Measurement of 5α-Reductase Activity Using Testosteron Total/Dehydrotestosteron Ratio

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Abstract

Introduction **:** DHT is essential for the development of the male sex characteristics before birth. In the adult, DHT is needed to develop and maintain male gender characteristics.DHT is derived from conversion of testosterone by 5α–Reductase and DHT has approximately 3 times greater affinity for androgen receptors than testosterone. The aims of this study is to measure 5alpha-reductase activity using TT / DHT ratio. ***Method :*** In 100 male patients was measured levels of TT and DHT. In 70 cases the level of TT and DHT was measured before and after HCG stimulation.TT/DHT ratio was calculating in order to reflect 5alpha-reductase activity. ***RESULTS*****:** In serum of patients was measured level of TT (0.81±0.11), DHT (16.4±4.3) and TT/DHT ratio (5.4±0.55). In cases aged 8-13 years after HCG stimulation TT and DHT levels rose respectively TT (1.43±0.34), DHT (29.4±6.5) and TT/DHT ratio (11.8±1.3). In puberty boys the TT/DHT ratio was (6.7 ± 3.5) and after HCG stimulation (,9.1±5.4).In neonates patient with ambiguous genitalia we observed a high level of TT and DHT .After the HCG stimulation we observed an increase in the level of TT in all cases and in 2 of them we had a TT/DHT ratio >27. ***CONCLUSION*****:** In normal males, TT and DHT rise in parallel, and we have a constant TT/DHT ratio. After HCG stimulation TT , DHT, and the TT/DHT ratio rose significantly. T/DHT ratio can help in investigation of 46, XY patients with ambiguous genitalia and normal testosterone synthesis

KEYWORDS: DHT, TT, 5α-reductases activity, TT/DHT ratio

1.Introduction :

The 5α-Reductases is produced in many tissues in both males and females, in the reproductive tract, testes and ovaries, skin, seminal vesicles, prostate, epididymis and many organ, including the Nervous System–There are three isoenzymes of 5α-reductase: steroid 5α-reductase 1, 2, and 3 (SRD5A1, SRD5A2 and SRD5A3)(26)

Specific substrates include testosterone, progesterone, androstenedione,[9] epitestosterone, cortisol, aldosterone, and deoxycorticosterone. Outside of dihydrotestosterone, much of the physiological role of 5α-reduced steroids is unknown .Beyond reducing testosterone to dihydrotestosterone, 5alpha-reductase enzyme isoforms I and II reduce progesterone to dihydroprogesterone (DHP) and deoxycorticosterone to dihydrodeoxycorticosterone (DHDOC).

5α-Reductase is most known for converting testosterone, the male sex hormone, into the more potent dihydrotestosterone. Metabolism of androgens is expressed in the following figure:

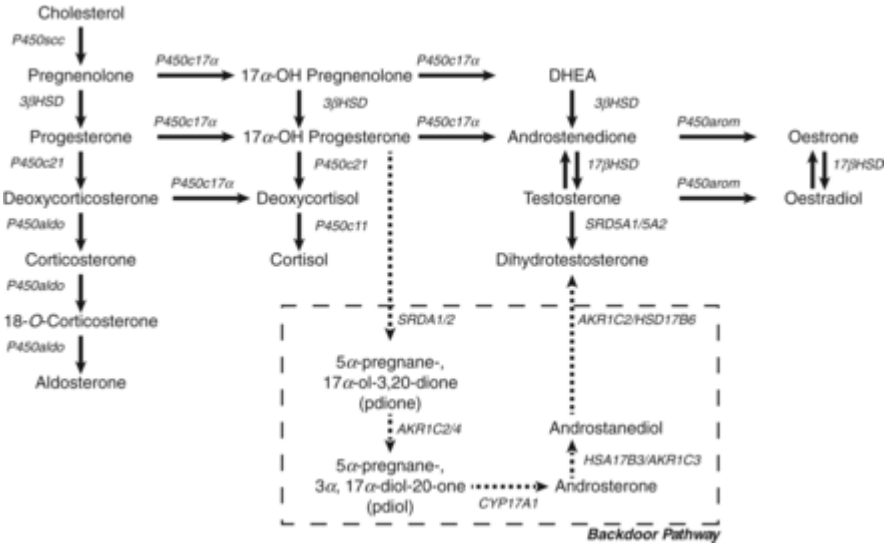

Figure 1.

Biosynthesis of androgens.

The mechanism of action of 5-alpha reductase is complex but its activity is regulated by the action of sulphite dihidroepiandrostenedion DHEA and progesterone that inhibit the action of this enzyme while activity of 5 alpha reductase is increased by obesity and insulin resistance in females and men. Increased activity in women is associated with polycystic ovary and hirsutism and I men is associated with hypertrophy of the prostate. (1)
The disorders of activity to 5-α reductase will be expressed with increase or decrease to the action of DHT in peripheral tissues.

1.1 5-Alpha Reductase Deficiency

5-alpha-reductase deficiency (5-ARD) is an autosomal recessive sex-limited condition resulting in the inability to convert testosterone to the more physiologically active dihydrotestosterone (DHT). Because DHT is required for the normal masculinization of the external genitalia in utero, genetic males with 5-alpha-reductase deficiency are born with ambiguous genitalia (ie, 46,XY DSD).(2,12)

Patients with 5-alpha-reductase deficiency classically present with striking ambiguity of the genitalia, with a clitoral-like phallus, markedly bifid scrotum, pseudovaginal perineoscrotal hypospadias, and a rudimentary prostate.(13)

In this condition, a person with male (XY) chromosomes has a body that appears female before puberty. After puberty begins, other testosterone-activating enzymes become available and the body soon takes on a masculine appearance, Children with 5-alpha reductase deficiency are often raised as girls. About half of these individuals adopt a male gender role in adolescence or early adulthood.(9,12)

The conversion of testosterone to DHT is essential for the formation of the male phenotype during embryogenesis. 5-alpha-reductase type 2 deficiency is characterized by normal levels of testosterone l and low levels of DHT (the ratio T / DHT is increased) .

1.2 Hirsutism

Hirsutism is defined as the growth of hair in women on the face and body which appears in the same pattern and with the same temporal development as in men. Such hair growth in women is caused either by an underlying state of androgen overproduction, possibly due to body mass, or enhanced local sensitivity to androgens. Studies looking at differential expression of 5*α* reductase isoenzymes in hair follicles have been hampered by detection of 5*α* reductase type 2 which is constitutively expressed in the hair follicle *in vivo* but not during *in vitro* culture. (16)

The majority of women with hirsutism with or without the PCOS have normal concentrations of circulating androgen hormones, yet increased 5*α* reductase activity has been demonstrated in ovarian tissue and in the urine metabolites of women with PCOS. The concentrations of DHT in serum from female patients are at the lower limit of currently available immunoassays and, therefore, distinguishing between possible normal and pathogenic concentrations is not possible. Newer mass spectrometric methods may enable this differentiation but currently DHT is not recommended as part of the investigations for PCOS.

1.3 Polycystic ovary syndrome

Polycystic ovary syndrome (PCOS) is a set of symptoms due to elevated androgens (male hormones) in women. Signs and symptoms of PCOS include irregular or no menstrual periods, heavy periods, excess body and facial hair, acne, infertility .Associated conditions include type 2 diabetes, obesity, obstructive sleep apnea, heart disease, mood disorders, and endometrial cancer.(3,4)

PCOS is due to a combination of genetic and environmental factors. Risk factors include obesity, not enough physical exercise, and a family history of someone with the condition. Diagnosis is based on two of the following three findings: no ovulation, high androgen levels, and ovarian cysts. Cysts may be detectable by ultrasound. Other conditions that produce similar symptoms include adrenal hyperplasia, hypothyroidism, and hyperprolactinemia.(10)

2.Materials and Methods

We measured total levels of testosterone and DHT in 100 male patients The patient was divide in 3 grups, neonate, childhood, puberty. In order to reflect 5alpha-reductase activity TT/DHT ratio was calculating .
In 70 cases the level of TT and DHT was measured before and after HCG stimulation. HCG is a double polypeptide hormone and shares a common subunit with LH. It stimulates testicular Leydig cells to secrete androgens via the LH receptors. A single injection of hCG is adequate as it has a long half life (2.5 days) and produces a progressive but modest rise in plasma testosterone for 72-120 hours.Procedure:

1. day 0: take blood for testosterone and dihydrotestosterone
2. administer 1500 units (infants) or 5000 units (over 2 years) hCG subcutaneously or im
3. day 4: take blood for testosterone and dihydrotestosterone

3.Results

In serum of patients was measured level of TT,DHT and TT/DHT ratio was calculating to measure 5alpha-reductase activity before and after HCG stimulation.

In serum of patient in childhood was measured level of TT (mean ± sd, 0.81±0.11), DHT (mean ± sd, 16.4±4.3) and TT/DHT ratio (mean ± sd, 5.4±0.55). After HCG stimulation TT and DHT levels rose respectively TT (mean ± sd 1.43±0.34), DHT (mean ± sd 29.4±6.5) and TT/DHT ratio (mean ± sd 11.8±1.3).

There was a parallel rise in DHT and T to pubertal levels.Both DHT and TT rose significantly ($P < 0.01$) in puberty boys and the TT/DHT ratio was (mean ± sd 6.7 ± 3.5) and after HCG stimulation (mean ± sd 9.1±5.4).

In 4 neonate male patient with with ambiguous genitalia TT,DHT and TT/DHT ratio was calculating before and after HCG stimulation.In 2 neonate we observed a high level of TT and DHT , and TT/DHT ratio was normal(<8.5) and in 2 other cases the level of TT and DHT was normal. After the HCG stimulation we observed an increase in the level of TT in all cases and in 2 of them we had a TT/DHT ratio >27.

4.Conclusion

5alpha-reductase has a role in various pathologies and the clinical indications for its measure activity are the diagnosis of 5*α*-reductase deficiency in an infant with ambiguous genitalia in a male with delayed puberty and/or undescended testes and to confirm the presence of active testicular tissue, all in combination with an hCG stimulation test. In women is more importane in hirsutism and PCOS.

The hCG stimulation test has three main indications in the differential diagnosis of DSD: (1) to confirm the presence of active testicular tissue, (2) to aid in the diagnosis of a male with delayed puberty and/or undescended testes and (3) to aid diagnosis in an infant with ambiguous genitalia and palpable gonads. The test is recommended for the diagnosis of 5*α* reductase deficiency.

In normal childhood males, TT and DHT rise in parallel, and we have a constant TT/DHT ratio. It is a significant rise in the T/DHT ratio in puberty. After HCG stimulation TT , DHT, and the TT/DHT ratio rose significantly.

T/DHT ratio can help in investigation of 46, XY patients with ambiguous genitalia and normal testosterone synthesis. T/DHT ratio can be used to select newborns affected by 5alpha-reductase deficiency.In 5-alpha Reductase deficiency TT/DHT ratio is higher than normal male and rose significantly after HCG stimulation and the diagnosis is decide with an TT/DHT ratio >27.

5α- reductase deficiency should be included in the differential diagnosis of all newborns with 46,XY DSD with normal testosterone production.

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