

## 21-Hydroxylase Deficiency

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### Abstract:

21-hydroxylase deficiency is one among a group of problems known as congenital adrenal hyperplasias that impair hormone production and disrupt sexual improvement. 21-hydroxylase deficiency is liable for about ninety five percent of all instances of congenital adrenal hyperplasia. 17-Hydroxyprogesterone (17-OHP) may be converted to dihydrotestosterone (DHT) through an opportunity “backdoor” route that bypasses the traditional intermediates androstenedione and testosterone. on this backdoor pathway, 17-OHP is transformed to 5 $\alpha$ -pregnane-3 $\alpha$ ,17 $\alpha$ -diol-20-one (pdiol), that's an wonderful substrate for the 17,20 lyase activity of CYP17A1 to provide androsterone.

### Objective and Hypotheses:

The goal of this observes become to attain proof for the presence of the backdoor pathway in sufferers with 21-hydroxylase deficiency (21-OHD).

### Methods:

We as compared urinary steroid hormone profiles determined by fuel chromatography-mass spectrometry of 142 untreated 21-OHD sufferers (age range, 1 d to 25.four year; fifty one men) with 138 control topics. The hobby of the backdoor pathway become assessed the usage of the ratios of the urinary concentrations of pdiol to those of the metabolites of the traditional  $\Delta$ four and  $\Delta$ 5 pathways. In evaluation to etiocholanolone, which originates nearly completely from the conventional pathways, androsterone may be derived moreover from the backdoor pathway. therefore, the androsterone to etiocholanolone ratio can be used as a hallmark for the presence of the backdoor pathway.

The main route of androgen synthesis in humans is the traditional  $\Delta$ 5 pathway. In this pathway, the 17,20 lyase pastime of CYP17A1 ends in the conversion of 17-hydroxypregnenolone to dehydroepiandrosterone (DHEA). DHEA is then metabolized to androstenedione ( $\Delta$ 4A) and testosterone (T). In androgen goal tissues, T is converted by means of five $\alpha$ -reductase to the stronger androgen receptor agonist dihydrotestosterone (DHT). So-known as “susceptible” androgens, together with  $\Delta$ 4A, characteristic as androgens only to the extent that they're transformed to T or DHT. The five $\alpha$ -reduction of T is, but, not the simplest biosynthetic pathway to supply DHT. Within the testes of tamar wallaby pouch younger, DHT may be correctly fashioned from the five $\alpha$ -decreased 21-carbon (C21) precursor five $\alpha$ -pregnane-3 $\alpha$ ,17 $\alpha$ -diol-20-one (pdiol) through an opportunity so-called “backdoor” pathway. In wallaby pouch young testes, 17-hydroxyprogesterone (17-OHP) can be transformed to pdiol via sequential activities of 5 $\alpha$ -reductase and reductive 3 $\alpha$ -hydroxysteroid dehydrogenase (HSD). Human CYP17A1 has a high affinity for pdiol, that's an extraordinary substrate for its 17,20 lyase activity, leading to androsterone generation Pdiol is

a better substrate for CYP17A1 than 17-OHP and even the favored substrate 17-hydroxypregnenolone. Additionally, 17-OHP is advanced to T as a substrate for five $\alpha$ -reductase (eight). Hence, the backdoor pathway may also provide a more green route to DHT underneath situations of huge 17-OHP accumulation as in 21-hydroxylase deficiency (21-OHD) (four). 21-OHD is the maximum common form of congenital adrenal hyperplasia and is caused by mutations inside the CYP21A2 gene (nine). CYP21A2 converts 17-OHP to eleven-deoxycortisol and progesterone to deoxycorticosterone, each of which are precursors for cortisol and aldosterone biosynthesis, respectively. It has been speculated that the backdoor pathway could be involved in androgen synthesis in patients with 21-OHD; however, no facts have yet been published supporting this hypothesis.

### Pathways of internal secretion synthesis:

In regular physiology, the principal course for androgenic hormone synthesis is the pathway from 17-hydroxypregnenolone to DHEA. Within the chance backdoor pathway in 21-OHD, the conversion of 17-OHP to five $\alpha$ -pregnane-17 $\alpha$ -ol-3,20-dione (pdione) is mediate with the help the help kind one or two (SRDA1 or SRDA2). it has been proved that the type the type isoform (encoded via SRD5A1) is responsible for androstanediol producing in immature mouse testes through the backdoor path. In assessment, best the Type 2 isoenzyme (encoded with the help of SRD5A2) become detectable in craniate sex organ skin. In differentiation, it's been valid recently that SRD5A2 is expressed at low degrees in craniate testes but is very expressed in the flesh testes, which SRD5A1 expression is higher in craniate than grownup testes. But, it is not however incontestable that isoenzyme is accountable for accountable for 17-OHP to pdione in patients with 21-OHD. eventually, pdione is regenerate to pdiol by subtractive subtractive. In people, kind one one (AKR1C4), that is expressed dominantly within the liver but to boot within the craniate and person adrenal, is that the main subtractive. It's been valid that sort 3 3 (AKR1C2) to boot catalyzes the discount of pdione to pdiole, and also the primary forty six, XY disorders of sexual development patient with a AKR1C2 mutation has been recently mentioned. As a result of AKR1C2 participates simplest within the chance backdoor but no longer within the standard front door pathway, it has been suggested that the backdoor pathway is needed for normal human male sexual differentiation. AKR1C2 is wide expressed and is determined in liver, lung, testes, and adrenal tissues. The following step at the backdoor path to DHT is that the cleavage of the C21 steroid pdiol to form the C19 steroid androgenic hormone by the seventeen, 20-lyase interest of CYP17A1. Subsequent, androgenic hormone is reduced to androstanediol by the kind the kind (HSD17B3), that is dominantly expressed

among the testes. besides this protein, sort two (AKR1C3; to boot observed as sort five 3) indicates a pattern of recreation a bit like that of the sex gland kind 3  $17\beta$ -HSD, and to boot reduces androgenic hormone to androstenediol with excessive chemical change performance in humans. AKR1C3 is expressed in uncountable distinctive tissues that embrace liver and also the adrenals and is answerable for the formation of androgens in women. The three $\alpha$ -oxidation of androstenediol to the spirited internal secretion DHT is mediate by suggests that of AKR1C2 in vitro. However, it's miles endorsed that AKR1C2 works generally as a subtractive protein among the backdoor pathway. The three-hydroxypimerase (additionally observed as retinol/sterol dehydrogenase encoded via HSD17B6) to boot oxidizes androstenediol to DHT. Therefore, the adrenals specific all needed enzymes for the conversion of pdione to DHT. The urinary metabolites of the standard pathways ar proved with the help of dotted strains. Remember that androgenic hormone may be derived from every the standard and backdoor pathways, whereas etiocholanolone is generated nearly entirely from the standard pathway. doc, 11-Deoxycorticosterone; 11OH-An, eleven-hydroxyandrosterone; AKR, aldo-keto-reductase.

The goal of this verify become to get proof for the presence of the backdoor pathway in 21-OHD through the retrospective analysis of urinary internal secretion profiles during a immense cohort of sufferers.

#### Outcomes:

Untreated 21-OHD subjects confirmed elevated urinary quantitative relations of pdiol to the  $\Delta 4$  and  $\Delta 5$  pathway metabolites and an improved androgenic hormone to etiocholanolone ratio.

#### Conclusions:

The extended quantitative relations of pdiol to the  $\Delta 4$  and  $\Delta 5$  pathway metabolites moreover because the higher androgenic hormone to etiocholanolone ratio in sufferers with 21-OHD indicate postnatal recreation of the backdoor pathway with most interest for the period of early infancy. Our records supply new insights into the pathophysiology of androgenic hormone synthesis of 21-OHD.

The pee samples of the manage subjects additional than one time period ( $n = 60$ ) had been received from the DONALD (Dortmund dietary and measure long ways Designed) observe, as delineate somewhere else. The median age of these controls become eight.6 twelve months, with twenty fifth and seventy fifth percentiles of seven. Zero and eleven. Four twelve months, severally. Those subjects are chosen to be age- and sex-matched to the 21-OHD sufferers WHO were bigger than one twelve month's vintage, having a median age of eight. Three time period with twenty fifth and seventy fifth percentiles of half-dozen.6 and eleven. Five twelve months, severally. The pee samples of the controls aged but one twelve months had been obtained from our own pool of standard patients. Urinary internal secretion concentrations are determined with the help

of GC-MS analysis of 24-h or random spot pee samples as delineate antecedently (14, 16). In quick, unfastened and conjugated urinary steroids are extracted by means of strong-section extraction (Sep-Pak C18 cartridge; Waters associates, Milford, MA). The conjugated steroids were enzymatically hydrolyzed (Sigma sort I pulverised Helix pomatia; letter Chemical Co., St. Louis, MO) and recovered with the help of Sep-Pak extraction. recognized quantities of three inner necessities (five $\alpha$ -androstane-3 $\alpha$ ,17 $\alpha$ -diol, stigmaterol, and cholesteryl butyrate) had been delivered to an element of every extract prior formation of methyloxime-trimethylsilyl ethers. petrol natural action become dead the usage of associate Optima-1 amalgamate silicon oxide column with argonon because the carrier petrol at a waft rate of one ml/min. initial kitchen appliance temperature become eighty C, control {for two}for two} min; the temperature became then elevated through twenty C/min to 100 ninety C (1 min) and so dilated by 2.5 C/min to 272 C. The fuel chromatograph (Agilent 6890 series GC; Agilent 7683 assortment machine-controlled liquid sampler; Agilent technology, city, CA) was immediately interfaced to a mass selective detector (Agilent 5973N mass selective detector) operated in elect particle chase mode. Standardization of the GC-MS becomes achieved by victimization analysis of a reference combination containing acknowledged quantities of all of the separation compounds. When standardization, values of man or girl steroids had been determined by activity the chosen particle prime regions against the inner necessities.

After the first year of existence, the androgenic flux in the conventional pathways now not favors the  $\Delta 4$  pathway in sufferers with 21-OHD. The hobby of the 17, 20 lyase reaction of CYP17A1 for the classic pathways is stronger by using the expression of cytochrome b5 on the onset of adrenarche. Adrenarche is a continuous developmental technique, beginning at least as early as three year of age.

Consequently, the alternate in androgenic flux in sufferers with 21-OHD will be because of an improved expression of cytochrome b5 with increasing age. Because the  $\Delta 4$  pathway has a low  $V_{max}$  for the 17,20 lyase response, a similarly enhancement of the 17,20 lyase hobby might lead to a greater marked increase within the Five pathway hobby.

Concerning our results, our records were presented according to age businesses. Because it is much more likely that older sufferers on the time of the diagnosis had a milder shape of 21-OHD, this could have encouraged the consequences. for instance, the decrease androsterone to etiocholanolone ratio in older patients can be a effect of a milder deficiency of the 21-hydroxylase in these subjects. Furthermore, due to the fact that is a postnatal analysis, it cannot provide statistics about the position of the backdoor pathway in prenatal existence. Due to the fact person adrenals explicit markedly more AKR1C2 than fetal adrenals, it's far suggested that the alternative backdoor pathway may not be a massive pathway in the fetal adrenal

however might be essential inside the grownup adrenal in patients with 21-OHD. However, besides the low expression of AKR1C2, fetal adrenals explicit ample AKR1C1, AKR1C3, and AKC1C4.

In summary, this have a look at demonstrates for the primary time and in a large cohort of patients that the opportunity backdoor pathway exists in 21-OHD patients. 21-OHD subjects have higher urinary concentrations of metabolites of the backdoor pathway, and they have an increased pathway activity, in both absolute and relative terms, in comparison with the traditional pathway activities. Therefore, the alternative backdoor pathway ought to contribute to androgen biosynthesis in patients with 21-OHD. Because the metabolites of the alternative backdoor pathway aren't measured in plasma samples for the duration of habitual tracking, the backdoor pathway may additionally explain divergences between virilization and androgen tiers visible in clinical exercise. Our facts are critical for the knowledge of androgen biosynthesis and virilization in sufferers with 21-OHD and provide new insights into the pathophysiology of 21-OHD.