0021-972X/99/$03.00/0
The Journal of Clinical Endocrinology & Metabolism

Vol. 84, No. 9
Printed in U.S.A.

Metabolism of Dihydrotestosterone in Human Liver: Importance of 3α- and 3β-Hydroxysteroid Dehydrogenase*

EDYTA C. PIROG AND DELWOOD C. COLLINS

Department of Pathology, Cornell University School of Medicine and Veterans Administration Medical Center, and the Department of Obstetrics and Gynecology, University of Kentucky College of Medicine, Lexington, Kentucky 40536

ABSTRACT

This study compared the enzyme activity of 3α-hydroxysteroid dehydrogenase (3αHSD) and 3β-hydroxysteroid dehydrogenase (3βHSD) in the human liver. 3αHSD was found in both microsomal and cytosolic liver fractions. Contrary to that in rat liver, microsomal 3αHSD activity was 12-fold higher than cytosolic 3αHSD activity, and 3αHSD was not inhibited by indomethacin (10 μmol/L). The rate of 5α-dihydrotestosterone (DHT) reduction to 5α-androstane-3α,17β-diol (3αDIOL) by 3αHSD was 2 times higher than the rate of 3αDIOL oxidation to DHT. 3βHSD was present primarily in the microsomal fraction of the human liver, and the rate of DHT reduction to 5α-androstane-3β,17β-diol (3βDIOL) by 3βHSD was 3 times higher than the rate of 3βHSD oxidation to DHT. When 3αHSD and 3βHSD activities were compared, the rate of DHT reduction by 3βHSD was 3-fold lower than the rate of DHT reduction by 3αHSD. No sex or age differences were found in either 3αHSD or 3βHSD activity. As the activity of DHT-metabolizing enzymes is not sex dependent, the sex differences in plasma levels of 3αDIOL glucuronide probably reflect differences in DHT production rather than in DHT metabolism. Comparison of the activities of 3αHSD, 3βHSD, and androgen UDP-glucuronyl transferase suggests that the major pathway of DHT metabolism in human liver involves 3αHSD reduction in the liver, followed by subsequent glucuronidation and clearance via the kidney. (*J Clin Endocrinol Metab* **84:** 3217–3221, 1999)

THE SYNTHESIS and metabolism of an active androgen, 5α-dihydrotestosterone (DHT), takes place in extragonadal, androgen target tissues. For this reason, DHT is considered to be a paracrine or an autocrine hormone. DHT is synthesized from testosterone in an irreversible reaction catalyzed by the microsomal enzyme, 5α-reductase. Modification of DHT at the 17β-hydroxy or 3-keto position renders it inactive (Fig. 1). The reactions carried out by DHT-metabolizing enzymes are reversible, and the relative activities of these enzymes determines tissue exposure to the active androgen, DHT. One of the enzymes involved in DHT metabolism is 3α-hydroxysteroid dehydrogenase (3αHSD), which reduces DHT to 5α-androstane-3α,17β-diol (3αDIOL). Interestingly, 3αHSD activity favors net production of DHT in certain tissues, such as the prostate (1). Inadequate metabolism of DHT by 3αHSD may contribute to the development of prostate hyperplasia (1). Skin fibroblasts show different patterns of 3αHSD activity in different skin areas (2). In nongenital skin, DHT formation from 3αDIOL was approximately equal to DHT reduction to 3αDIOL (2). However, in genital skin, the rate of DHT formation from 3αDIOL was twice as high as DHT reduction (2). Little information is available about the regulation of 3αHSD activity in other tissues. It is tempting to speculate that abnormal DHT metabolism may contribute not only to disorders such as prostate hyperplasia, but also to other androgen-related disorders such as acne and hirsutism.

Other enzymes involved in DHT metabolism are 3βHSD, which reduces DHT to 5α-androstane-3β,17β-diol (3βDIOL) and microsomal UDP-glucuronyl transferase (UDPGT), which conjugates DHT to glucuronic acid. This reaction can be reversed by another enzyme, β-glucuronidase. The relative contributions of the above-mentioned enzymes to the overall metabolism of DHT are poorly understood.

The equilibrium of reductive and oxidative activities of the hydroxysteroid dehydrogenases may be important in the regulation of intracellular levels of DHT. This study was designed to determine the activity and kinetic properties of 3αHSD and 3βHSD in the human liver. In addition, the activities of these enzymes were measured in liver homogenates from five females and five males to determine whether there are sex- or age-specific differences in the expression of these enzymes in the liver.

Subjects and Methods

Subjects

Liver samples from female (34–60 yr) and male (33–75 yr) organ donors were obtained from the National Disease Research Interchange (Philadelphia, PA). Samples were procured postmortem (within 5 h), snap-frozen upon removal, and stored at −70 C until used.

Chemicals

The radioactive androgens, [1,2-^{3}H]3α-DIOL (SA, 30.5 Ci/mmol) and [1,2,4,5,6,7-^{3}H]DHT (SA, 119.6 Ci/mmol) were purchased from Amersham Pharmacia Biotech (Arlington Heights, IL). The nonradioactive steroids were obtained from Sigma Chemical Co. (St. Louis, MO) or Steraloids, Inc. (Wilton, NH). The purity of each androgen was con-

Received January 28, 1999. Revision received May 19, 1999. Accepted May 21, 1999.

Address all correspondence and requests for reprints to: Dr. D. C. Collins, 204 Health Sciences Research Building, University of Kentucky Medical Center, Lexington, Kentucky 40536-0305.

* This work was supported by NIH Grant R01-DK-41879 and the Department of Veterans Affairs.

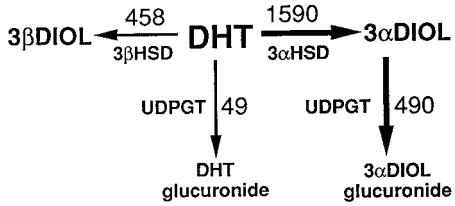

FIG. 1. Major pathways for DHT metabolism leading to glucuronyl metabolites. Minor pathways leading to sulfonated metabolites are not shown.

firmed by high pressure liquid chromatography. The protein assay reagents were obtained from Bio-Rad Laboratories, Inc. (Richmond, CA). All other chemicals were analytical grade.

Tissue fractionation

Liver samples were homogenized with a Polytron homogenizer (Brinkmann Instruments, Inc., Westbury, NY) in 4 vol ice-cold 50 mmol/L phosphate buffer, pH 7.4, containing 250 mmol/L sucrose and 0.5 mmol/L dithiothreitol. Aliquots of the liver homogenates were taken for analysis of 3αHSD and 3βHSD activities. The homogenates were centrifuged for 15 min at 12,000 × *g*, followed by ultracentrifugation of the supernatant at 100,000 × *g* for 60 min. The 100,000 × *g* pellets were rinsed once with 250 mmol/L sucrose and 50 mmol/L phosphate buffer, pH 7.4, and re-homogenized in 250 mmol/L sucrose and 50 mmol/L phosphate buffer, pH 7.4, with 20% glycerol. The microsomal and cytosolic fractions were stored at −80 C for up to 2 months with no significant change in enzyme activity. At the time of analysis, the viability of the microsomal fractions was confirmed by determining the activity of the microsomal marker enzyme, glucose-6-phosphatase, as described by Aronson and Touster (3), using 100 μmol glucose-6-phosphate as a substrate.

Enzyme kinetics

The enzyme activity studies were performed using liver homogenates or subcellular fractions. The K_m and V_{max} values were determined in the microsomal and cytosol fractions using the Michaelis-Menten equation with a nonlinear regression data analysis software program (Enzfitter 1.05, Biosoft, 1987). 3αHSD activity was determined using tritiated substrates (DHT or 3αDIOL). The incubation mixture (600 μL) contained 150,000 dpm of either [^{3}H]DHT or [^{3}H]3αDIOL and cold androgens (final concentration, 0.1–20 μmol/L), an aliquot of the cell fraction (100 μg total homogenate protein, 150 μg cytosol protein, or 50 μg microsomal protein), 200 μmol/L NADP or NADPH, and 0.05% Tween-80 (vol/vol) in 25 mmol/L phosphate buffer, pH 7.4. The androgens were added to the tubes in ethanol solution before the assay, and the ethanol was evaporated before the other components of the incubation mixture were added. Control incubations were carried out without adding the cofactors, NADP or NADPH. Samples were incubated at 37 C in a shaking water bath for 0.5–5 min. DHT or 3αDIOL was used as substrate at a concentration of 50 nmol/L (100 times the physiological concentration). The conditions of the assay used both cofactor (NADPH or NADP) and substrate at excess concentrations to ensure that product formation reflected maximal 3αHSD activity and was linear with time and protein concentration.

The reaction products were extracted from the incubation mixture using reverse phase C_{18} SPC columns. Aliquots of the incubation mixture (150 μL) were applied to the columns and eluted with 1.5 mL water, 0.5 mL hexane, and finally, 1.5 mL ethanol. All of the radioactivity was recovered in the ethanol eluate. The ethanol fractions were evaporated using a Speed-Vac evaporator (Savant Instruments, Farmingdale, NY), reconstituted in ethanol containing cold tracer (100 μg each of DHT, 3αDIOL, and 3βDIOL), and separated by high pressure liquid chromatography (Waters, Millipore Corp., Millford, MA) using a C_{18} column (Waters, Millipore Corp.) and a mobile phase of acetonitrile-water (45: 55) at a flow rate of 1 mL/min. The retention times for the androgens were: 3αDIOL, 9.46; 3β-DIOL, 7.32; and DHT, 10.95. The radioactivity in the eluted samples was quantitated using a Radio-Chromatography Detector (Radiomatic FLO-ONE Beta, Series A-500, Radiomatic Instruments & Chemical Co., Inc., Meriden, CT) and analyzed with the Radiomatic FLO-ONE Software program (Radiomatic Instruments & Chemical Co., Inc.).

3βHSD activity was determined as described above, except that 3βDIOL was the substrate for the oxidative reaction, and 150 μg microsomal protein, 450 μg cytosolic protein, or 100 μg total homogenate protein were used in the incubation.

TABLE 1. Michaelis-Menten constants (K_m), maximal velocities (V_{max}), and V_{max}/K_m ratios of 3αHSD and 3βHSD in liver microsomes and cytosol of a 65-yr-old Caucasian male

Enzyme	Substrate	K_m (μmol/L)	V_{max} (nmol/min·mg)	V_{max}/K_m ratio
3αHSD				
Microsomes				
Reduction	DHT	2.2	2.50	1.136
Oxidation	3αDIOL	2.4	1.57	0.654
Cytosol				
Reduction	DHT	2.2	0.20	0.091
Oxidation	3αDIOL	2.5	0.21	0.084
3βHSD				
Microsomes				
Reductive	DHT	1.13	0.37	0.327
Oxidative	3βDIOL	3.35	0.39	0.116
Cytosol				
Reduction	DHT	ND	ND	ND
Oxidation	3βαDIOL	ND	ND	ND

DHT was added in excess as the substrate for reduction, and 3αDIOL or 3βDIOL was added in excess as the substrate for oxidation. ND, Not detectable.

Protein determination

Protein concentrations of the homogenates and cellular fractions were determined as described by Bradford (4), using BSA as a standard.

Results

Kinetic characteristics of 3αHSD and 3βHSD in liver

The K_m and maximum velocity (V_{max}) values of 3αHSD and 3βHSD for both the reduction of DHT and the oxidation of 3αDIOL and 3βDIOL in the microsomes and cytosol are shown in Table 1 for a 65-yr-old Caucasian male. A similar pattern of K_m constants and reductive and oxidative activities was noted in the other samples that were fractionated. 3αHSD activity was expressed in both the microsomal and cytosolic fractions. The V_{max} value was 12-fold higher in the microsomes than in the cytosol for both the oxidative and reductive reactions. The K_m values for DHT and 3αDIOL were similar in both cellular fractions. The V_{max}/K_m ratio represents the relative reaction velocity (see Table 1), assuming that the initial concentrations of the substrates, DHT and 3αDIOL, are equal (see Table 1). This is, in fact, the physiological situation, as plasma levels of DHT and 3αDIOL are approximately equal (5). The relative rate of DHT reduction was 2 times the relative rate of 3αDIOL oxidation in the microsomes. In cytosol, the V_{max}/K_m ratios for reduction and oxidation were similar. Neither microsomal nor cytosolic 3αHSD activity was inhibited by indomethacin at a concentration as high as 10 μmol/L (data not shown).

3βHSD activity was present mainly in the microsomal fraction of the liver. The K_m and V_{max} values of microsomal 3βHSD for reduction of DHT to 3βDIOL and oxidation of 3βDIOL to DHT are shown in Table 1. 3βHSD activity in the cytosol was below the assay detection limit. DHT reduction

to 3βDIOL was 3 times faster than DHT formation from 3βDIOL. In the microsomes, the relative rate of DHT reduction by 3βHSD was 3-fold lower than the relative rate of DHT reduction by 3αHSD. This suggests that the major product to DHT metabolism in the human liver is 3αDIOL.

Activity of 3αHSD and 3βHSD in liver homogenates from males and females

3αHSD activity in liver homogenates from five male and five female subjects is shown in Fig. 2. In all subjects, the velocity of DHT reduction to 3αDIOL was 2 times higher than the velocity of 3αDIOL oxidation to DHT. There was no significant difference between the mean rate ± SE for 3αHSD reduction in men (38.5 ± 16.9 pmol/min·mg protein) and women (38.3 ± 10.6; Table 2). 3αHSD activity (DHT reduction) was 9.4–103.7 pmol/min·mg for men and 10.0–73.1 pmol/min·mg for women. Furthermore, no significant correlation was found between 3αHSD activity and the age of the subjects.

As suggested by the results in Table 1, the enzyme activity of 3βHSD was about 10-fold lower than that of 3αHSD and was near the sensitivity of the assay for the oxidation of 3βDIOL to DHT. The mean 3βHSD activity in liver homogenates for DHT reduction in men (8.71 ± 2.3 pmol/min·mg protein; range, 6.4–12.6) was not significantly different from the mean activity in women (6.78 ± 1.7 pmol/min·mg protein; range, 4.4–8.9; Table 2). In addition, there was no significant correlation between the age of the subjects and 3βHSD activity.

Discussion

The need to understand the pathways for metabolism of DHT in human tissues was brought about by the long and unsuccessful search for a reliable biochemical test of androgen excess. The concentration of DHT in plasma does not correlate with the clinical symptoms of hyperandrogenism (6). Horton *et al.* (7) proposed that plasma levels of 3αDIOL glucuronide reflected the formation and metabolism of DHT in extrahepatic, androgen target tissues, such as skin and prostate (7). However, only limited studies of the enzymes involved in DHT metabolism in human tissues other than skin and prostate have been performed, and the relative contributions of these tissues to plasma levels of androgen conjugates were not clearly established. In a recent study of human subjects, Duffy *et al.* (8) demonstrated that percutaneously applied DHT is more readily converted to 3αDIOL than DHT infused iv, suggesting significant activity of 3αHSD in skin. On the other hand, conversion of 3αDIOL to 3αDIOL glucuronide was more efficient after iv infusion of DHT than with percutaneous application. Giagulli *et al.* (9) reported that the plasma level of 3αDIOL glucuronide was an order of magnitude higher after oral administration of testosterone compared to transdermal application. These observations suggested the importance of the splanchnic organs for androgen glucuronidation and support *in vitro* data obtained from several studies. Recent studies of *in vitro* androgen UDPGT activity revealed that androgen glucuronidation occurs almost exclusively in the liver (10–13). These results from several different laboratories raise serious concerns about the hypothesis that plasma levels of 3αDIOL glucuronide reflect exclusively androgen action and metabolism in extrahepatic tissues (8–13).

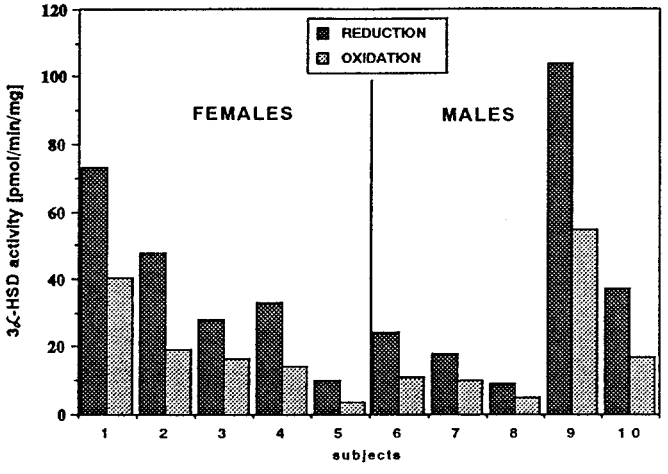

FIG. 2. 3αHSD activity (picomoles per min/mg protein) in liver homogenates from five male and five female subjects. DHT (50 nmol/L) was the substrate for the reductive reaction (DHT→3αDIOL). 3αDIOL (50 nmol/L) was the substrate for the oxidative reaction (3αDIOL→DHT).

TABLE 2. Mean activity (picomoles per min/mg ± SE) of 3αHSD and 3βHSD in liver homogenates from five male and five female subjects

Enzyme	Activity	Substrate	Mean ± SE	
			Males	Females
3αHSD	Reduction	DHT	38.5 ± 16.9	38.5 ± 10.6
3αHSD	Oxidation	3αDIOL	19.5 ± 9.0	18.9 ± 6.0
3βHSD	Reduction	DHT	8.7 ± 2.3	6.7 ± 1.7

DHT (50 nmol/L) or 50 nM 3αDIOL was used as the substrate. Values are expressed as the mean ± SE for three determinations performed in duplicate.

The level of DHT in androgen target tissues is regulated mainly by 5α-reductase and 3αHSD. 3αHSD was initially purified from rat liver cytosol, and subsequently, the gene was cloned from human liver and prostate (14–16). Although 3αHSD activity from rat liver and rat and human prostate has been extensively characterized (17–20), no comparative studies of the enzyme oxidative and reductive activities in human liver have been conducted.

We present here a comparative study of the kinetic properties of 3αHSD and 3βHSD involved in DHT metabolism in the human liver. 3αHSD, the most active DHT-metabolizing enzyme, was present in both the microsomal and cytosol fractions. The V_{max} of microsomal 3αHSD was 12-fold higher than that of cytosolic 3αHSD. This is in contrast to rat liver, where cytosolic 3αHSD was 10 times more active than microsomal 3αHSD (18). Another distinctive feature was that 3αHSD in the human was not sensitive to indomethacin, a strong inhibitor of 3αHSD in the rat (18, 21). Our laboratory and others (18, 21) have shown that rat 3αHSD possesses high affinity binding and sensitivity to inhibition by nonsteroidal antiinflammatory drugs. If human 3αHSD was inhibited by nonsteroidal antiinflammatory drugs, significant clinical consequences would be expected. However, we

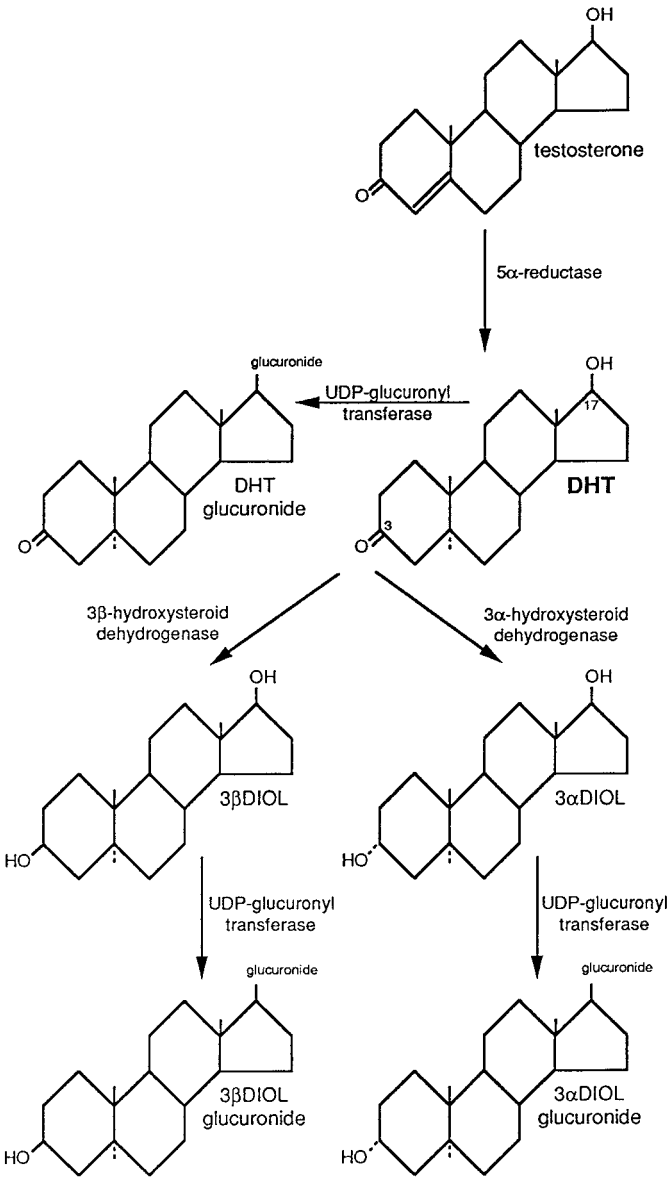

FIG. 3. Proposed scheme for DHT metabolism in human liver. The values are the enzyme velocities at a physiological concentration ($V_{max}/K_m \times$ [S] in femtomoles per min/mg protein).

found no inhibition of this enzyme activity even with a high concentration of indomethacin (10 μmol/L).

The reductive activity of 3αHSD (DHT→3αDIOL) was 2 times higher than the oxidative activity (3αDIOL→DHT). This indicates that 3αHSD in the human liver favors reduction of DHT to 3αDIOL. Liver samples from both male and female subjects showed a wide range of values in 3αHSD activity. However, the mean activity in men and women was not statistically different. This suggests that human 3αHSD is not regulated by sex steroids. In contrast, rat liver 3αHSD exhibits sexual dimorphic expression. The enzyme activity is elevated in female rat liver and appears to be under the control of estrogens (22). In addition, no change in 3αHSD activity was found to be related to age in our samples.

3βHSD activity was found primarily in the human liver microsomes and was 3-fold lower than the activity of microsomal 3αHSD. DHT reduction to 3βDIOL was 3 times higher than the oxidation of 3βDIOL to DHT. This indicates that 3βHSD in liver contributes to the clearance of DHT, but to a lesser extent than 3αHSD. These *in vitro* observations support *in vivo* studies of the metabolic clearance and origin of DHT in humans (23). Mahoudeau *et al.* (23) also found that the ratio for *in vivo* conversion of DHT to 3αDIOL was greater than that to 3βDIOL, as we report here in the liver *in vitro*. In contrast, Dijkstra *et al.* (24) found that 3βDIOL was the major product of DHT metabolism in the sebaceous glands in skin. This result is particularly interesting because plasma levels of 3βDIOL have been reported to be 3-fold higher than plasma levels of 3αDIOL in both men and women (5). There are at least two potential explanations for this observation. First, a tissue other than the liver may be the primary source for plasma 3βDIOL (*e.g.* skin). Second, the clearance of 3βDIOL may be much slower than the clearance of 3αDIOL, perhaps due to the low affinity of UDP-glucuronyl transferase for 3βDIOL. This is supported by the observation that plasma levels of 3βDIOL glucuronide are lower than those of 3αDIOL glucuronide (5). No sex or age differences in 3βHSD activity were found in the human liver.

The scheme for DHT metabolism leading to formation of glucuronated metabolites in the human liver is shown in Fig. 3. The numbers represent calculated enzyme velocities (velocity = $V_{max}/K_m \times$ [S] with a physiological substrate concentration for males). The values for androgen UDPGT velocity were calculated from previous studies from our laboratory (13). The scheme was created using results from *in vitro* kinetic studies of enzyme activity in subcellular fractions. The values represent relative enzyme velocities *in vitro* and may not represent the *in vivo* state. A well controlled perfusion study would be required to determine *in vivo* dynamics.

In summary, the results of our *in vitro* kinetic studies suggest that the main pathway of DHT metabolism leading to glucuronated metabolites in human liver is 3α-reduction, followed by subsequent conjugation to glucuronic acid. Furthermore, no sex or age differences were observed in either 3αHSD or 3βHSD activity in human liver.

References

1. **Isaacs JT, Brendler CB, Walsh PC.** 1983 Changes in the metabolism of dihydrotestosterone in the hyperplastic human prostate. J Clin Endocrinol Metab. 56:139–146.
2. **Morimoto I, Eto S, Inoue S, Izumi M, Nagataki S, Saito Y, Hara T.** 1991 DHT formation and degradation in cultured human skin fibroblasts: DHT accumulation in genital skin. J Steroid Biochem Mol Biol. 38:227–232.
3. **Aronson NN, Touster O.** 1974 Isolation of rat plasma membrane fragments in isotonic sucrose. In: Fleischer S, ed. Methods in enzymology. New York: Academic Press; vol 31:90–102.
4. **Bradford M.** 1976 A rapid sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem. 72:248–254.
5. **Belanger A, Couture J, Caron S, Roy R.** 1990 Determination of nonconjugated and conjugated steroid levels in plasma and prostate after separation on C-18 columns. Ann NY Acad Sci. 595:251–259.
6. **Giagulli VA, Giorgino R, Vermeulen A.** 1991 Is plasma 5α-androsterone-3α,17β-diol glucuronide a biochemical marker of hirsutism in women? J Steroid Biochem. 39:55–61.
7. **Horton R.** 1992 Dihydrotestosterone is a peripheral paracrine hormone. J Androl. 13:23–27.
8. **Duffy DM, Legro RS, Chang L, Stanczyk FZ, Lobo RA.** 1995 Metabolism of DHT to 5α-androstane-3α,17β-diol glucuronide is greater in the peripheral compartment than in the splanchnic compartment. Fertil Steril. 64:736–739.

9. **Gaigulli VA, Verdonck L, Deslypere JP, Giorgino R, Vermeulen A.** 1993 Comparison of androgen glucuronide levels after percutaneous or perioral androgen treatment in men: evidence for important splanchnic contribution to plasma 17β-hydroxyandrogen glucuronides. J Clin Endocrinol Metab. 76:429–431.
10. **Rittmaster RS, Zwicker HJ, Thompson DL, Konok G, Norman RW.** 1993 Androgen glucuronide production in human liver, prostate and skin. Evidence for the importance of the liver in 5α-reduced androgen conjugation. J Clin Endocrinol Metab. 76:977–982.
11. **Pirog EC, Clark RV, Collins DC.** 1993 Androgen UDP-glucuonryl transferase is found primarily in liver in the rat. J Androl. 14:2–8.
12. **Rittmaster RS, Leopold CA, Thompson DL.** 1989 Androgen glucuronyl transferase activity in rat liver, evidence for the importance of hepatic tissue in 5α-reduced androgen metabolism. J Steroid Biochem. 33:1207–1212.
13. **Pirog EC, Collins DC.** 1997 Androgen UDP-glucuronyl transferase activity is found primarily in the liver in the human. J Androl. 18:116–122.
14. **Penning TM, Mukharji I, Barrows S, Talalay P.** 1984 Purification and properties of a 3α-hydroxysteroid dehydrogenase of rat liver cytosol and its inhibition by anti-inflammatory drugs. Biochem J. 222:601–611.
15. **Khanna M, Qin KN, Wang RW, Cheng KC.** 1995 Substrate specificity, gene structure and tissue specific distribution of multiple human 3α-hydroxysteroid dehydrogenases. J Biol Chem. 270:20162–20168.
16. **Lin HK, Jez JM, Schlegel BP, Peehl DM, Pachter JA, Penning TM.** 1997 Expression and characterization of recombinant type 2 3α-hydroxysteroid dehydrogenase (HSD) from human prostate: demonstration of bifunctional 3α/17β-HSD activity and cellular distribution. Mol Endocrinol. 11:1971–1984.
17. **Penning TM, Bennett MJ, Smith-Hoog S, Schlegel BP, Jez JM, Lewis M.** 1997 Structure and function of 3α-hydroxysteroid dehydrogenase. Steroids. 62:101–111.
18. **Pirog EC, Collins DC.** 1994 3α-Hydroxysteroid dehydrogenase activity in rat liver and skin. Steroids. 59:259–264.
19. **Biswas MG, Russell DW.** 1997 Expression cloning and characterization of oxidative 17β- and 3α-hydroxysteroid dehydrogenases from rat and human prostate. J Biol Chem. 272:15959–15966.
20. **Span PN, Sweep CGJ, Bernraad TJ, Smals AGH.** 1996 3α-Hydroxysteroid oxidoreductase activities in dihydrotestosterone degradation and back-formation in rat prostate and epididymis. J Steroid Biochem Mol Biol. 3:319–324.
21. **Smithgall TE, Penning TM.** 1985 Indomethacin-sensitive 3α-hydroxysteroid dehydrogenase in rat tissues. Biochem Pharmacol. 34:831–835.
22. **Penning TM, Isaacson K, Lyttle CR.** 1992 Hormonal regulation of 3α-hydroxysteroid dehydrogenase in rat liver cytosol. Biochem Pharmacol. 43:1148–1151.
23. **Mahoudeau JA, Bardin CW, Lipsett MB.** 1971 The metabolic clearance rate and origin of plasma DHT in men and its conversion to 5α-androstanediol. J Clin Invest. 50:1338–1344.
24. **Dijkstra AC, Goss CMAA, Cunliffe WJ, Sultan C, Vermorken AJM.** 1987 Is increased 5α-reductase activity a primary phenomenon in androgen-dependent skin disorders? J Invest Dermatol. 89:87–92.