

The Source of Plasma Dihydrotestosterone in Man

T. Ito, R. Horton

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Research Article

The source of plasma dihydrotestosterone (DHT) (17β -hydroxy- 5α -androstan-3-one) in humans has been investigated by infusing two potential peripheral precursors, testosterone (T) and androstenedione (A). Metabolic clearance rates (MCR), conversion ratios (CR), transfer constants (ρ), and blood production rates (P_B) were calculated. Plasma testosterone and dihydrotestosterone were measured by competitive binding techniques. The MCR^{DHT} was 652 ± 35 (SD) liters/day in five males and 314 ± 63 (SD) liters/day in four adult females. In each individual, the MCR^{DHT} was significantly lower than MCR^T as predicted by testosterone-binding protein affinity studies. The P_B^{DHT} was 302 ± 65 (SD) $\mu\text{g/day}$ in males and 56 ± 26 $\mu\text{g/day}$ in females. Testosterone and androstenedione are precursors (prehormones) for plasma dihydrotestosterone. The conversion ratio CR_{BB}^{T-DHT} , calculated as the ratio of counts per minute per liter of plasma of product to precursor after infusion of labeled precursor, was 5.6 ± 0.6 (SD)% (six subjects) in the male and 3.5 ± 0.4 (SD)% (four subjects) in the female. CR_{BB}^{A-DHT} after androstenedione infusion to three female subjects averaged 9.2%. No dihydrotestosterone back conversion was detected ($< 0.2\%$). The transfer constants were $[\rho]BB^{T-DHT}$, $3.9 \pm 1.0\%$ (male) and $1.7 \pm 0.6\%$ (female), and $[\rho]BB^{A-DHT}$ average was 13.3% in three female subjects. Using either plasma testosterone and dihydrotestosterone values from our subjects and mean androstenedione values as reported in the literature, approximate contributions can be calculated. Testosterone conversion accounts for [...]

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T. ITO and R. HORTON

From the Department of Medicine, University of Southern California, School of Medicine, Los Angeles, California 90033

ABSTRACT The source of plasma dihydrotestosterone (DHT) (17β -hydroxy- 5α -androstan-3-one) in humans has been investigated by infusing two potential peripheral precursors, testosterone (T) and androstenedione (A). Metabolic clearance rates (MCR), conversion ratios (CR), transfer constants (ρ), and blood production rates (P_B) were calculated. Plasma testosterone and dihydrotestosterone were measured by competitive binding techniques. The MCR^{DHT} was 652 ± 35 (SD) liters/day in five males and 314 ± 63 (SD) liters/day in four adult females. In each individual, the MCR^{DHT} was significantly lower than MCR^T as predicted by testosterone-binding protein affinity studies. The P_B^{DHT} was 302 ± 65 (SD) μ g/day in males and 56 ± 26 μ g/day in females. Testosterone and androstenedione are precursors (prehormones) for plasma dihydrotestosterone. The conversion ratio CR_{BB}^{T-DHT} , calculated as the ratio of counts per minute per liter of plasma of product to precursor after infusion of labeled precursor, was 5.6 ± 0.6 (SD) % (six subjects) in the male and 3.5 ± 0.4 (SD) % (four subjects) in the female. CR_{BB}^{A-DHT} after androstenedione infusion to three female subjects averaged 9.2%. No dihydrotestosterone back conversion was detected ($< 0.2\%$). The transfer constants were $[\rho]_{BB}^{T-DHT}$, $3.9 \pm 1.0\%$ (male) and $1.7 \pm 0.6\%$ (female), and $[\rho]_{BB}^{A-DHT}$ average was 13.3% in three female subjects. Using either plasma testosterone and dihydrotestosterone values from our subjects and mean androstenedione values as reported in the literature, approximate contributions can be calculated. Testosterone conversion accounts for at least 70% of plasma DHT in the male, but less than 20% in the normal female. Androstenedione appears to be a major prehormone of plasma dihydrotestosterone accounting for at least two-thirds plasma dihydrotestosterone by peripheral conversion in adult females. In three normal women undergoing tubal ligation, there was an unimpressive gradient

between ovarian vein and peripheral plasma dihydrotestosterone. It is suggested that dihydrotestosterone in the blood does not arise from direct secretion but may reflect events occurring in peripheral androgen target tissues.

INTRODUCTION

It is now clearly established that certain sex steroids are interconvertible in the body. The rates of interconversion vary from tissue to tissue. Of great importance is the observation that conversion rates as measured in blood differ from those calculated by the analysis of urinary metabolites (1). The study of interconverting steroid pairs has indicated that a biologically secreted active steroid hormone in blood may be derived from an inactive secreted precursor (prehormone). Examples are the interconverting pairs of androstenedione-testosterone (2),¹ estrone-estradiol (3), and androgen-estrogen (4).

In a series of in vitro studies, Wilson, Bruchovsky, and Chatfield found that testosterone was efficiently converted by androgen target tissues to the reduction product, dihydrotestosterone.¹ This steroid is bound with much higher affinity to nuclear chromatin than is testosterone and may be the effective tissue androgen (5).

We have recently published an accurate competitive binding method for the measurement of dihydrotestosterone in peripheral plasma (6). This biologically active androgen may be a secreted hormone or may reflect peripheral conversion of other secreted androgens. Our study concludes that blood dihydrotestosterone arises from the peripheral conversion of two different secretory products.

¹Systematic nomenclature for trivial names used in this paper: dihydrotestosterone, 17β -hydroxy- 5α -androstan-3-one; 5β -DHT, 17β -hydroxy- 5β -androstan-3-one; testosterone, 17β -hydroxy-4-androsten-3-one; androstenedione, 4-androstene-3,17-dione; estradiol, 1,3,5(10)-estratriene-3,17 β -diol; estrone, 1,3,5(10)-estratriene-3-ol-17 β -one.

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METHODS

Infusion procedure. Normal adult men and women were infused with testosterone-1,2-³H (T),² androstenedione (A), or dihydrotestosterone (DHT) by the constant infusion technique as previously described (2). 2-hr infusions were given after the patients were kept supine for a 1 hr control period. All experiments were started between 8 and 9 a.m. A priming dose of 1–3 μ Ci was given intravenously, and a constant infusion of 10–30 μ Ci of the ³H-labeled steroid in 50 ml of a 6% ethanol solution in normal saline was administered through Teflon tubing as previously described (2). Control blood samples were drawn for measurement of plasma testosterone and dihydrotestosterone, and infusion samples were obtained at 105 and 120 min. The plasma was then frozen immediately at -20° C.

Analysis of precursor and product radioactivity. 20–30 ml of plasma was added to a flask containing testosterone-4-¹⁴C (300 cpm) and dihydrotestosterone (100 cpm) for the T-1, 2-³H studies and androstenedione-4-¹⁴C for the androstenedione-1,2-³H infusions. The plasma was then alkalinized with 2 ml of 4 N NaOH, and the steroids were extracted two times, each time with three volumes of methylene chloride. The extract was then washed twice with 10 ml of 1 N NaOH and twice again with 10 ml water, transferred to a round bottom flask, and evaporated in a Rinco type vacuum setup. 6 ml of 70% methanol and 12 ml of petroleum ether (bp 75.8–96.4 $^{\circ}$ C) were added, shaken, and the petroleum ether was removed and discarded. The remaining methanol was evaporated to dryness.

Paper chromatographic purification was carried out using the Bush B-3 and the Bush A paper systems as previously described (6). In a typical 2 hr run in the B-3 system, testosterone moved 15 cm and DHT ran 23 cm. The samples corresponding to ± 1.5 cm from the center of the parallel standards (which were located by scanning) were then cut out and eluted with 5 ml methanol using the syringe technique.

After the first chromatogram, the separated steroid samples were acetylated overnight by adding 0.2 ml pyridine and 0.2 ml acetic anhydride. After drying the acetylated samples, they were separately spotted on Whatman paper No. 2 and run in the A system for 2 hr. The acetylated steroids were located against parallel ³H-acetylated standards passed through a strip scanner. Although androstenedione moves the same as dihydrotestosterone in the Bush 3 system, these two testosterone conversion products are completely separated by the acetylation procedure (androstenedione runs 10 cm, dihydrotestosterone acetate runs 20 cm).

The following infusion studies were performed: (a) testosterone-³H, six males and four females; (b) dihydrotestosterone-³H, five males and four females; and (c) androstenedione-³H, three females.

Radioactive counting. All counting was performed with a Nuclear-Chicago scintillation spectrometer. The characteristics for counting tritium and carbon-14 are as described previously (4). An analysis of counting errors for this type of study has been extensively described elsewhere (7).

Radioactive steroids. Testosterone-1,2-³H, androstenedione (SA 44 Ci/mmoles per liter), and steroids-¹⁴C were obtained from New England Nuclear Corp., Boston, Mass.

² Abbreviations used in this paper: A, androstenedione; BB, blood to blood; CR, conversion ratio; DHT, dihydrotestosterone; MCR, metabolic clearance rate; P_b, blood production rate; PR, production rate; ρ , transfer constant; T, testosterone.

All steroids used were purified by paper chromatography in the Bush B-3 system. Evidence for radiochemical purity has been previously presented (4, 6).

Dihydrotestosterone-4-¹⁴C was made from previously purified testosterone-4-¹⁴C. Approximately 100 μ Ci testosterone-¹⁴C was taken to dryness under vacuum, and then 0.1 ml ethanol and 1 ml NH₄OH were added to the flask. Potassium metal (J. T. Baker Chemical Co., Phillipsburg, N. J.) previously cut into small strips (1–2 mm) under petroleum ether, was slowly added to the flask. Addition was continued for several hours to generate H₂. 1 ml water was then added to stop this reaction, and the ammonium hydroxide was evaporated under vacuum. The solution was neutralized with dropwise acetic acid and steroids extracted with two volumes of methylenedichloride, and the dihydrotestosterone-¹⁴C was purified by repeated paper chromatography. The other possible reduction product, 5 β -DHT, was separated from DHT in both the B-3 and Bush A paper systems because of its greater polarity.

Proof of radiochemical purity of the dihydrotestosterone-³H and dihydrotestosterone-¹⁴C was obtained by mixing the purified DHT-1,2-³H with the prepared and purified DHT-4-¹⁴C (³H/¹⁴C ratio 3.18). The remaining mixture was chromatographed in the B-3 system (³H/¹⁴C ratio 3.16). Evidence for attainment of equilibrium after infusion for 100 min for both testosterone and androstenedione has been previously reported (2).

Estimation of plasma testosterone and dihydrotestosterone. Plasma testosterone and dihydrotestosterone were measured in samples obtained at the beginning of the infusion by competitive binding methods developed in our laboratory (6, 8). Peripheral and ovarian vein plasma from three women undergoing tubal ligation were assayed for dihydrotestosterone.

Evidence for radiochemical purity of dihydrotestosterone-³H isolated from plasma during infusion. After a constant infusion of testosterone-³H, a pool of plasma was made, and DHT-4-¹⁴C and T were added. The sample was then extracted, chromatographed on the Bush B-3 system, acetylated, and run as in the standard method in the Bush A system. The remaining acetylated sample was then chromatographed in the B-3 system. The calculated isotope ratios were 4.0 and 3.7, respectively, which could be accounted for by the error of counting at that level.

Proof of the radiochemical purity of DHT-³H after infusion of androstenedione-³H was obtained by the same technique. A pool of plasma after an infusion of androstenedione-³H was prepared. The plasma with dihydrotestosterone-¹⁴C was purified as in the regular procedure which included Bush B-3 paper chromatography, acetylation of the sample running parallel to standard dihydrotestosterone, and rechromatography in the Bush A system. One-third was taken for counting (³H/¹⁴C = 3.05). The remaining sample was then further purified in the Bush A system. No significant change was observed in the isotope ratio (³H/¹⁴C = 3.02).

Evidence that equilibrium in plasma was attained during the constant infusion. Analysis of radioactivity as the purified steroid-³H corrected for recovery after the priming dose was determined as previously described for testosterone and androstenedione (2, 4). In the six infusions where the value at 105 min is compared with the respective 120-min samples, the mean value at 120 min was 96.6 ± 2.7 (SE) student's test ($t = 1.16$). Also, if an individual ratio (120/105 min value) for each infusion is calculated and if the mean ratio is 0.96, the 95% confidence interval is equal to 0.83–1.09. This indicates that this ratio is not significantly different from 1.0.

Calculation of metabolic clearance, production rates, and interconversion rates. Metabolic clearance rates (MCR), conversion ratios (CR), and transfer constants, were calculated as previously described (2). The blood production rate (PR) is the product of the mean metabolic clearance rate and plasma concentration. In this study as in previous investigations, the production rate (PR) was calculated using the steroid concentration in plasma and metabolic clearance obtained during the 2 hr morning infusion.

In previous studies only the labeled precursor was infused, and calculations of the transfer constants ($[\rho]^{\text{Pre-Prod}}$) used the mean metabolic clearance value of the product from similar individuals (2-4) since:

$$[\rho]^{\text{Pre-Prod}} = \frac{\text{MCR}^{\text{Prod}} \cdot \bar{x}^{\text{Prod}}}{R^{\text{Pre}}} = \frac{\text{MCR}^{\text{Prod}} \cdot \bar{x}^{\text{Prod}}}{\text{MCR}^{\text{Pre}} \cdot \bar{x}^{\text{Pre}}}$$

where \bar{x} is the mean level of radioactivity as precursor (Pre) or product (Prod) and R is the rate of infusion of precursor (2). In the present investigation, both labeled precursor and product were separately infused into the same patient under this same condition which should yield more accurate data for the group studied.

The fraction of the blood production rate of dihydrotestosterone ($P_{\text{B}}^{\text{DHT}}$) coming from the blood production rate of testosterone (P_{B}^{T}) is given by the expression:

$$\frac{P_{\text{B}}^{\text{T}} \cdot [\rho]_{\text{BB}}^{\text{T-DHT}}}{P_{\text{B}}^{\text{DHT}}} \div 1 - [\rho]_{\text{BB}}^{\text{T-DHT}} [\rho]_{\text{BB}}^{\text{DHT-T}} \quad (2)$$

where $P_{\text{B}}^{\text{T}} \cdot [\rho]_{\text{BB}}^{\text{T-DHT}}$ is the production of dihydrotestosterone in the blood arising from the production of testosterone (9). Usually in this type of calculation, a correction ($1/1 - [\rho]_{\text{BB}}^{\text{T-DHT}} [\rho]_{\text{BB}}^{\text{DHT-T}}$) has to be made because some of the precursor has arisen initially from the product. However, with the result presented here, $[\rho]_{\text{BB}}^{\text{DHT-T}}$ is undetectable and the correction is unnecessary, and so the fraction of total $P_{\text{B}}^{\text{DHT}}$ made in this manner is expressed by the formula

$$\frac{P_{\text{B}}^{\text{T}} \cdot [\rho]_{\text{BB}}^{\text{T-DHT}}}{P_{\text{B}}^{\text{DHT}}}$$

RESULTS

Metabolic clearance rates. Metabolic clearance rates of dihydrotestosterone (MCR^{DHT}) by continuous infusion were 652 ± 35 (SD) liters/day in five male subjects and 314 ± 63 (SD) liters/day in four female adults (Table I). The different values in males and females are significantly different ($P < 0.02$) and remain so even if corrected for sex differences in surface area.

Metabolic clearance of testosterone (MCR^{T}) was 976 ± 57 (SD) liters/day in six males and 667 ± 70 (SD) liters/day in four females. These MCR^{T} values were similar to those previously reported by our group (2, 4). There is a significant difference between MCR^{DHT} and MCR^{T} values in males and females ($P < 0.05$).

Plasma concentration of dihydrotestosterone and testosterone. Plasma dihydrotestosterone levels were 46 ± 19 (SD) $\mu\text{g}/100$ ml in five males and 17 ± 7 (SD) $\mu\text{g}/100$ ml in four females. Plasma testosterone was 544 ± 208 (SD) $\mu\text{g}/100$ ml in six male and 40 ± 9 (SD) $\mu\text{g}/100$ ml in the four female subjects. These plasma values for both androgens are indistinguishable from normal values reported by us from a much larger group of normal individuals (6).

Blood production rates. Production rates were calculated individually as the product of MCR and plasma concentration. The blood production rate of dihydrotestosterone was 302 ± 65 $\mu\text{g}/\text{day}$ in adult males and 56 ± 26 $\mu\text{g}/\text{day}$ in females. Testosterone production rates were 5130 ± 740 $\mu\text{g}/\text{day}$ in males and 267 ± 37 $\mu\text{g}/\text{day}$ in females.

Conversion ratios and transfer constants in blood. The conversion ratio, $\text{CR}_{\text{BB}}^{\text{T-DHT}}$, was calculated as the ratio of counts per minute per liter of plasma of product

TABLE I
Data for the Determination of MCR^{DHT} and PR^{DHT} in Men and Women

Subject	Sex	Infused DHT-3H	DHT-3H			MCR^{DHT}	Plasma DHT	PR^{DHT}
			105 min	120 min	Mean			
			<i>cpm/day</i>	<i>cpm/liter</i>		<i>liters/day</i>	<i>μg/liter</i>	<i>μg blood/day</i>
S.	M	19×10^6	27,000	29,000	28,000	691	556	384
R.	M	24×10^6	34,000	37,000	35,900	689	409	282
Sc.	M	23×10^6	35,000	38,000	36,500	629	756	475
A.	M	19×10^6	25,000	27,000	26,000	728	389	283
I.	M	40×10^6	77,000	75,000	76,000	526	163	86
Mean \pm SD						652 ± 35	455 ± 196	302 ± 65
L.	F	43×10^6	188,000	173,000	80,000	243	188	46
V.	F	80×10^6		Pool	219,000	372	89	33
W.	F	57×10^6		Pool	120,000	437	277	121
Sp.	F	18×10^6		Pool	90,000	204	126	24
Mean \pm SD						314 ± 63	170 ± 71	56 ± 26

TABLE II
*Metabolic Clearance, Conversion Ratio, Transfer Constant, and Production Rate of Testosterone,
 Dihydrotestosterone, and Androstenedione, Measured in Human Plasma*

Subject	Sex	Infused precursor, testosterone	Precursor after infusion			MCRT	Mean DHT after infusion			CR _{BBT-DHT}	[ρ] _{BBT-DHT}	Plasma T <i>m</i> μ g/liter	PR _{BT} <i>mg/day</i>
			90 min	120 min	Mean		105 min	120 min	Mean				
I.	M	14×10^7	134,900	150,000	142,700	993	7,200	7,000	7,100	5.0%	2.7	3515	3.49
S.	M	85×10^6	Pool			818	Pool			5.7	4.4	6524	5.34
A.	M	50×10^6	36,000	41,000	38,500	1300	7,900	7,500	7,700	6.4	5.2	4572	5.95
C.	M	97.6×10^6	118,000	121,000	119,500	817	8,400	7,600	8,000	4.6	3.0	9614	7.85
Sp.	M	110×10^6	127,300	127,300	127,300	867	Pool			5.6 \pm 0.6	3.9 \pm 1.0	4085	3.54
R.	M	87×10^6	Pool			1062	Pool			3.5	1.4	4347	4.61
						976 \pm 57						5443 \pm 2085	5.13 \pm 0.74
L.	F	170×10^6	295,000	282,000	288,500	598	10,500	9,500	10,000	3.5	1.8	509	0.30
V.	F	141×10^6	153,000	169,000	161,000	835	6,200	7,200	6,700	4.2	2.6	303	0.25
W.	F	150×10^6	291,000	247,000	269,000	559	8,500	9,700	91,000	3.4	0.9	326	0.18
Sp.	F	55×10^6	91,000	72,400	82,100	678	3,200	2,000	2,600	3.2	1.7 \pm 0.6	484	0.33
						667 \pm 71				3.5 \pm 0.4		405 \pm 92	0.26 \pm 0.40
Androstenedione													
			MCR _A				[ρ] _{BBT-DHT} *						
V.	F	23×10^7	99,670	109,190	104,430	2228	5,860	5,980	5,920	5.7	8.0		
L.	F	22×10^7	94,970	117,950	106,460	2152	11,520	11,530	11,525	10.8	15.7		
Sp.	F	32×10^7	Pool			1723	Pool			11.3	16.3		

* Calculated from $\text{Ln} \frac{\text{CR}^{\text{Pre-Prod}}}{\text{MCR}^{\text{Pre}}} = \text{MCR}^{\text{Pro}} (4)$.

to precursor both corrected for recovery after infusion of the labeled precursor. The conversion ratio of dihydrotestosterone/testosterone (CR_{BB}^{T-DHT}) after infusion of labeled testosterone was 5.6 ± 0.6 (SD) % (six subjects) in males and 3.5 ± 0.4 (SD) % (four subjects) in females. CR_{BB}^{A-DHT} after infusion of androstenedione to three females averaged 9.2%. After infusion of labeled dihydrotestosterone, no tritium was detectable as testosterone indicating that the conversion CR_{BB}^{DHT-T} is negligible ($< 0.2\%$). The transfer constants for testosterone $[p]_{BB}^{T-DHT}$ and androstenedione $[p]_{BB}^{A-DHT}$ are illustrated in Table II. $[p]_{BB}^{T-DHT}$ is $3.9 \pm 1.0\%$ in the male and $1.7 \pm 0.6\%$ in the female. $[p]_{BB}^{A-DHT}$ averages 13.3 in the female.

The contribution of testosterone and androstenedione to blood dihydrotestosterone. Taken together the data allow calculation of the contribution of each blood precursor to product since:

$$\%DHT \text{ from T} = \frac{\text{Plasma T} \cdot CR_{BB}^{T-DHT}}{\text{Plasma DHT}}.$$

The contribution of testosterone to plasma concentration would then be 31 $\mu\text{g}/100 \text{ ml}$ in the male but only 1.4 $\mu\text{g}/100$ in the female. No detectable ($< 0.2\%$) back conversion was found. These data suggest that approximately 70% of blood DHT in the male but only 8% in the female is derived from the peripheral conversion of testosterone.

If the conversion ratio for androstenedione (CR_{BB}^{A-DHT}) is considered with normal plasma androstenedione values in the female of 159 $\mu\text{g}/100 \text{ ml}$ previously reported (10), then the data suggest that approximately two-thirds of blood dihydrotestosterone in the female is derived from androstenedione. The direct measurement of DHT in peripheral and ovarian vein plasma indicates minimal ovarian secretion (Table III).

DISCUSSION

Steroids, when compared with proteins, are relatively simple planar molecules. Yet studies with androgens, estrogens, or mineralocorticoids have demonstrated a high degree of stereospecificity with the tissue-binding (receptor) proteins (11-14). The sex steroids are also bound to plasma proteins. In general, there has been excellent correlation between binding affinity and biological activity. It was surprising when we and others (11, 12) observed that dihydrotestosterone had approximately twice the binding affinity to the sex hormone-binding plasma protein as did testosterone, although testosterone was thought to be the circulating androgen in blood.

TABLE III
*Dihydrotestosterone in Ovarian Vein and Peripheral Plasma of Women during Laparotomy**

Subject	Peripheral	Ovarian vein
	<i>m$\mu\text{g}/100 \text{ ml}$</i>	
1	8.2	7.8
2	6.0	12.9
3	16.0	23.0

* Plasma samples obtained from Doctors J. Lobotsky and C. W. Lloyd.

Hormone binding at the site of action is considered an essential event in hormone action. Wilson and co-workers observed that dihydrotestosterone, a potent androgen, binds with higher affinity to nuclear chromatin of prostate and seminal vesicle than does testosterone (5, 15). Their in vitro work suggests that testosterone is rapidly and efficiently converted to dihydrotestosterone by a 5α -reductase located in the nucleus. In vitro dihydrotestosterone is formed in large amounts only in accessory sex tissues. In one tissue, skin, this group reported a much higher conversion by perineal sexual skin than skin from other sites (16). It should be noted that lack of conversion by muscle from 11 different species indicates that formation is not required to explain all known androgen effects (17). After an injection of labeled testosterone into the intact animal, small amounts of labeled dihydrotestosterone appeared in blood.

Using techniques developed to quantitate steroid conversion rates in the circulation and a sensitive plasma dihydrotestosterone assay, we have attempted to measure blood production or secretion rates and to determine whether plasma dihydrotestosterone is the result of direct secretion or reflects peripheral events.

As suggested by the affinity studies with the plasma-binding protein, the metabolic clearance rates in both males and females are significantly lower in each respective sex than the testosterone clearance rates. Dihydrotestosterone blood production rates were $302 \pm 65 \mu\text{g}/\text{day}$ in six males and only $56 \pm 26 \mu\text{g}/\text{day}$ in four females. Blood production rates of dihydrotestosterone were also much lower than testosterone blood production in the respective sex. The testosterone/dihydrotestosterone production rate ratio in the female was approximately 5:1. In both males and females therefore, the plasma concentration and production rate of DHT are only fractions of testosterone production.

A significant amount of testosterone infused into the general circulation is converted to dihydrotestosterone. The conversion ratio, C_{BB}^{T-DHT} , is $5.6 \pm 0.6\%$ in the male and 3.5 ± 0.4 in the female. The transfer factor $[\rho]_{BB}^{T-DHT}$ is $3.9 \pm 1.0\%$ in the male and $1.7 \pm 0.6\%$ in the female. As has been observed in previous studies of androstenedione-testosterone (2) and testosterone-estradiol and androstenedione-estrone conversions (4), there is a significantly higher conversion ratio or transfer constant in the male. In three females, the conversion ratio of androstenedione to dihydrotestosterone (C_{BB}^{A-DHT}) was 9.2%. Since plasma testosterone and dihydrotestosterone were measured simultaneously with the conversion data in these patients, a direct calculation indicating the contribution of the plasma precursor to dihydrotestosterone in plasma can be obtained. Despite the difference in conversion ratios seen in the male and female, the major factor appears to be the excess of precursor testosterone present in male plasma. As a result of this, approximately three-fourths of the plasma dihydrotestosterone is derived from the peripheral conversion of testosterone in the male, but less than 20% in the female. The studies of androstenedione-dihydrotestosterone conversion indicate that androstenedione is also a precursor (prehormone) for plasma dihydrotestosterone. When the conversion ratio (CR_{BB}^{A-DHT}) is considered together with published values for plasma androstenedione (mean 116 $\mu\text{g}/100$ ml in male and 159 $\mu\text{g}/100$ ml in the female [10]) these data suggest that the remaining plasma dihydrotestosterone, not accounted for by testosterone, may be derived from peripheral conversion of androstenedione. Testosterone is the major source (prehormone) of plasma dihydrotestosterone in the male, while androstenedione appears to be the major prehormone for blood dihydrotestosterone in the adult female. This study therefore would suggest that blood dihydrotestosterone is not a secretion product in either of the sexes. Further support for this concept is provided by a direct comparison of peripheral and ovarian vein dihydrotestosterone levels where a minimal gradient was detected in three women.

As expected from consideration of steroid structure and in vitro experience (18), no back conversion of the 5α -androstane steroid to the 4-androstene type steroid was detected from these blood studies.

This study suggests that there is a different kind of hormone circulating in plasma. Previous conversion products, such as testosterone in the female and estradiol in the male, appear to be the results of both peripheral conversion and direct secretion (1). The precursor (prehormone) has been inactive or has possessed differ-

ent biological properties than the product in blood. In the present situation of testosterone-dihydrotestosterone in the male, there is conversion of an active androgen into a similarly active androgen. However, in the female the previous pattern again appears. Testosterone is a minor prehormone, but androstenedione, a very weak androgen, appears to be the major precursor for plasma dihydrotestosterone. This investigation suggests that dihydrotestosterone is not a secreted steroid in females but that blood dihydrotestosterone is derived from both androstenedione and testosterone peripheral conversion. Although this work indicates a peripheral, nonendocrine tissue source for dihydrotestosterone, it is not possible to indicate whether synthesis occurs in hepatic or androgen target tissues. This is now an important question since blood dihydrotestosterone may be an important reflection of androgen events occurring in target tissues.

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