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Molecular Genetics of Androgenic 17β-Hydroxysteroid Dehydrogenases

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17β-Hydroxysteroid dehydrogenase (17β-HSD) type 2 catalyzes the NAD$^+$-dependent oxidation of androgens, estrogens and progestins, predominantly in the secretory endometrium, placenta, liver and small intestine. 17β-HSD type 3 catalyzes the NADPH-dependent conversion of androstenedione to testosterone in the testis, and the genetic disease 17β-HSD deficiency is caused by mutations in the 17β-HSD3 gene.

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INTRODUCTION

17β-Hydroxysteroid dehydrogenase (17β-HSD) is an enzyme which catalyzes the oxido/reduction at carbon 17 of C-18 and C-19 steroids. Based on substrate specificity studies with organ cultures and tissue homogenates, it was suggested many years ago that different isozymes of 17β-HSD exist. Human tissues that have been thoroughly characterized in respect of 17β-HSD enzyme activities are endometrium, placenta and testis.

MICROSOMAL 17β-HSD OF HUMAN ENDOMETRIUM AND PLACENTA

Tseng and Gurpide [1] were the first to show that the NAD$^+$-dependent oxidation of estradiol to estrone and 20α-dihydroprogesterone to progesterone in human endometrium is much higher in tissue from the luteal phase of the menstrual cycle (secretory endometrium), i.e. during the time of high plasma progesterone levels, as compared to tissue from the follicular phase (proliferative endometrium). Consequently, in experiments with cultured endometrial cells it was shown that addition of progesterone or synthetic progestins to the tissue culture medium causes a striking increase in the oxidative 17β/20α-HSD activities, using estradiol [2] as well as testosterone, 5-androstene-3β,17β-diol and 20α-dihydroprogesterone as substrates [3]. It was proposed that the parallelism in enhancement of 17β/20α-HSD activities in response to the progestins *in vitro* could be due to induction of a single enzyme that catalyzes both the 17β-HSD and 20α-HSD activities. Also, the enzyme induction of endometrial 17β-HSD is also observed *in vivo* in women treated with synthetic progestins [4]. Furthermore, Pollow and colleagues reported on partial purification and characterization of human endometrial 17β-HSD, and showed that the enzyme is membrane bound, preferentially utilizes NAD$^+$ as cofactor, and catalyzes the oxidative reaction with estradiol and testosterone as substrates [5, 6].

The placenta was shown to have a 17β-HSD enzyme similar to the enzyme in the endometrium. The enzymology of this enzyme has been extensively characterized by Blomquist and colleagues [7], who presented strong evidence that the microsomal 17β-HSD of the human placenta also possesses 20α-HSD activity, and utilizes NAD$^+$ as cofactor. In addition to this microsomal enzyme, placenta was also shown to contain a soluble 17β-HSD, termed estradiol 17β-dehydrogenase because of its preference for estrogens as substrates [7–9]. The estradiol 17β-dehydrogenase was the first 17β-HSD to be purified to homogeneity and cloned, herein designated 17β-HSD type 1 [10–12].

The microsomal 17β-HSD of human endometrium and placenta, designated 17β-HSD type 2, was recently cloned by expression cloning, and found to be equally active on androgens and estrogens as substrates. The recombinant 17β-HSD type 2 catalyzes the oxidative reaction, i.e. the inactivation of estradiol to estrone and testosterone to androstenedione. In addition, the type 2 isozyme efficiently catalyzes the conversion of the inactive progestin, 20α-hydroxyprogesterone, to the active progestin, progesterone, i.e. the enzyme possesses 20α-HSD activity. The amino acid sequence of 17β-HSD type 2 indicates an extended hydrophobic amino terminus of approx. 60 amino acids, which is characteristic of a transmembrane signal anchor. Cell

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fractionation experiments with recombinant protein demonstrate association of the enzyme with the membranes of the endoplasmic reticulum [13]. Also, in consonance with earlier observations of high enzyme activities in placenta and secretory endometrium, the mRNA encoding 17β-HSD type 2 is present at high levels in these tissues. High levels of type 2 mRNA are also found in the liver and small intestine, tissues that are known to inactivate steroid hormones [14].

TESTICULAR 17β-HSD

Testis has been shown to contain a unique isoform of a microsomal 17β-HSD that shows high specificity for NADPH as cofactor [15]. The physiological substrate for the testicular 17β-HSD is androstenedione, i.e. the enzyme catalyzes the reductive reaction. Over 20 years ago, Saez and colleagues [16, 17] reported on male pseudohermaphroditism caused by deficiency of the testicular 17β-HSD enzyme, consequently this inborn error of metabolism was termed 17β-HSD deficiency. This rare disease is inherited in an autosomal recessive fashion and approx. 50 cases have been described. The phenotype of afflicted subjects is normal Wolffian duct-derived genitalia but external genitalia female in character (blind-ending vagina), and strikingly elevated post-pubertal plasma levels of androstenedione [18].

The gene encoding the testicular 17β-HSD which catalyzes the 17β-reduction of androstenedione to testosterone, designated 17β-HSD type 3, was recently cloned. The recombinant enzyme utilizes NADPH as cofactor and is associated with the microsomal fraction of transfected cells. It is also established that 17β-HSD deficiency is caused by mutations in the gene encoding the 17β-HSD type 3 enzyme [19]. To date, we have identified 14 different mutations in the 17β-HSD3 gene in 17 unrelated subjects with 17β-HSD deficiency. The mutations characterized include ten missense mutations, three splice junction abnormalities, and one small deletion that results in a frame-shift [20]. A puzzling clinical feature of this disorder is that the male internal structures are apparently normal. It is conceivable that isozyme(s) other than 17β-HSD type 3 may locally convert testes-derived androstenedione to testosterone in the anlage of the internal genitalia. Alternatively, androstenedione may act by itself as an androgen in the Wolffian ducts during fetal development.

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