues) and exogenous factors (nutrition, physical activity and psychosocial influences), through which living creatures, at the same time as increasing their size, they physiologically mature and progressively acquire a complete functional capacity ***(Martínez, 2011).***

Growth is a biological process in which, as well as an increase in body weight, there is a progressive morphologic and functional maturing of the individual. Therefore, it is a quantitative process, with respect to the increase in number and cell size and extracellular substances, but it is also a qualitative process as it entails a progressive specialization of all the organisms´ systems ***(Saz-Parkinson et al., 2013).***

Growth factors

- They can be classified into four different types:

- Determinant factors.
- Permissive factors.
- Regulatory factors.
- Achieving factors ***(Saz-Parkinson et al., 2013).***

A) **Determinant factors:** They are the genetic ones, those responsible for maximum growth potential. They are determined by the parents´ size and by the rhythm of growth throughout the different sequential stages of life. Height is determined in a polygenic way which is why chromosome disorders are almost always associated with undergrowth, on many occasions presenting prenatally. The genes which regulate size in the sexual chromosomes have been found to be located in the short arm of chromosome X and the long arm of chromosome Y ***(Saz-Parkinson et al., 2013).***

B) **Permissive Factors:** These are the ones which make it possible to achieve a genetically determined growth. We can distinguish two different types ***(Saz-Parkinson et al., 2013)***:

1. **Nutrition-metabolic factors:** In order to achieve a normal growth, the nutrients and oxygen provided to the organism must be sufficient, and the absorption – digestion functions, as well as the organism´s metabolism, needs to be adequate. Malnutrition is still the main cause of a high percentage of cases of undergrowth. On the other hand, in obesity there is a transient acceleration of the growth rate, reaching a size above the mean and an increase in the insulin growth factor (IGF-I) is observed, although the final size achieved is that which is genetically expected ***(Saz-Parkinson et al., 2013).***

2. **Environmental factors**: The environmental surrounding of the subject is included here, such as the socio-economic status, lifestyle, climate, rural or urban setting, and in particular the family setting (number of children, emotional relationships, etc.). The lack of emotional support cannot be forgotten when evaluating the possible causes of short stature, even more so taking into account that it can lead to severe undergrowth ***(Cassorla, 2002)***.

C) **Regulatory factors:** They coordinate the determinant and permissive factors so growth can take place. There are two different groups ***(Saz-Parkinson et al., 2013):***

1. **Hormonal factors**: The hormones which are most involved in growth, in addition to growth hormone (GH), are thyroid hormones, cortisol, sexual steroids, either from the gonads or adrenal glands, PTH, vitamin D and its metabolites, and insulin, as well as all hypothalamic factors that regulate synthesis and secretion of the fore-mentioned compounds ***(Saz-Parkinson et al., 2013).***

2. **Autocrine and paracrine factors**: They are peptide factors which act by stimulating or inhibiting cell proliferation and growth at a local level. If the action takes place on the same cell that has synthesized them, it is referred to as autocrine control, whereas if it occurs on nearby cells, it is paracrine control. In both cases, the action mechanism is due to interaction with cell membrane receptors, which induce physical-chemical

changes in the cell. IGFs or somatomedins are included in this group, and their synthesis depends on age, GH and nutrition state ***(Saz-Parkinson et al., 2013).***

D) Achieving factors: They are represented by the "target" organs on which the rest of the growth factors act. The most important one is bone, and within the bone, the growth cartilage ***(Saz-Parkinson et al., 2013).***

Growth during childhood is a relatively stable process. Until about the age of 4 years, girls grow slightly faster than boys and both sexes then average a rate of 5–6 cm/yr and 2.5 kg/yr until the onset of puberty. A general rule of thumb is that a child grows (10 inches or 25 cm) in the first year of life, half that (5 inches or 12-13 cm) in the second year, and then (2.5 inches or 5 to 6 cm) until puberty, and this is called Growth Velocity (table (1)). Assuming an average birth length of (20 inches or 51 cm) results in an average 1-year-old being (30 inches or 76 cm), a 2-year-old (35 inches or 89 cm), a 4-year-old (40 inches or 102 cm), and an 8-year-old 50 in (127 cm) ***(Alan et al., 2014).***

Table (1): Normal growth velocity at various life stages

Life stage	Growth velocity per year
- First year - Second year - Fourth year - Pre-pubertal nadir - Pubertal growth spurt	- 23 to 27 cm (9 to 11 in) - 10 to 14 cm (4 to 6 in) - 6 to 7 cm (2 to 3 in) - 5 to 5.5 cm (2 to 2.2 in) - Girls: 8 to 12 cm (3 to 5 in) Boys: 10 to 14 cm (4 to 6 in)

(Benjamin and Mary, 2008)

Short stature

Growth disturbances manifest as abnormal absolute height or growth velocity ***(Grimberg and Lifshitz, 2007).***

Short stature is defined as height that is two standard deviations below the mean height for age and sex (less than the 3rd percentile) or more than two standard deviations below the mid-parental height ***(Grimberg and Lifshitz, 2007)*** (table (2)).

Table (2): Mid-Prental height calculation

• *Boys: [father's height in cm + (mother's height in cm + 13 cm)]/2*
• *Girls: [(father's height in cm - 13 cm) + mother's height in cm]/2*

(Benjamin and Mary, 2008)

A growth velocity disorder is defined as an abnormally slow growth rate, which may manifest as height deceleration across two major percentile lines on the growth chart. Growth velocity looks at the change in height measured on two separate occasions, relative to the time that lapsed between the two measurements. It is annualized, by the following equation, to enable comparisons:

Growth velocity (cm/yr) =

$$\frac{\text{(Height (cm) measured at time2 \_ Height (cm) measured at time1)}}{\text{Number of months between time2 and time1}}$$

x 12 (months/yr) ***(Haymond et al., 2013)***

In some cases, short stature or slow growth is the initial sign of a serious underlying disease in an otherwise healthy-appearing child ***(Boersma et al., 2002).***

The different causes of short stature are listed in the table below (table (3)) and how to approach a diagnosis is discussed in (figure (1)).

Table (3): Causes of short stature according to the ESPE classification.

A Primary growth disorders
- *A1 Clinically defined syndromes*
  - Turner syndrome
  - Cornelia de Lange syndrome
  - DiDeorge syndrome (velocardiofacial syndrome)
  - Down syndrome
  - Noonan syndrome
  - Prader-Willi-Labhart syndrome
  - Von Recklinghausen's disease (neurofibromatosis type 1)
  - Silver-Russell syndrome
- *A2 Small for gestational age with failure of catch-up growth*
  - IGF-I deficiency, IGF resistance
  - Due to known cause, e.g. prenatal infections, drugs, smoking, alcohol
  - Idiopathic
- *A3 Skeletal dysplasias*
  - Achondroplasia
  - Hypochondroplasia
  - Dyschondrosteosis (Leri-Weill and other defects in the SHOX gene)
  - Osteogenesis imperfecta I–VI
  - Mucopolysaccharidosis (type IH, IS, II–VII)
  - Mucolipidosis (type II and III)
- *A4 Dysplasias with defective mineralization*

B Secondary growth disorders
- *B1 Insufficient nutrient intake (malnutrition)*
- *B2 Disorders in organ systems*
  - Cardiac disorders
  - Pulmonary disorders, e.g. cystic fibrosis
  - Liver disorders
  - Intestinal disorders, e.g. Crohn's disease, malabsorption syndromes
  - Short bowel syndrome
  - Renal disorders, e.g. Fanconi syndrome, renal acidosis
  - Chronic anemia
- *B3 Growth hormone deficiency (secondary IGF-I deficiency)*
  - Idiopathic
  - Genetic (HESX1, PROP1, POU1F1, LHX3, LHX4, GHRHR, GH)
  - Associated with syndromes or cerebral or facial malformations, e.g. septo-optic dysplasia, empty sella syndrome
  - Associated with prenatal infections, e.g. rubella
  - Acquired (craniopharyngioma, other pituitary tumors, e.g. germinoma, hamartoma)
  - Head trauma
  - Central nervous system infections
  - Granulomatous diseases, e.g. histiocytosis
- *B4 Other disorders of the growth hormone-IGF axis (primary IGF-I deficiency and resistance)*
  - Bioinactive growth hormone
  - Abnormalities of the growth hormone receptor (growth hormone insensitivity syndrome, Laron syndrome)
  - Abnormalities of GH signal transduction, e.g. STAT5B defect
  - ALS (acid-labile subunit) deficiency
  - IGF-I deficiency
  - IGF resistance (IGF1R defects, postreceptor defects)
- *B5 Other endocrine disorders*
  - Cushing syndrome
  - Hypothyroidism
  - Leprechaunism
  - Diabetes mellitus (poorly controlled)
  - Short adult stature caused by accelerated bone maturation, e.g. precocious puberty, hyperthyroidism, congenital adrenal hyperplasia, exogenous estrogens or androgens
- *B6 Metabolic disorders*
  - Disorders of calcium and phosphorus metabolism
  - Disorders of carbohydrate metabolism
  - Disorders of lipid metabolism
  - Disorders of protein metabolism
- *B7 Psychosocial*
  - Emotional deprivation
  - Anorexia nervosa
  - Depression
- *B8 Iatrogenic*
  - Systemic glucocorticoid therapy
  - Local glucocorticoid therapy (inhalation, intestinal, other)
  - Other medication
  - Treatment of childhood malignancy
  - Total body irradiation
  - Chemotherapy
  - Other specified iatrogenic causes

C Idiopathic short stature
- *C1 Familial (idiopathic) short stature*
- *C2 Non-familial (idiopathic) short stature*

(Wit, 2007)

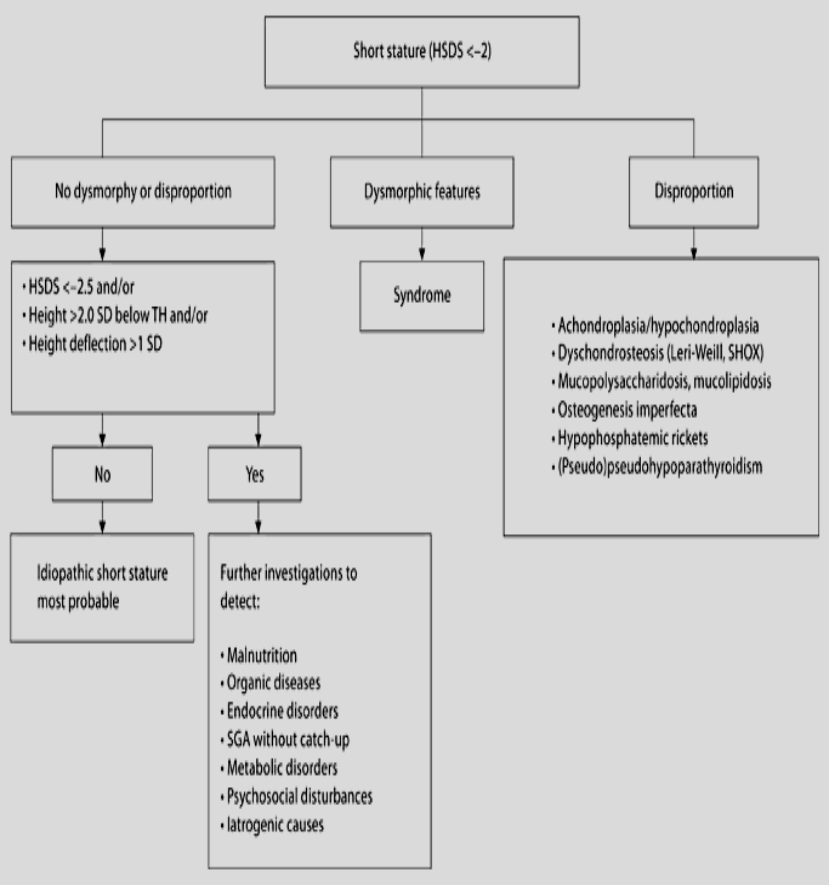

Figure (1): Diagnostic approach in children with short stature. TH = Target height; SGA = small for gestational age ***(Wilma et al., 2009)***.

Growth hormone

Growth hormone or somatotrophin is a polypeptide of 191 amino acids with a molecular weight of 22,000 KDa produced in the anterior pituitary gland ***(Saz-Parkinson et al., 2013).*** Growth hormone (GH) is a particularly important player in human physiology as it controls many physiological processes such as growth, bone mineralization, sugar and lipid metabolism, protein synthesis and stimulation of the immune system, and is in turn influenced by important external factors such as stress, sleep, exercise and food intake ***(Widdowson et al., 2009)***.

This peptide is secreted from the somatotropes cells in the anterior pituitary gland and is released in a pulsatile manner into the blood stream under the regulation of the two hypothalamic peptides: GH-releasing hormone (GHRH), which is stimulatory, and GH-inhibiting factor (GHIF; somatostatin), which is inhibitory ***(Kojima et al., 1999).*** The blood delivers GH to the liver and other target organs where it binds to the GH receptor (GHR) and mediates signal transduction through at least three pathways: phosphatidylinositol-3 kinase (PI3K), extracellular signal related kinases (ERK) 1 and 2, and Janus kinase-2 (JAK-2)-signal transducers and activators of transcription-5 (STAT-5) ***(Herrington and Carter-Su, 2001)***, the latter being a particularly important pathway for the growth-mediating actions of GH ***(Herrington, 2000)***.

Structure of growth hormone:

GH belongs to a family of proteins with structural similarity and certain common functions that include prolactin (Prl), somatolactin (SL), chorionic somatomammotropin (CS), proliferin (PLF) and proteins related to Prl (PLP).This family represents one of the most physiologically diverse protein groups that have evolved by gene duplication. The two most studied members of this family have been GH and Prl, which have been described from primitive fish to mammals; however, other members of the family are not so amply distributed or studied ***(Ascacio-Martínez and Barrera-Saldaña, 2012)***.

GH (Figure (2)), in general, has a molecular weight of around 22,000 Daltons (22 kDa or simply 22k) and does not require post-translational modifications. It is synthesized in somatotrophs in the hypophysis, intervening as an important endocrine factor in postnatal somatic growth and lactation ***(Ascacio-Martínez and Barrera-Saldaña, 2012).***

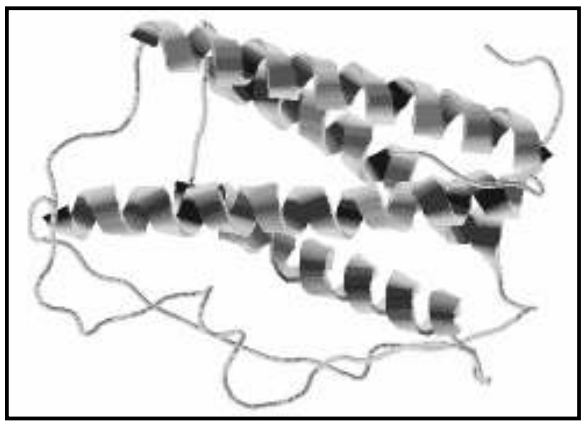

Figure (2): Growth hormone's consensus tridimensional structure. GH has in general 190 aminoacidic residues, four alpha helixes, and two sulphide bonds ***(Ascacio-Martínez and Barrera-Saldaña, 2012).***

Growth hormone secretion and regulation:

GH is secreted in a pulsatile fashion from the anterior hypophysis, beneath the hypothalamus in the brain ***(Rosen, 2000)***.

During human development, GH secretion is maximal during periods of growth, most obviously adolescence; thereafter both the periodicity and amplitude of GH secretion falls at a relatively low rate, for example, the total amount of GH secreted by a 60 year old man each day may be about half that secreted by a 20 year old ***(Rosen, 2000)***.

GH secretion usually occurs nocturnally, but may be stimulated during the day by high protein foods, especially those containing arginine ***(Rennie, 2003)***, and by exercise of both the aerobic and resistance types ***(Consitt et al., 2002)***, Apart from sleep, exercise is the most potent physiological stimulus of GH secretion. Obesity and aging also diminishes normal GH secretion and the response to stimuli such as arginine and clonidine ***(Rosen, 2000)***. The ability to increase GH with exercise is diminished with obesity and aging, but is certainly not abolished in either case ***(Copeland et al., 2002)*** (figure (3))**.**

GH secretion from the pituitary is under neural control from the hypothalamus through at least three hypothalamic factors: Growth Hormone Releasing Hormone (GHRH),

somatostatin (SS) and ghrelin. The GHRH neurons are located in the arcuate and ventromedial nuclei, and SS neurons are located in the anterior periventricular area. GHRH and SS release are controlled by a complex neuronal network, in which alfa-adrenergic, dopaminergic and serotoninergic signals stimulate GH secretion ***(Takaya et al., 2000)*** (figure (3)).

Ghrelin is expressed in the hypothalamus and the pituitary as well as the stomach, and is considered as a potential physiological regulator of GH secretion. The action of ghrelin on GH secretion is dependent on a functional GHRH system, and GHRH and ghrelin have synergic actions in vivo. The principal site of ghrelin action on GH release is the hypothalamus, but a minor effect is also present at the pituitary level ***(Takaya et al., 2000)***.

Thyrotropin (TSH) releasing hormone (TRH) plays not only a stimulating role in TSH and prolactin (PRL), but also an inhibitory action on GH secretion induced by a number of stimuli and during sleep. The inhibitory mechanisms remain to be elucidated ***(Tsumori et al., 1999).***

Once GH is released from the pituitary gland, it circulates in the blood to increase IGF-I production in many tissues, leading to a rise in blood IGF-I, which provides a long-term inhibition of further pituitary GH secretion ***(Roelfsema and Clark, 2001)*** (figure (3)).