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Novel Steroid 5α-Reductase Inhibitor FK143: Its Dual Inhibition against the Two Isozymes and Its Effect on Transcription of the Isozyme Genes

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SUMMARY

Recent cloning of the cDNAs for the two isozymes of steroid 5α-reductase (EC 1.3.99.5) allowed individual expression of the isozymes and permitted us to investigate the action of steroid 5α-reductase inhibitors against the individual isozymes without any ambiguity that may be caused by coexistence of the isozymes in tissue preparations. We examined the kinetic characteristics of FK143 (4-[3-[3-[bis(4-isobutylphenyl)methylamino]benzoyl]-1*H*-indol-1-yl]butyric acid), a novel nonsteroidal steroid 5α-reductase inhibitor against cloned human and rat steroid 5α-reductase isozymes. FK143 was shown to inhibit both isozymes equally. The mode of the inhibition of FK143 against both isozymes was noncompetitive. The inhibition constants K_{ie} and K_{ies} of FK143 for human types 1 and 2 were 27.0 and 19.6 nM and 19.9 and 14.5 nM, respectively. Species selectivity between human and rat of the inhibitory activity of FK143 against both isozymes was not found. We also examined the effect of FK143 on the *in vivo* expression of the genes encoding for the rat steroid 5α-reductase isozymes. FK143 reduced the testosterone-induced increase in the amount of the type 1 mRNA in castrated rat, whereas it did not substantially affect the amount of the type 2 mRNA.

Steroid 5α-reductase is the enzyme that converts T into its active form, DHT (1). DHT is essential for the growth and development of the prostate gland and is assumed to be a major factor in the development of BPH (2, 3). Accordingly, steroid 5α-reductase has been a promising target of therapeutic agents for BPH (4, 5).

Recently, human steroid 5α-reductase cDNAs were cloned (6), revealing unambiguously the presence of two isozymes (7). Subsequent extensive investigation into the biochemical properties and tissue distribution of both human and rat isozymes showed that the two isozymes exhibit quite distinct characteristics, leading us to assume that they have different physiological roles (6–13).

The type 2 enzyme had a 10–15-fold higher affinity than the type 1 isozyme for T and several other steroid substrates (8, 12). Northern hybridization of RNA from various rat tissues indicated that the type 1 mRNA was predominant in nonandrogen target tissues, such as liver, whereas the type 2 mRNA was predominant in male reproductive tissues, such as testis, vas deferens, and epididymis (9, 10, 12, 13). These enzymatic properties and tissue-selective distribution of the two isozymes are consistent with the hypothesis that type 1 enzyme has a catabolic role in steroid metabolism and type 2 isozyme has an anabolic role (12).

However, the role of the isozymes in prostate are complicated by their distribution in the prostate of humans and rats. Rat ventral prostate has been shown to comparatively express two isozymes (12), whereas human type 2 is expressed predominantly in the prostate (9). *In situ* hybridization of rat prostate RNA revealed the cell-specific expression of the two isozyme mRNAs (13); rat type 1 was expressed in basal epithelial cells, whereas the expression of type 2 was confined to stromal cells. This observation indicates that two rat isozymes also have distinct roles in the prostate, suggesting that type 2 participates during embryonic development and type 1 participates in glandular growth and maintenance during adulthood. Further evidence is needed to define the roles of the rat and human isozymes in prostate and the participation of human isozymes in the development of BPH.

Several steroid 5α-reductase inhibitors, including FK143, have recently been developed as therapeutic agents for BPH (14–16). These studies on steroid 5α-reductase have enabled us to examine the action of these inhibitors against the individual isozymes of the enzyme and will provide a basis

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ABBREVIATIONS: T, testosterone; DHT, dihydrotestosterone; BPH, benign prostatic hyperplasia; FK143, (4-[3-[3-[bis(4-isobutylphenyl)methylamino]benzoyl]-1*H*-indol-1-yl]butyric acid); PCR, polymerase chain reaction; TLC, thin layer chromatography.

for analysis of the efficacy of inhibitors against BPH. In the present, we investigated the kinetic characteristics of FK143 against cloned isozymes of human and rat steroid 5α-reductases and the effect of FK143 on the expression of the rat isozyme genes *in vivo*.

Experimental Procedures

Materials. FK143 and finasteride were prepared by the Exploratory Research Laboratories of Fujisawa Pharmaceutical Co., Ltd. [α-^{32}P]-dCTP was purchased from Amersham International (Bucks, UK), and 1,2,6,7-^{3}H-T was purchased from Dupont-New England Nuclear (Boston, MA). Restriction endonucleases and DNA ligase were obtained from Takara Shuzo (Kyoto, Japan). We obtained the TA cloning kit from Invitrogen (San Diego, CA), the PCR amplification kit from Takara Shuzo, and the Sequenase DNA sequencing kit from United States Biochemical Corporation (Cleveland, OH). Human and rat testis and liver cDNA were purchased from Clonetech (Palo Alto, CA). Primers for PCR amplification were synthesized with a Model 380 synthesizer (Applied Biosystems, Foster City, CA).

Cloning of cDNAs for human and rat steroid 5α-reductase isozymes. The cDNA for human steroid 5α-reductase type 1 was kindly provided by Dr. D. W. Russell of the University of Texas (11). The cDNAs for human steroid 5α-reductase type 2 and rat steroid 5α-reductase types 1 and 2 were cloned using the PCR amplification method.

Human and rat steroid 5α-reductase type 2 cDNAs were amplified individually with the use of human and rat testis cDNA, respectively. Each cDNA was amplified using two 20-mer primers homologous to the 5′ and 3′ untranslated sequence of the corresponding cDNA. The amplified products were cloned into pCRII (Invitrogen), yielding pCRhT5αR2–1 and pCRrT5αR2–1.

Rat steroid 5α-reductase type 1 cDNA was amplified with the use of rat liver cDNA as template and two 30-mer primers. The sense primer was homologous to the 5′ untranslated sequence of the type 1 cDNA and included a *Hind*III recognition site, whereas the antisense primer was homologous to the 3′ untranslated sequence and included an *Eco*RI recognition site. After digestion with *Hind*III and *Eco*RI, the amplified products were cloned into pBSIISK(+) (Stratagene, La Jolla, CA), yielding pBSrT5αR1–1. The nucleotide sequence of each cDNA cloned was determined by the M13/dideoxy chain termination method (17) and verified to be identical with the reported sequence (6, 7, 11, 12).

Construction of expression plasmids. All of the cDNAs for human and rat steroid 5α-reductase types 1 and 2 were cloned into the mammalian expression vector pCDM8 (Invitrogen).

The plasmid p45–1, which contains cDNA for human type 1, was kindly provided by Dr. Russell. It was cleaved with *Not*I, and the resulting fragment of cDNA insert was isolated and ligated with *Not*I cleaved pCDM8, yielding pCDMhT5αR1–1. The expression plasmid for human type 1, pCDMhT5αR1–101, was produced by deleting the *Xho*I fragment containing stuffer from pCDMhT5αR1–1. The plasmid pCRhT5αR2–1 containing cDNA for human type 2 was digested with *Hind*III and *Not*I, and the fragment containing the cDNA was isolated and ligated into the pCDM8 vector to give pCDMhT5αR2–1, an expression plasmid for human type 2.

The plasmid pBSrT5αR1–1 containing cDNA for rat type 1 was digested with *Hind*III and *Xho*I, and the fragment of the cDNA was ligated into pCDM8 to give pCDMrT5αR1–1, an expression plasmid for rat type 1. The plasmid pCRrT5αR2–1 containing cDNA for rat type 2 was digested with *Hind*III and *Not*I. The cDNA fragment was ligated into pCDM8 to give pCRrT5αR2–2 with the cDNA insert in the reverse orientation with respect to the promoter. This plasmid was digested with *Eco*RI, and the resulting two fragments were religated to produce the expression plasmid for rat type 2, pCRrT5αR2–1.

Transfection and preparation of enzymes. Human embryonic kidney 293 cells (American Type Culture Collection, Rockville, MD) were transfected by the calcium phosphate precipitation method (18) with 20 μg of expression plasmids pCDMhT5αR1–101, pCDMhT5αR2–1, pCDMrT5αR1–1, or pCDMrT5αR2–1. After incubation for 60 hr, the cells were washed twice with phosphate-buffered saline and collected with a rubber policeman. The cell suspension was centrifuged, and the resulting cell pellets were resuspended in homogenizing buffer consisting of 0.32 M sucrose, 0.1 M dithiothreitol, and 20 mM sodium phosphate buffer, pH 6.5, and disrupted in an ice bath by sonication at maximal power three times for 15 sec. The resulting sonicate was used as the enzyme preparation.

Enzyme assays. The activities of steroid 5α-reductase were assayed with TLC (19). The reaction mixtures contained the following components in a final volume of 200 μl: 200 nmol dithiothreitol, 8 mmol sodium phosphate buffer, pH 7.5, 400 nmol NADPH, 0.8 pmol–2 nmol T including 0.4 pmol 1,2,6,7-^{3}H-labeled T (3.44 TBq/mmol), and enzyme preparation except where otherwise specified. Reaction mixtures were incubated at 37° for 20 min, and the reaction was terminated by the addition of 300 μl ethylacetate. After subsequent extraction with ethylacetate, a portion of the extract was subjected to TLC with silica gel (5748–7, E. Merck, Darmstadt, Germany) using ethylacetate/cyclohexane (1:1, v/v) as the solvent system. After TLC, spots corresponding to T and DHT were cut out and subjected to liquid scintillation counting. Enzymatic activities were expressed as the corrected values of DHT produced (in picomoles) per minute, which were calculated from the ratio of the radioactivity of DHT to the sum of the radioactivities of T and DHT.

Protein concentrations were determined with the BioRad protein assay kit (Bio-Rad Co., Hercules, CA) with bovine serum albumin as a standard.

The inhibitory assay data were processed with Cricket Graph software for Macintosh to draw Lineweaver-Burke plots and to calculate the kinetic parameters K_{ie} and K_{ies}. K_{ie} and K_{ies} are dissociation constants for enzyme-inhibitor complex and enzyme-substrate-inhibitor complex, respectively. K_{ie} and K_{ies} were calculated as the horizontal intercepts that were obtained by replotting the slopes and vertical intercepts of Lineweaver-Burke plots versus inhibitor concentrations, respectively.

Measurement of mRNA. Northern blot hybridization and reverse-transcription PCR were used for the measurement of the mRNA of the rat isozymes. Total RNA was isolated from rat prostate by a guanidinium isothyocyanate procedure, purified by centrifugation through CsCl, and quantified spectrophotometrically (20). Aliquots of total RNA (20 μg) were separated on a 1.2% formaldehyde agarose gel and transferred to a nylon membrane (Biodyne, Pall BioSupport, East Hills, NY). After UV cross-linking of the RNA, the membranes were prehybridized and hybridized at 42° with labeled DNA fragments in 50% formamide, 1 M NaCl, 10% dextran sulfate, 1% SDS, 50 mM Tris, pH 7.5, and 200 μg/ml denatured salmon sperm DNA. The plasmids pBSrT5αR1–1 and pCRrT5αR2–1 were labeled with [α-^{32}P]-dCTP (New England Nuclear) by random priming (Takara Shuzo) and used as probes for steroid 5α-reductase types 1 and 2, respectively. After hybridization, the membranes were washed twice at room temperature with 2×SSC, once at 65° for 30 min with 0.2×SSC/0.1% SDS, and once at room temperature with 0.2×SSC. Autoradiography was performed with Kodak XAR-5 film and two intensifying screens (DuPont).

Administration of the steroid 5α-reductase inhibitors in castrated mature rats. Mature male Wistar rats (Nihon Clea, Tokyo, Japan) were housed and fed for 1 week and then anesthetized with pentobarbital and castrated. After 7 days, the rats were given once daily oral administration of 3.2 and 32 mg/kg body weight of drug suspension for 5 consecutive days. FK143 and finasteride were suspended in 0.5% methylcellulose solution at final concentrations of 0.64 or 6.4 mg/ml and 0.064 or 0.64 mg/ml, respectively. T propionate in sesame oil was subcutaneously injected into the rats at a dose of 300 μg/kg body weight at the same time of drug administration. Six hours after the last dose administration, rats were sacrificed by exposure to carbon dioxide. The ventral prostates and seminal ves-

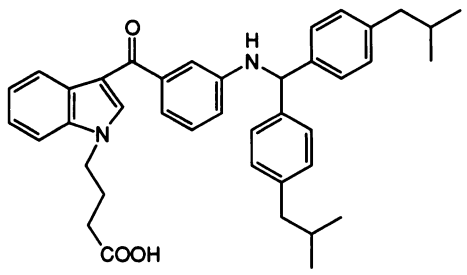
icles were removed, weighed, dipped into liquid nitrogen, and stored at −80°.

Results

Inhibitory activity and mode of inhibition of FK143 against rat and human isozymes of the steroid 5α-reductase. We prepared each isozyme from the 293 cell lines expressing cDNA encoding the steroid 5α-reductases to elucidate the inhibitory activity and mode of inhibition of FK143 against human and rat isozymes of the steroid 5α-reductase. The cDNAs for human type 2 and rat types 1 and 2 isozymes were cloned with the use of PCR amplification, and their sequences were confirmed to be identical with those reported previously. The specific enzyme activities of human types 1 and 2 and rat types 1 and 2 of the lysates for T transiently expresed in 293 cells were 41.1 ± 3.5, 208 ± 16, 492 ± 16, and 40.5 ± 1.8 pmol/min/mg, respectively. The pH profiles of the 5α-reductase activities of the human and rat types 1 and 2 were confirmed to be similar to those previously reported (Fig. 2; data for rat not shown). The optimum pH of the activity of the human type 1 was 7.0, whereas that of the human type 2 was shown to depend on the concentration of the substrate, namely, it shifted from 4.5 at 1 μM T to 6.5 at 4 nM T. The latter result coincides with the finding by Thigpen *et al.* (8) that the V_{max}/K_m of the human type 2 was maximal at neutral pH although V_{max} was maximal at acidic pH. Assays for the rat and human type 2 isozymes were performed at both pH 5.0 and 6.5.

FK143 is characterized by its nonsteroidal chemical structure (Fig. 1), which is in contrast with that of the known steroid 5α-reductase inhibitor finasteride, a 4-azasteroid (14). This structural characterization prompted us to investigate whether the mode of inhibition by FK143 is different than that of finasteride, which is known to be a competitive inhibitor. Our kinetic studies explicitly showed that FK143 inhibited noncompetitively both types 1 and 2 human and rat steroid 5α-reductases, whereas finasteride inhibited the enzyme in a competitive fashion (Figs. 3 and 4).

The inhibition constants of FK143 for human and rat steroid 5α-reductase isozymes were measured and compared with those of finasteride (Table 1). The K_i of finasteride for human type 1 was 321 nM, whereas that for human type 2 was 1.48 nM at pH 6.5 and 0.91 nM at pH 5.0. Therefore, finasteride is a type 2 selective inhibitor, as previously reported (7, 8). The inhibition constants (K_{ie} and K_{ies}) of FK143 for human type 1 isozyme were 27.0 and 19.6 nM, whereas those for human type 2 were 19.9 and 14.5 nM at pH 6.5 and 24.1 and 20.4 nM at pH 5.0, respectively. In contrast with finasteride, FK143 is characterized by its dual inhibition against both human isozymes.



4-[3-[3-[bis(4-isobutylphenyl)methylamino]benzoyl]-1H-indol-1-yl]butyric acid

Fig. 1. Structure of FK143.

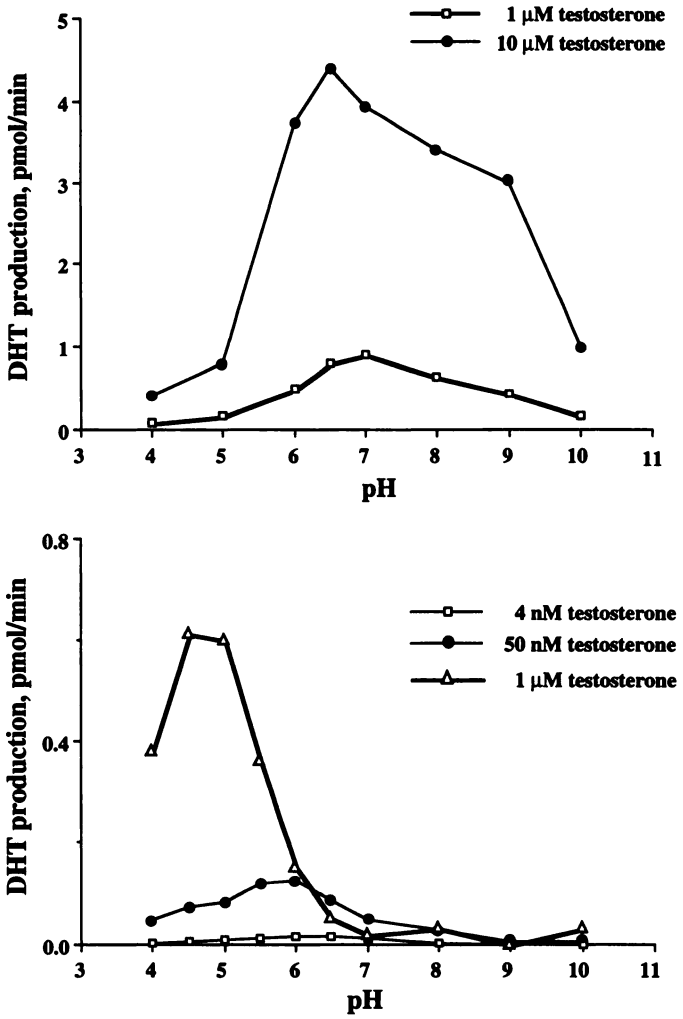


Fig. 2. Effect of substrate concentrations on pH profiles of types 1 and 2 isozymes. Each isozyme was prepared as described in the text from 293 cells transiently expressing the individual isozyme cDNA. The enzyme assays were performed as described in the text. The buffers used in the assay were sodium phosphate, pH 4–7, and Tris-citrate, pH 8–10. The protein concentrations in the enzyme preparations of types 1 and 2 used in each assay were 200 and 5 μg, respectively.

Species differences in the inhibitory activity of finasteride between human and rat were correlated to published data (6). Namely, the K_i of finasteride for rat type 1 was 11.0 nM, 29-fold less than that for human type 1. In contrast, the inhibition constant values of FK143 for rat types 1 and 2 were comparable to those for human isozymes.

It is also noteworthy that pH had negligible influence on the K_i values of FK143 and on those of finasteride for human and rat type 2, whereas the K_m values of T for human and rat type 2 isozymes were decreased 7.2- and 5.1-fold, respectively, when pH was shifted from 5 to 6.5.

Effect of FK143 on the transcription of the genes coding for the isozymes of the rat 5α-reductase. We studied the effects of FK143 on the *in vivo* expression of the rat steroid 5α-reductase genes. Male rats were castrated and, after a 7-day interval, given once daily a subcutaneous injection of T propionate, with or without oral administration of FK143 or finasteride, for 7 consecutive days. The ventral

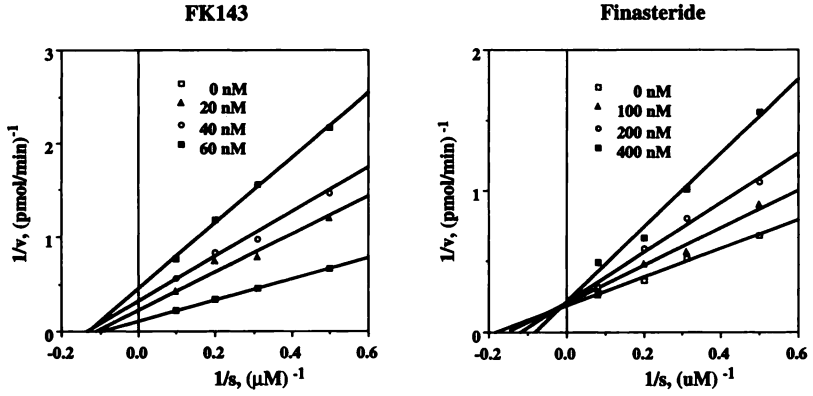


Fig. 3. Noncompetitive inhibition of human steroid 5α-reductase type 1 by FK143. Human steroid 5α-reductase type 1 was prepared as described in the text from the transiently expressing 293 cells. Aliquots of the enzyme preparation of 200 μg were assayed at pH 7.5 in the presence of varying concentrations (2–12 μM) of T and 2 mM NADPH for 20 min at 37° with or without varying indicated concentrations of FK143 or finasteride. *Points*, mean of triplicate determinations.

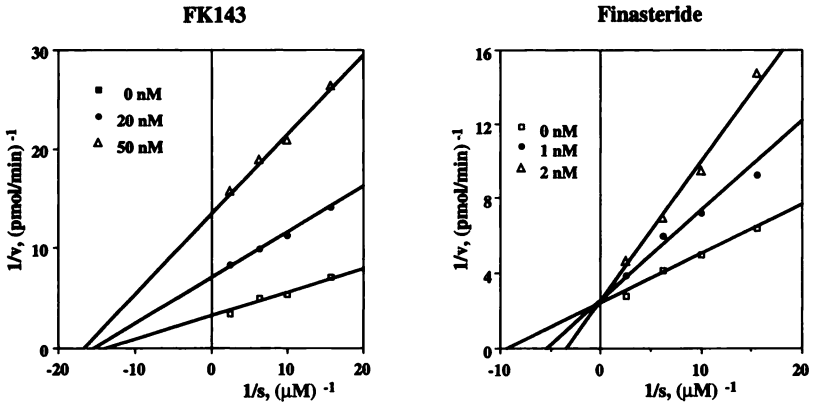


Fig. 4. Noncompetitive inhibition of human steroid 5α-reductase type 2 by FK143. Human steroid 5α-reductase type 2 was prepared as described in the text from the transiently expressing 293 cells. Aliquots of the enzyme preparation of 200 μg were assayed at pH 5 in the presence of varying concentrations (64–400 nM) of T and 2 mM NADPH for 20 min with or without varying indicated concentrations of FK143 or finasteride. *Points*, mean of triplicate determinations.

TABLE 1

Kinetic constants of the inhibitors against human and rat steroid 5 α-reductases

Human and rat steroid 5 α-reductase isozymes were prepared as described in the text. The enzyme assays were performed as described in the text. Human and rat type 1 isozymes were assayed at pH 7.5, and type 2 isozymes were assayed at pH 5 and 6.5. Protein quantities of the enzyme preparations used were 200 μg for human type 1, 5 μg for human type 2, 10 μg for rat type 1, and 2 μg for rat type 2. Each value represents the mean ± SE of three or four determinations for K_{ie} and K_{ies} and of seven to nine determinations for K_m.

Inhibitor/ substrate	Human								
	Type 1			Type 2					
				pH 5			pH 6.5		
	K_{ie}	K_{ies}	K_m	K_{ie}	K_{ies}	K_m	K_{ie}	K_{ies}	K_m
	nM	nM	μM	nM	nM	μM	nM	nM	μM
FK143	27.0 ± 4.3	19.6 ± 1.8		24.1 ± 1.4	20.4 ± 7.3		19.9 ± 4.3	14.5 ± 10.9	
Finasteride	321 ± 67.0			0.91 ± 0.02			1.48 ± 0.08		
Testosterone			9.1 ± 0.75			0.115 ± 0.009			0.016 ± 0.003
	Rat								
	Type 1			Type 2					
				pH 5			pH 6.5		
	K_{ie}	K_{ies}	K_m	K_{ie}	K_{ies}	K_m	K_{ie}	K_{ies}	K_m
	nM	nM	μM	nM	nM	nM	nM	nM	nM
FK143	97.2 ± 32.1	34.8 ± 2.0		15.3 ± 2.0	6.9 ± 1.3		16.6 ± 3.0	12.5 ± 0.5	
Finasteride	11.0 ± 0.6			0.38 ± 0.05			0.54 ± 0.08		
Testosterone			1.90 ± 0.06			14.5 ± 0.9			2.84 ± 0.24

prostates were removed, weighed, and analyzed for types 1 and 2 5α-reductase mRNAs. Both FK143 and finasteride exhibited inhibitory effects on the prostate growth with this experimental protocol (data not shown). Northern blot hybridization showed that type 1 5α-reductase mRNA was induced markedly by the administration of T and that this elevation of the level of type 1 mRNA was reduced by the administration of finasteride, as reported by George *et al.*

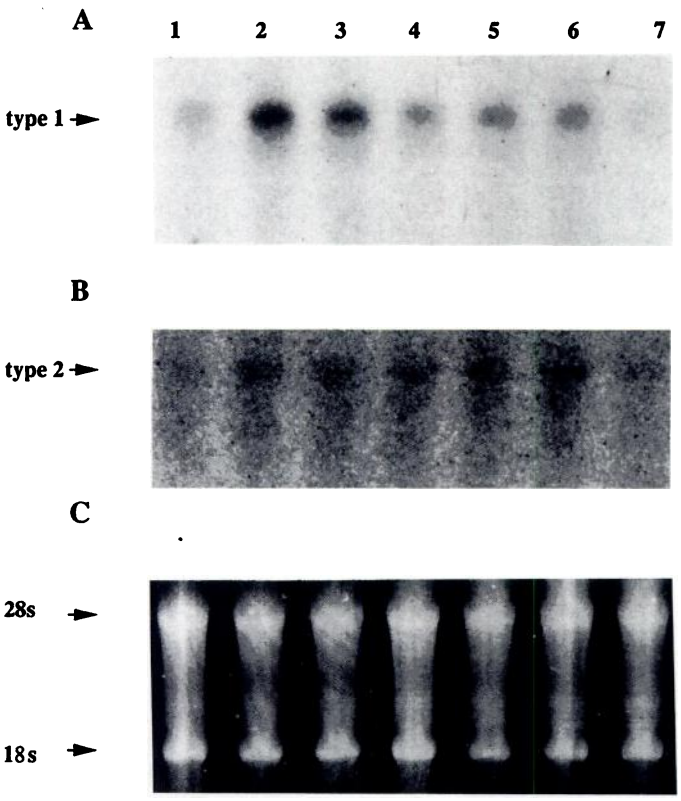


Fig. 5. Effect of FK143 on steroid 5α-reductase types 1 and 2 mRNA levels in rat prostate. Northern blot hybridization was performed as described in the text. Total RNAs were prepared from the pooled prostates of the rats that were castrated (n = 15) (*lane 1*), castrated and administered T (n = 3) (*lane 2*), castrated and administered T and FK143 (3.2 mg/kg) (n = 3) (*lane 3*), castrated and administered T and FK143 (32 mg/kg) (n = 3) (*lane 4*), castrated and administered T and finasteride (0.32 mg/kg) (n = 3) (*lane 5*), or castrated and administered T and finasteride (3.2 mg/kg) (n = 3) (*lane 6*) and of normal rats (n = 3) (*lane 7*). *Bottom*, loading control gel; *Top*, samples, in which 20 μg of the total RNA from each group was electrophoresed.

(21) (Fig. 5). FK143 was also shown to reduce the level of type 1 mRNA when induced by T.

Type 2 mRNA was shown to be induced by T, but contrary to the report by Normington *et al.* (12), Northern blot hybridization revealed that neither finasteride nor FK143 substantially affected the level of type 2 mRNA when induced by T (Fig. 5).

Discussion

FK143, a novel steroid 5α-reductase inhibitor, is characterized by its nonsteroidal chemical structure, which is in contrast with the existing steroid-based inhibitor finasteride. In the present study, FK143 was shown to noncompetitively inhibit both steroid 5α-reductases human and rat types 1 and 2, whereas finasteride inhibited the enzymes in a competitive manner. These results indicate that FK143 inhibits the enzymes by combining with the site or sites that are different from the active site where the substrate analogue inhibitors are bound. FK143 was also shown to be uncompetitive against the steroid 5α-reductase from the prostate with respect to NADPH, a cofactor of the steroid 5α-reductases.[1] Because of these properties, FK143 would be expected to exert inhibitory activity specifically on the steroid 5α-reductases and not on the other androgen-related enzymes or NADPH-requiring enzymes. FK143 was confirmed to uninhibit several androgen-related and NADPH-requiring enzymes (15).

FK143 was shown to inhibit both human isozymes equally, whereas finasteride inhibited type 2 enzyme approximately 300-fold more potently than type 1 enzyme. Because the type 2 isozyme is predominant over type 1 isozyme in human prostate and type 2 is suggested to have an anabolic role in steroid metabolism (12), a type 2-selective inhibitor may be sufficiently effective to treat BPH. However, although type 1 enzyme has lower activity than type 2 in human prostate, its existence in human prostate was demonstrated by Northern blot hybridization (6, 9). We cannot exclude the possibility that human type 1 isozyme activity may to some extent contribute to growth and maintenance of the prostatic gland. The rat type 1 isozyme was assumed to participate in growth and maintenance of the rat prostatic gland based on the finding of its localization in the basal epithelial cells of the rat ventral prostate. Alternatively, DHT synthesized by the human type 1 isozyme in the liver and skin can act as a true endocrine hormone and may contribute to androgen-dependent growth of the prostate (22). According to these propositions, a dual inhibitor of the steroid 5α-reductase isozymes may be more efficacious in treating BPH than type 1- or 2-selective agents.

The inhibition of the steroid 5α-reductase isozymes was found to vary with the species studied. Namely, finasteride was selective for the human type 2 enzyme, whereas it inhibited the rat types 1 and 2 equally. In contrast, FK143 was shown to be a dual inhibitor against both human and rat isozymes. This finding, taken together with the observation that the expression patterns of the isozymes in prostate differed human and rat, highlights how the clinical efficacy of the inhibitors may be postulated from data of their efficacy in the *in vivo* rat model.

FK143 and finasteride were shown in the present study to reduce the expression of rat type 1 mRNA. The reduction in the expression of type 1 mRNA by FK143 provided further support to the hypothesis of feedforward regulation by androgen of rat type 1 transcription (21). Furthermore, the action of FK143 against steroid 5α-reductase type 1 may be enhanced by synergism of its inhibitory activity with its suppression of the type 1 gene. In contrast to type 1 mRNA, we could not show a substantial reduction in rat type 2 mRNA by FK143 and finasteride. This observation is contradictory to the report by Normington *et al.* (12). One possible reason for the difference between our data and those of Normington *et al.* may be the difference in the experimental protocol of drug administration. Normington *et al.* administered daily subcutaneous injections of finasteride (25 mg/kg body weight/day) to Sprague-Dawley rats for 3 days, whereas we administered oral finasteride (3.2 mg/kg body weight/day) daily to Wistar rats for 7 days. However, there appears to be some difference in the regulation of gene expression in prostate between the steroid 5α-reductase types 1 and 2 as the differential profiles of the mRNAs of the two isozymes were obtained despite the use of the same prostates. Our results are consistent with the finding by Silver *et al.* (22) that short-term finasteride therapy did not substantially decrease steady state 5α-reductase type 2 protein levels, although the

[1] J. Hirosumi and O. Nakayama, unpublished observations.

species differences in regulation of the type 1 gene between human and rat should also be considered. Further studies will be required to clarify this contradiction.

In conclusion, we showed with the use of cloned isozymes that FK143 is a potent, noncompetitive, and dual inhibitor against types 1 and 2 isozymes of human and rat steroid 5α-reductases. FK143 will be useful in elucidating the participation of steroid 5α-reductase isozymes in the pathogenesis of BPH.

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