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PNU 157706, a Novel Dual Type I and II 5α-reductase Inhibitor

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PNU 157706 is a novel dual inhibitor of 5α-reductase (5α-R), the enzyme responsible for the conversion of testosterone (T) to 5α-dihydrotestosterone (DHT). Tested on a crude preparation of human or rat prostatic 5α-R, PNU 157706 caused enzyme inhibition with IC_{50} values of 20 and 34 nM, respectively, compared to the values of 32 and 58 nM shown by finasteride. Furthermore, PNU 157706 was highly potent in inhibiting human recombinant 5α-R type 1 and 2 isozymes, showing IC_{50} values of 3.9 and 1.8 nM and, therefore, it was several folds more potent than finasteride (IC_{50} values of 313 and 11.3 nM), particularly on the type I isozyme. PNU 157706 was shown to have no binding affinity for the rat prostate androgen receptor (RBA 0.009% that of DHT). In adult male rats, a single oral dose of 10 mg/kg of PNU 157706 caused a marked and longer lasting reduction of prostatic DHT than did finasteride (at 24 h inhibition by 89 and 47%, respectively). In prepubertal, T- or DHT-implanted castrated rats, PNU 157706, given orally for 7 days at the dose of 10 mg/kg/day, markedly reduced ventral prostate weight in T- but not in DHT-implanted animals, thus showing to be devoid of any anti-androgen activity. In adult rats treated orally for 28 days, PNU 157706 resulted markedly more potent (16-fold) than finasteride in reducing prostate weight, the ED_{50} values being 0.12 and 1.9 mg/kg/day, respectively. These results indicate that PNU 157706 is a promising, potent inhibitor of both type II and 1 human 5α-R with a very marked antiprostatic effect in the rat. © 1998 Elsevier Science Ltd. All rights reserved.

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INTRODUCTION

The steroid 5α-reductase (5α-R) enzyme catalyzes the irreversible reduction of testosterone (T) to the tissue-specific androgen 5α-dihydrotestosterone (DHT). T, the androgen secreted by the testis, is the main circulating androgen in man responsible for the increase in muscle mass, spermatogenesis and libido. In some tissues, such as the skin or the prostate, T functions as a prohormone which is converted to the more potent androgen DHT, through local 5α-R. DHT is recognized as the principal mediator of benign prostatic hyperplasia (BPH) and has also been implicated in the etiology of prostatic cancer and of some skin disorders, such as acne, androgenic alopecia and hirsutism [1, 2]. Inhibition of 5α-R provides a novel and selective approach to androgen deprivation in DHT-target tissues without affecting T-target structures. Finasteride [3], the prototype of 5α-R inhibitors, has been introduced into the market since 1992 for the therapy of symptomatic BPH [4], whereas another steroidal 5α-R inhibitor, epristeride [5, 6], is undergoing clinical evaluation.

Following the discovery of these inhibitors, the existence of at least two isoforms of 5α-R, namely type I and II, characterized by distinct molecular genetics, structural and biochemical properties and by different tissue localization, was reported [7, 8]. In these initial studies, the 5α-R type II was found to be mainly located in human urogenital tissues, such as the prostate, whereas the type I enzyme was found to be predominant in human nongenital skin, such as the scalp sebaceous glands and in liver [9, 10]. However, recent immunohistochemical data have shown that the type I isozyme is also expressed in the

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human normal, hyperplastic and cancerous prostatic tissue [11, 12]. In addition, high levels of human type I 5α-R enzymatic activity have been recently demonstrated in various human prostatic cancer cell lines, including the androgen-insensitive lines DU 145 and HPC-36M [13, 14] and the androgen-sensitive LNCaP [15]. Furthermore, type II 5α-R has been found in human scalp hair follicles [16]. Therefore, the respective physiological and pharmacological roles of 5α-R type I and II isozymes in the DHT-target tissues are yet to be fully elucidated.

Finasteride and epristeride have been subsequently characterized to be selective type II inhibitors [17–19]. However, finasteride has been found to have a lower clinical effectiveness than originally expected in the therapy of symptomatic BPH. This has been attributed to an incomplete suppression (60–70%) of circulating DHT [4, 20], due to the residual DHT synthesis through type I isozyme not inhibited by the compound. Therefore, a dual inhibitor of both type I and II 5α-R isoforms is expected to cause a better suppression of circulating DHT, that could result in a more efficient therapy of BPH [21].

Recently, non-steroidal [22, 23] and steroidal [21, 24] derivatives with dual inhibitory activity on human 5α-R have been described. In the last years we have evaluated the 5α-R inhibitory properties of a number of 4-azasteroids with fluoro-substituted 17β-amidic side chains [25, 26]. In the present paper we describe the compound PNU 157706 [*N*-(1,1,1,3,3,3-hexafluorophenylpropyl)-3-oxo-4-aza-5α-androst-1-ene-17β-carboxamide] (Fig. 1) which has been identified as a potent dual type I and II inhibitor of human 5α-R. The compound's inhibitory activity *in vitro*, as well as the preliminary evaluation of its pharmacological activity *in vivo* are here reported, in comparison with those of the reference compound finasteride.

MATERIALS AND METHODS

Chemicals

PNU 157706 and finasteride were synthesized at the Medicinal Chemistry Department of Pharmacia and Upjohn (Italy). Flutamide was kindly donated by Schering-Plough (Italy). [4-^{14}C]T (50 mCi/mmol), [1,2,6,7-^{3}H]T (97 Ci/mmol), [1,2,4,5,6,7-^{3}H]DHT (110 Ci/mmol) were purchased from Dupont® NEN, U.S.A. All the other unlabelled steroids and the protease inhibitors aprotinin and leupeptin were obtained from Sigma Chemical Co. (U.S.A.). Celite ready-to-use for steroid chromatography (Chromatolithe A®) was supplied by bioMérieux (France).

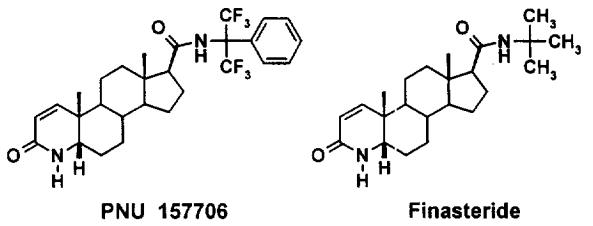

Fig. 1. Chemical structures of PNU 157706 and finasteride.

Preparation of the solutions

For the *in vitro* tests, stock solutions of the compounds were prepared in methanol and diluted with the assay buffer. The amount of methanol in the final incubation volume was always ≤3% and a similar amount of solvent was included in the control sample of each assay. For the *in vivo* studies the compounds were suspended in 0.5% Methocel (A-4C Premium, Dow Chemical, U.S.A.) containing 0.4% Tween 80 (Merck, U.S.A.).

Animals

Prepubertal 22-day-old, or adult (250 g body weight) male Crl:CD®(SD)BR rats were supplied by Charles River, Italy. Animals were fed a commercially available chow (Altromin MT, supplied by Rieper, Italy) and water was available *ad libitum*.

Inhibition of native prostatic 5α-R

Inhibition of the conversion of T to the 5α-reduced products DHT and 3α(β),17β-androstanediol was evaluated using the rat prostate and the human hyperplastic prostate as the enzyme sources. The particulate fraction (140,000*g* pellet) from homogenate of adult rat prostates was obtained as described by Liang *et al.* [27]. The assay was performed in a final incubation volume of 0.5 ml, in 40 mM phosphate buffer pH 6.5, containing 1 mM dithiothreitol, 0.5 mM NADPH and ≈700 μg of protein. The human prostatic tissue was homogenized in a w/v ratio of 1:3 with 100 mM Tris–HCl buffer, pH 7.0, containing 20% glycerol, 100 mM sodium citrate, 100 mM KCl, 1 mM EDTA and 5 mM dithiothreitol (buffer *a*). To protect the enzyme during the preparation process, 0.5 mM NADPH, 1 μM T and 10 μg/ml of the protease inhibitors aprotinin and leupeptin were also added to buffer *a* [28]. The microsomes were then isolated, the pellet resuspended in buffer *a*, also containing the protease inhibitors and stored at −80°C. Incubations were performed in a final volume of 0.5 ml in 100 mM Tris–HCl buffer, pH 5.5, containing the same ingredients as in buffer *a*, 0.5 mM NADPH and ≈300 μg of microsomal proteins. For both enzyme preparations, various concentrations of the inhibitors, in duplicate, were incubated with 1 μM [^{14}C]T for 30 min at 37°C. The samples were then processed as described previously [29].

Inhibition of human recombinant type I and type II 5α-R

Recombinant human prostatic 5α-R type I and type II were independently expressed using the baculo-

virus-directed insect cell expression system as previously described [30, 31]. The inhibitory properties of PNU 157706 and finasteride on the two isozymes were evaluated as described by Iehlé *et al.* [31], using 3 or 1 μM of [^{3}H]T as substrate for type I or II isozyme, respectively.

Binding affinity to the androgen receptor

Binding of PNU 157706 to cytoplasmic androgen receptor (AR) of rat prostate was determined by standard dextran-coated charcoal adsorption techniques, as described previously [29]. The assay was performed at 0°C for 2 h, in a final incubation volume of 0.4 ml containing an aliquot of cytosol, 1 nM of [^{3}H]DHT and various concentrations, in duplicate, of the tested compounds or unlabelled DHT. The relative binding affinity (RBA) of each compound was calculated according to the following formula: %RBA = 100×IC_{50} of DHT/IC_{50} of tested compound.

Effect on prostatic DHT and T in rats

The specific *in vivo* 5α-R inhibitory activity of PNU 157706 in comparison to finasteride was studied by measuring the prostatic concentrations of DHT and T at 6 and 24 h after single oral dosing (10 mg/kg) in adult rats. In order to avoid time-related differences in control hormone levels, groups of 6–7 adult rats were treated with test compounds 6 or 24 h before being sacrificed in a single setting. Control animals received the vehicle (2 ml/kg of 0.5% Methocel containing 0.4% Tween 80). At sacrifice, prostates were removed and immediately frozen on dry ice, then stored at −20°C for androgen hormone assays.

Effect on prostate weight in androgen supplemented rats

The *in vivo* inhibition of 5α-R was further studied by evaluating the antiprostatic effect in castrated rats supplemented with T, locally converted to DHT by prostatic 5α-R. In addition, a possible direct antiandrogenic effect, mediated through antagonism at the androgen receptor level, was evaluated in castrated rats supplemented with DHT. In both conditions, the effect of PNU 157706 was compared to that of the androgen receptor antagonist flutamide. 22-day-old male rats were castrated by scrotal incision in light ether anaesthesia. 7 days later, animals were implanted with a 1 cm long Silastic® tubing (602-265, Dow Corning, U.S.A.) filled with a mixture of 25% T (in cholesterol) or 100% DHT and treated orally for 7 consecutive days with PNU 157706 or flutamide, at the oral dose of 10 mg/kg/day (8 rats/group). 24 h after the last dose, rats were sacrificed and the ventral prostate and the seminal vesicles removed and weighed. T- or DHT-implanted rats treated with 0.5% Methocel containing 0.4% Tween 80 (5 ml/kg) served as controls, whereas castrated rats implanted with a Silastic tubing containing 100% cholesterol and treated with the vehicle served as castrated-only controls. The mean percentage of inhibition of the T- or DHT-induced hypertrophic response was calculated according to the following formula: %inhibition = $100 \times (C_A - D)/(C_A - C_C)$, where C_A, C_C and D are the mean organ weights of androgen-implanted control, castrated control and drug treated group, respectively.

Antiprostatic effect in adult rats

Groups of 8–10 adult male rats were treated orally with PNU 157706 at doses of 0.003, 0.03, 0.3 or 3 mg/kg, or finasteride (0.03, 0.3 or 3 mg/kg) once daily for 28 consecutive days. Rats treated with 0.5% Methocel containing 0.4% Tween 80 (2 ml/kg) served as controls. 24 h after the last dose the animals were sacrificed. The following organs were removed and weighed: ventral prostate, seminal vesicles, testes, epididymides and adrenals.

T and DHT determinations

Prostatic concentrations of T and DHT were measured by specific radioimmunoassays (RIA), after sample extraction and purification on celite columns. Each prostate sample was thawed and homogenized in 4 ml of acetone:acetonitrile mixture (1:1) with a Polytron apparatus. After extraction and centrifugation, the organic phase was dessicated, the dried extract was dissolved in 5% methanol aqueous solution and purified on C-18 Amprep minicolumns (Amersham, U.K.), using ethylacetate as the eluting solvent. The dried extract was then dissolved in 0.5 ml isooctane and applied to a celite column (1.2 g of Chromatolithe A® in a 5 ml glass pipette). Elution was then performed with increasing concentrations of ethylacetate in iso-octane (0, 6 and 22%). The fractions containing DHT (6% ethylacetate) and T (22% ethylacetate) were evaporated under N_2 and the dried samples were processed for RIA. T and DHT levels in the resuspended samples were estimated, in duplicate, by using the [^{3}H]T and [^{3}H]DHT RIA Kits supplied by bioMérieux (France) and ICN Biomedicals (U.S.A.), respectively. The final sensitivity for T and DHT assays were 0.3 and 1 ng/g of prostate, respectively (for a sample of at least 0.2 g).

RESULTS

Inhibition of native prostatic 5α-R

The abilities of PNU 157706 and finasteride to inhibit 5α-R from crude rat and human prostate preparations are reported in Table 1. PNU 157706 inhibited rat and human enzymes with IC_{50} values of 34 and 20 nM, respectively. In the same assay system finasteride showed IC_{50} values of 58 and 32 nM for the rat and the human enzyme, respectively.

Table 1. Inhibition of native rat and human prostatic 5α-reductases

Compound	IC_{50} (nM)	
	Rat	Human
PNU 157706	34 ± 3	20 ± 3
Finasteride	58 ± 7	32 ± 5

Results are the mean ± SE of 4 separate assays. Incubations were performed in the presence of 1 μM [^{14}C]testosterone.

Inhibition of human recombinant type I and II 5α-R

Data reported in Table 2 indicate that PNU 157706 is a very potent inhibitor of both human recombinant type I and II 5α-R, with IC_{50} values of 3.9 and 1.8 nM, respectively. As a comparison, finasteride showed IC_{50} values of 313 and 11.3 nM for the type I and II isozymes, thus resulting 80 and 6.3 times less potent than PNU 157706, respectively.

Androgen receptor binding affinity

The binding affinities of PNU 157706 and finasteride to the rat prostate androgen receptor are reported in Table 3. The reference standard DHT displaced [^{3}H]DHT from the AR with an IC_{50} of 1.7 nM. Both PNU 157706 and finasteride were found to have a very low binding affinity, with IC_{50} values of 20 317 and 6 370 nM, respectively (RBA 0.009 and 0.027% that of DHT).

Effect on prostatic DHT and T in rats

In a preliminary investigation of the time-course of DHT inhibitory activity by PNU 157706 in comparison to finasteride, prostatic DHT and T were measured 6 and 24 h after dosing with 10 mg/kg of each compound. 6 h after dosing, PNU 157706 and finasteride similarly reduced prostatic DHT concentrations by 74 and 80%, respectively (Fig. 2). 24 h after treatment, however, PNU 157706 caused a further reduction (to 89%) of prostatic DHT, whereas the effect of finasteride was far less evident (47% inhibition). As expected for 5α-R inhibitors, prostatic T increased 550 or 470% 6 h after dosing with PNU 157706 or finasteride, respectively. At 24 h the increase in prostatic T was 688 and 380% for PNU 157706 or finasteride, inversely correlated to the degree of tissue DHT inhibition.

Table 2. Inhibition of recombinant human type I and II 5α-reductases

Compound	IC_{50} (nM)	
	Human type I	Human type II
PNU 157706	3.9 ± 0.1	1.8 ± 0.3
Finasteride	313 ± 74	11.3 ± 2.6

Results are the mean ± SE of 3 separate assays. Incubations were performed in the presence of 3 or 1 μM [^{3}H]testosterone, for type I or II isozyme, respectively.

Table 3. Binding affinity to the rat prostate androgen receptor

Compound	IC_{50} (nM)[a]	% RBA[b]
Dihydrotestosterone (DHT)	1.7 ± 0.1[c]	100
PNU 157706	20 317 ± 225	0.009
Finasteride	6 370 ± 11	0.027

[a]Displacement of 1 nM [^{3}H]DHT.
[b]Relative binding affinity (DHT = 100%).
[c]Mean ± SE of 2 separate assays.

Effect on T- and DHT-stimulated prostate growth in rats

Seven-day oral treatment with 10 mg/kg/day of PNU 157706 or flutamide in castrated, T-implanted prepubertal rats, effectively reduced the ventral prostate (by 72 and 76%) and seminal vesicle growth (by 81 and 81%) induced by T-implant (Fig. 3). In the experiment comparing the effect of both compounds, at 10 mg/kg/day, in DHT-implanted rats, no inhibitory effect on prostatic and seminal vesicle growth resulted from PNU 157706, as expected for a 5α-R inhibitor. The antiandrogen flutamide, as expected from its mechanism of action, caused a significant reduction of prostate and seminal vesicle growth also in DHT-stimulated rats (78 and 93%, respectively).

Antiprostatic effect in adult rats

The effect of a 28-day oral treatment of adult rats with various doses of PNU 157706 and finasteride on androgen target organ weights is reported in Table 4 and Fig. 4. Both compounds caused a dose-related

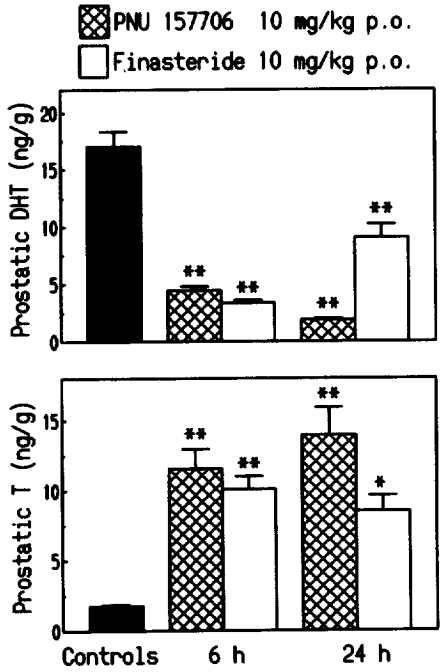

Fig. 2. Effect of a single oral dose (10 mg/kg) of PNU 157706 or finasteride on prostatic dihydrotestosterone (DHT) and testosterone (T) concentrations in adult rats. Rats were sacrificed 6 or 24 h after dosing. Mean ± SE of 6–7 rats per group. *$P < 0.05$; **$P < 0.01$ vs control group (Dunnett's test).

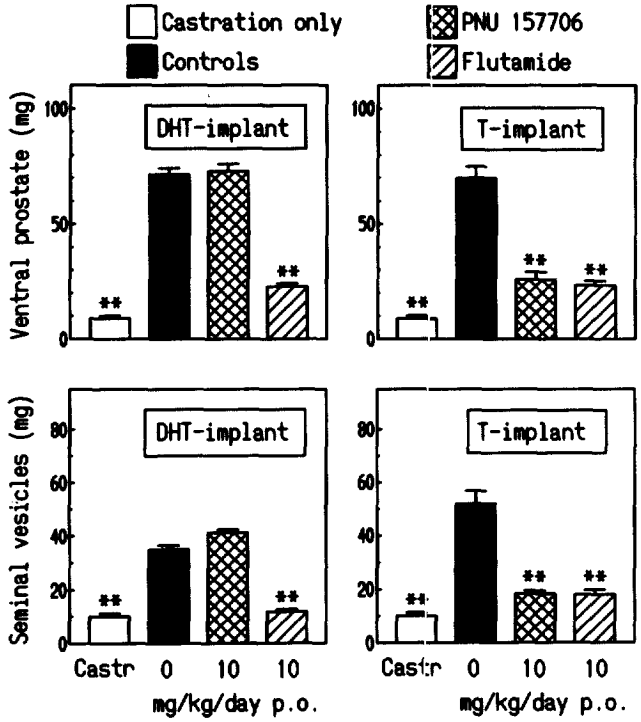

Fig. 3. Effect of 7-day oral dosing with 10 mg/kg/day of PNU 157706 or flutamide on ventral prostate and seminal vesicle weights in castrated, testosterone (T)- or dihydrotestosterone (DHT)-implanted rats. One group of castrated animals (Castr) was not treated with T or DHT. Mean ± SE of 8 rats per group. **$P < 0.01$ vs control group (Dunnett's test).

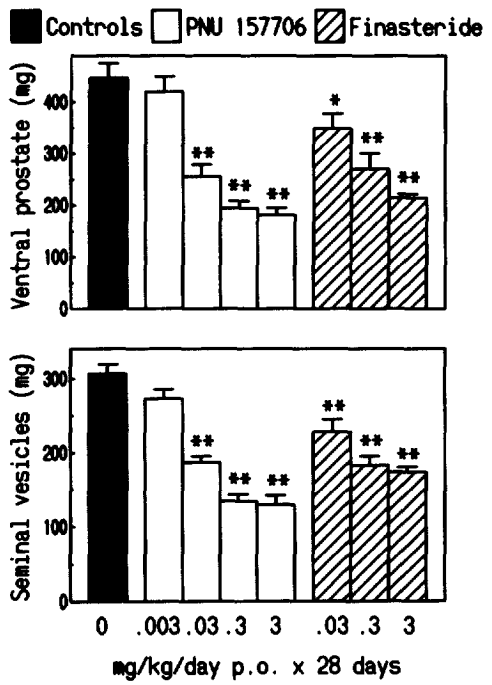

Fig. 4. Effect of 28-day oral dosing with various doses of PNU 157706 or finasteride on ventral prostate and seminal vesicle weights in adult rats. Mean ± SE of 8–10 rats per group. *$P < 0.05$; **$P < 0.01$ vs control group (Dunnett's test).

decrease in ventral prostate and seminal vesicle weights. PNU 157706 resulted 16 times more potent than finasteride in reducing prostate weight (ED_{50}, 0.12 and 1.9 mg/kg/day, respectively). Similarly, seminal vesicle weight was reduced more effectively by PNU 157706 (ED_{50}, 0.13 mg/kg/day) than by finasteride (ED_{50}>3 mg/kg/day). Both compounds did not show any effect on testicular and adrenal weights (data not shown), whereas a slight reduction of epididymis weight was observed at 3 mg/kg/day with both PNU 157706 and finasteride (8% for both compounds).

Table 4. Effect of 28-day oral dosing with PNU 157706 or finasteride on ventral prostate and seminal vesicle weights in adult rats

Compound	Dose (mg/kg/day)	Ventral prostate weight (mg)	Seminal vesicle weight (mg)
Vehicle	–	446.3 ± 28.7[a]	306.7 ± 12.4
PNU 157706	0.003	419.5 ± 29.8	272.6 ± 12.6
	0.03	255.3 ± 22.8[c]	186.5 ± 8.5[c]
	0.3	194.3 ± 14.1[c]	134.4 ± 9.3[c]
	3	181.5 ± 14.1[c]	129.3 ± 12.9[c]
Finasteride	0.03	348.3 ± 29.2[b]	228.0 ± 16.6[c]
	0.3	270.1 ± 29.5[c]	182.4 ± 12.7[c]
	3	214.0 ± 7.8[c]	173.5 ± 6.8[c]

[a]Mean ± SE (8–10 rats/group).
[b]$P < 0.05$ vs control group (Dunnett's test).
[c]$P < 0.01$ vs control group (Dunnett's test).

DISCUSSION

PNU 157706 is a novel 4-azasteroid selected among a series of compounds with fluoro-substituted-17β-amidic side chains for its potent *in vitro* and *in vivo* inhibitory activity on 5α-R. The compound has in fact been found to inhibit rat and human 5α-R in crude prostatic preparations with a potency respectively 1.7 and 1.6 times higher than that shown by finasteride. In this human prostate enzymatic preparation the type II isozyme is recognized to be the most prevalent form [32], whereas in the rat prostate enzymatic preparation both isozymes are represented [33], with a prevalence of the type I isozyme. Therefore, taking into account the experimental conditions (i.e. the pH) of enzyme activity evaluation, the *in vitro* results on crude enzyme preparations likely represent inhibition of the human type II isozyme for the human prostate and prevalent inhibition of the rat type I isozyme for the rat prostate. In this study, an efficient expression system for human 5α-reductases in eucaryotic cells was used to produce a biologically active 5α-R which exhibited a high specific activity and a high affinity for T [30, 31]. The kinetic characteristics of these recombinant 5α-reductases [31] were in agreement with those previously published in the literature [7]. The results here reported on recombinant human 5α-R type II in-

dicate that PNU 157706 is a potent inhibitor of this isozyme, its IC_{50} being 1.8 nM, thus confirming the data on the crude enzyme preparation. Very interestingly, PNU 157706 was also found to inhibit the recombinant human type I isozyme at low nanomolar concentrations (IC_{50}, 3.9 nM). In the same assay system finasteride was 6.3 and 80 times less potent than PNU 157706 on type II and I isozymes, respectively. PNU 157706 therefore appears to be a potent, dual inhibitor of human type I and II 5α-R. In addition, PNU 157706, like finasteride, was found to have no binding affinity to the rat prostate androgen receptor.

A preliminary evidence of *in vivo* inhibition of 5α-R activity by PNU 157706 was obtained by measuring the prostatic DHT and T concentrations after single oral dosing in adult rats. Because previous data [26, 34] had pointed out that oral finasteride maximally reduced prostatic DHT 4–8 h after dosing, the inhibitory effect of PNU 157706 or finasteride on prostatic androgen levels was studied at 6 and 24 h after a single oral treatment at 10 mg/kg. A similar degree of inhibition of prostatic DHT was observed 6 h after dosing with PNU 157706 and finasteride (74 and 80% respectively). Thereafter, at 24 h an even more marked inhibitory effect (89%) on prostatic DHT was observed in PNU 157706-treated rats, whereas finasteride was confirmed not to be a long lasting 5α-R inhibitor (inhibition of 47%).

In a further *in vivo* study, the inhibitory effect of PNU 157706 on 5α-R has been evaluated using the test for the antiprostatic effect in T-implanted, castrated rats. PNU 157706, at the oral dose of 10 mg/kg/day was found to reduce T-stimulated growth of the ventral prostate by 72%. A marked inhibitory effect on seminal vesicle growth (81%) was also observed. Such a test, however, does not discriminate between possible 5α-R inhibitory and androgen receptor antagonist effects. Therefore, the possible androgen receptor-mediated anti-androgenic activity of PNU 157706 has been evaluated in DHT-supplemented rats, in which prostatic hypertrophy results from direct DHT stimulation of the androgen receptor. In these animals, oral PNU 157706 dosing at 10 mg/kg/day did not antagonize the DHT-induced prostatic hypertrophy, whereas the androgen receptor antagonist flutamide was markedly effective. Therefore, PNU 157706 was found to be devoid of any direct androgen receptor antagonist property *in vivo*, thus confirming the *in vitro* results.

In addition, in intact adult rats, after repeated (28 days) oral dosing, PNU 157706 caused a very relevant reduction of prostate and seminal vesicle weights, without any effect on testicular and adrenal weights, thus showing to be a very selective 5α-R inhibitor. At the highest PNU 157706 dose tested, the compound's inhibitory effect on prostate and seminal vesicle weights was less marked than that usually caused by castration in similar experiments [29], as expected for a 5α-R inhibitor. In fact, it is well known that the marked DHT suppression in the rat prostatic tissue caused by 5α-R inhibition is generally coupled to increased T concentration [35–37], as also shown by PNU 157706 in the present study. Although T has a lower affinity for the androgen receptor than DHT [38], it can partly decrease the efficacy of a 5α-R inhibitor on prostate weight despite the marked suppression of DHT. The present results, however, indicate that the *in vivo* potency of PNU 157706 in reducing prostate weight (ED_{50}, 0.12 mg/kg/day) was 16-fold higher than that of finasteride (ED_{50}, 1.9 mg/kg/day), i.e. much higher than the 1.7-fold difference in potency observed in the *in vitro* study with rat 5α-R. To our knowledge no 5α-R inhibitor has been reported to have a prostate weight inhibitory potency higher than that shown by PNU 157706 in the present study.

All these data suggest that PNU 157706 can represent a major improvement in the field of 5α-R inhibitors, on account of its potent activity *in vitro* on both types of human 5α-R and of its long-lasting inhibitory effects on prostatic DHT *in vivo*, which can contribute to the marked antiprostatic effect it shows after repeated oral administration in rats.

Subsequently to the discovery of the 5α-R inhibitory property of finasteride and epristeride, both compounds have been characterized as selective inhibitors of the type II isozyme. More recently, a few compounds, such as LY 191704 [39], MK 386 [40] and L 751788 [41] have been described as selective inhibitors of the human type I isozyme. Clinical studies with selective type I or type II 5α-R inhibitors have shown that ≈30–40% of circulating DHT is formed by type I 5α-R [42], and ≈60–70% by the type II enzyme [4, 20, 42]. More recently, the first dual type I and II 5α-R inhibitor studied in man, i.e. the 4-azasteroid derivative GI 198745 [24], was found to suppress circulating DHT by ≈85–95% after single oral doses of 10–40 mg, thus resulting more effective than the standard dose of 5 mg finasteride [43]. As already mentioned, the residual circulating DHT, probably related to peripheral synthesis by type I 5α-R not inhibited by finasteride, may represent the cause for the limited clinical effectiveness of the compound. Therefore, dual inhibitors of human 5α-R (like GI 198745 and PNU 157706) could represent an advance in the treatment of BPH. Furthermore, these compounds could be considered an interesting approach to androgen dependent prostate cancer therapy; preclinical investigations on their possible role as single agents or as part of a combined therapy need to be performed.

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