Synthesis of Tricyclic Compounds as Steroid 5α-Reductase Inhibitors

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A series of 4-phenoxobutyric acid derivatives attached to a tricyclic skeleton were prepared and evaluated as 5α-reductase inhibitors. Structure activity relationships for these compounds in terms of rat epididymis (type 2) 5α-reductase inhibitory activities reveal that 1) the substitution pattern at the 11-position of dibenz[b,e]oxepin influenced potency, 2) higher lipophilicity of the tricyclic skeleton improved potency, whereas the existence of a basic nitrogen atom in this skeleton was detrimental to potency, and 3) isobutyl substitution at the 8 position of the azeptine skeleton was tolerated. Among the tricyclic compounds studied, 4-[3-[5-benzyl-8-(2-methyl)propyl]oxepin-2-carboxyamido]phenoxy]butyric acid (26) was the most potent inhibitor of rat type 2 5α-reductase at 0.1 μM.

Key words 5α-reductase; benign prostatic hyperplasia; structure–activity relationship; tricyclic compounds; substituent effect

5α-Reductase is an enzyme responsible for the conversion of testosterone (T) into the more potent androgenic metabolite, dihydrotestosterone (DHT). 5α-Reductase inhibitors may be a new type of drug for benign prostatic hyperplasia (BPH) and related disorders associated with elevated levels of DHT such as acne, 3) male pattern baldness, 4) and hirsutism. 5) With the discovery of two 5α-reductase isozymes, the physiological and pharmacological roles of these enzymes in BPH are the subject of current research. 6,7)

We previously reported indole derivatives, such as 1, exhibiting a potent inhibitory activity for rat prostatic (type 1) 5α-reductase with an IC50 value of 9.6±1.0 nM. 7) During the course of structure–activity relationship (SAR) studies of indole derivatives, we discovered that the bulky substituent at the N-1 position of indole was required for potent inhibition of type 1 5α-reductase. 8,9) These observations led us to design tricyclic compounds such as carbazoles and azeptines, which are bulkier than indoles.

When we started our research program, 2, (±)-ONOO-3805 10) was the only compound reported as a nonsteroidal 5α-reductase inhibitor. Consequently, we designed novel compounds analogous to 2 and considered that the lipophilic part of 2 corresponds to a steroidal skeleton. 10) The benzylxophenyl moiety of 2 can be transformed into dibenz[b,e]oxepin as follows: 1) the three bonds between two benzene rings in conformation a are appropriately rotated to conformation b, 2) then two benzene rings in b are bridged by a C1 unit to afford the dibenz[b,e]oxepin tricyclic system. Thus, we prepared dibenz[b,e]oxepins and evaluated them as 5α-reductase inhibitors.

The starting materials, 6,11-dihydrodibenz[b,e]oxepin-2-carboxylic acids (3–6) and 5,11-dihydrobenzoxepino[3,4-b]pyridine-7-carboxylic acid (7) were available by a known method. 11) Carbazole and azeptine carboxylic acids (8–13) were derived from the corresponding aldehydes. 12) Tricyclic carboxylic acids (3–13) were converted to butyric acids (15–27) by condensation with a 2-substituted aniline (14a,b) 13) using Mukaiyama’s reagent, 14) followed by hydrolysis of ethyl ester (see Chart 1).

The final tricyclic compounds (15–27) were evaluated for inhibitory activity against rat prostatic (type 1) and epididymis (type 2) 5α-reductases in the manner described previously. 7) Rat prostatic 5α-reductase showed a broad, neutral-to-basic, pH optimum, whereas the type 2 isozyme obtained from epididymis exhibited an optimal pH of 5.5. 12) Inhibitory activities, expressed as percent inhibition, are summarized in Table 1.

Initially, compounds 16 and 19 were tested for inhibitory effects both on rat prostatic (type 1) and epididymis (type 2) 5α-reductases. Though both compounds showed weak potency for type 1 isozyme, even at 1 μg drug concentration, they exhibited potent activity against type 2 isozyme at 0.1 μM. The 11-hydroxy derivative (19) exhibited 73% inhibition at 0.1 μM, which is better than the ketone (16). Thus, tricyclic compounds were more responsive to type 2 isozyme and discriminated between subtle structural differences in the same manner as our previous observation: non-steroidal inhibitors, such as indole derivatives and (±)-ONO-3805, more potently inhibited type 2 versus type 1 isozyme. 7,15) Consequently, we evaluated the tricyclic compounds for inhibitory activity against type 2 isozyme to investigate structure–activity relationships.

Oxepins with a methylene group at the 11-position showed more potent inhibitory activity than the ketone (17, 18 vs. 15, 16). Substitution of one of the benzene rings in dibenz[b,e]-oxepin with a pyridine ring apparently led to a loss of inhibitory activity (21 vs. 20). ClogP values (13) of tricyclic skeletons corresponding to 15, 17, 20 and 21, in which the left part of the molecule was replaced by the acetyl substituent, were calculated as 2.87, 3.98, 2.94 and 1.45, respectively. These results suggest that the higher lipophilicity in dibenz[b,e]oxepin is desirable for potent activity.

It is obvious that the substitution pattern at the 11-position of dibenz[b,e]oxepins influenced potency. Hydroxy and methoxy groups were more potent than ketone (19, 20 vs. 15, 16). From the result that compound 19 showed more potent activity than 16, it was concluded that substitution pattern rather than the lipophilicity of this moiety influenced the potency. The configuration of the 11-carbon atom affects the conformation of the dibenz[b,e]oxepin ring system, as we previously reported. 14) Ring inversion occurs easily in the case of the trigonal sp3 carbon, whereas such a conformational change hardly ever occurs in physiological conditions in tetrahedral sp3 carbon at the 11 position. 1H-NMR spectra
Fig. 1. Design of Tricyclic Compounds

Table 1. 5α-Reductase Inhibitory Activity of Tricyclic Compounds

<table>
<thead>
<tr>
<th>No.</th>
<th>A</th>
<th>X</th>
<th>Y</th>
<th>B</th>
<th>R²</th>
<th>R³</th>
<th>% inhibition (drug concentration)</th>
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<tr>
<td></td>
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<td></td>
<td></td>
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<tr>
<td>16</td>
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<td>S</td>
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<tr>
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a) % inhibition at 0.1 μM.
also indicated these phenomena. The C-6 methylene hydrogen atoms at the linkage of dibenz[b,e]azepin were observed as a singlet peak for the two protons in the case of ketoine at the 11-position (15—18). On the other hand, a doublet in the AB-pattern was observed in compounds possessing hydroxy or methoxy at the 11-position (19, 20). These observations suggest that one of the enantiomers in compound 19, 20 has a higher affinity against type 2 isozyme, and the ring inversion of dibenz[b,e]azepin might cause decreased potency. Replacement of the ether bond of the phenoxy part by thioether did not show any significant change in potency.

When compared with 22 and 23, dibenzepazine is relatively more potent than carbazol. The substituent \( R^2 \) on the nitrogen of azepine notably influenced inhibitory activity. The \( N \)-benzyl azepines (22 and 26) showed potent inhibitory activity. Non-substitution (\( N^3 H \)) retained potency, whereas substitution of the picolyl group caused a drop in potency, even though C log P values of the tricyclic moiety of 24 and 25 were over 4.5. From this result, the existence of a basic nitrogen atom of this moiety appears to be undesirable for potent inhibitory activity. As for the substituent \( R^3 \) in the 8 position of dibenzepazine, the introduction of isobutyl retained potency (26, 27 vs. 22, 24). This result indicates that bulkier substituents in the tricyclic ring systems are allowed as a lipophilic part of the molecule.

In conclusion, we designed tricyclic compounds, such as dibenzoxepins, carbazole and azepines, and evaluated them as \( \alpha \) reductase inhibitors. Several compounds showed potent inhibitory activity for rat type 2 \( \alpha \)-reductase, which were comparable with parent indole derivative 1 and \( \alpha \)-ONO-3805. These results reveal interesting features of the nonsteroidal \( \alpha \)-reductase inhibitors and provide a new prototype for novel synthetic candidates.

Experimental

Melting points were determined with a Büchi-510 melting point apparatus and are uncorrected. Infrared spectra (IR) were recorded on a Jasco IR-810 spectrometer. Proton nuclear magnetic resonance (1H-NMR) were recorded on a JEOL JNM GX-270 or EX-270 (270 MHz) spectrometer with Me\( _4 \)Si as an internal standard. Elemental analyses were performed by the analytical department of our laboratories.

8-Isobutyl-10,11-Dihydropyrido[1,2-a]pyrazine-5-carboxylic Acid (12) — To a solution of 12 (0.52 g, 1.30 mmol), 10 ml of AcOEt and 10 ml of AcOH, Pd/C (5 wt %, 0.13 g) was added. The reaction mixture was stirred under atmospheric hydrogen at 25°C for 48 h. After the reaction was complete, the mixture was filtered with Celite, and the filtrate was evaporated in vacuo to afford 12 (0.35 g, 62%) as a yellow amorphous. 1H-NMR (CDCl\(_3\)) : 6.90 (6H, d, \( J = 6.8 \) Hz), 1.80—1.85 (1H, m), 2.41 (2H, \( J = 6.9 \) Hz), 3.09—3.17 (4H, m), 6.91 (1H, dd, \( J = 2.0 \) Hz), 6.92 (1H, d, \( J = 2.0 \) Hz), 7.10 (2H, d, \( J = 8.6 \) Hz), 7.17—7.39 (5H, m), 7.51 (1H, dd, \( J = 1.9 \) Hz, 8.6 Hz), 7.57 (1H, d, \( J = 1.9 \) Hz), 9.80 (1H, s). To a solution of 2-aldehyde (0.4 g, 1.08 mmol) in methanol (20 ml), a 2.62 ml of 2M KOH in methanol and iodine (0.14 g, 0.54 mmol) were added and the resulting mixture was stirred at 0—5°C for 24 h. Methanol was evaporated in vacuo. The residue was dissolved in 1.01 of water and acidified with 4 M HCl to pH 5. The reaction mixture was extracted with AcOEt. The organic layer was washed with saturated aqueous Na\(_2\)SO\(_4\), and brine, successively, dried, and evaporated in vacuo. The yellow oil was chromatographed on silica gel and eluted with hexane–AcOEt (3:1) to afford 2-aldehyde (0.43 g, 74%), which was isolated as a pale yellow oil. 1H-NMR (CDCl\(_3\)) : 6.88 (6H, d, \( J = 6.7 \) Hz), 1.80—1.85 (1H, m), 2.39 (2H, d, \( J = 6.7 \) Hz), 3.19—3.27 (4H, m), 5.05 (2H, s), 6.91 (1H, dd, \( J = 9.1 \), 8.6 Hz), 6.92 (1H, d, \( J = 9.1 \) Hz), 7.79 (2H, d, \( J = 8.6 \) Hz), 7.51 (1H, dd, \( J = 1.9 \) Hz, 8.6 Hz), 7.57 (1H, d, \( J = 1.9 \) Hz), 9.80 (1H, s). To a solution of 2-aldehyde (0.4 g, 1.08 mmol) in methanol (20 ml), a 2.62 ml of 2M KOH in methanol and iodine (0.14 g, 0.54 mmol) were added and the resulting mixture was stirred at 0—5°C for 24 h. Methanol was evaporated in vacuo. The residue was dissolved in 1.01 of water and acidified with 4 M HCl to pH 5. The reaction mixture was extracted with AcOEt. The organic layer was washed with saturated aqueous Na\(_2\)SO\(_4\), and brine, successively, dried, and evaporated in vacuo. The yellow oil was chromatographed on silica gel and eluted with hexane–AcOEt (3:1) to afford 2-methoxyester (0.43 g, 74%), which was isolated as a pale yellow oil. 1H-NMR (CDCl\(_3\)) : 6.88 (6H, d, \( J = 6.7 \) Hz), 1.80—1.85 (1H, m), 2.41 (2H, d, \( J = 6.9 \) Hz), 3.09—3.17 (4H, m), 6.91 (1H, dd, \( J = 2.0 \) Hz), 6.92 (1H, d, \( J = 2.0 \) Hz), 7.10 (2H, d, \( J = 8.6 \) Hz), 7.17—7.39 (5H, m), 7.51 (1H, dd, \( J = 1.9 \) Hz, 8.6 Hz), 7.57 (1H, d, \( J = 1.9 \) Hz). The methyl ester was hydrolyzed to afford carboxylic acid (0.33 g, 59%) as tan amorphous. 1H-NMR (CDCl\(_3\)) : 1.78—1.84 (1H, m), 2.38 (2H, d, \( J = 6.8 \) Hz), 3.22 (4H, br s), 5.02 (2H, s), 6.87—6.91 (1H, m), 7.01—7.08 (2H, m), 7.16—7.28 (4H, m), 7.35 (2H, d, \( J = 6.9 \) Hz), 7.72 (1H, d, \( J = 2.0 \) Hz, 7.72 (1H, d, \( J = 2.0 \) Hz).
J = 7.1 Hz), 2.94 (2H, t, J = 7.1 Hz), 5.40 (2H, brs), 7.15—7.32 (5H, m), 7.40—7.70 (5H, m), 7.74 (1H, d, J = 8.8 Hz), 8.17 (1H, dd, J = 2.2, 8.6 Hz), 8.78 (1H, d, J = 2.2 Hz). IR (KBr) cm\(^{-1}\): 3430, 3335, 1732, 1668, 1643, 1519, 1487, 1300, 1254, 1174, 763. Anal. Calcd for C\(_{31}\)H\(_{29}\)N\(_3\)O\(_4\)· 0.5H\(_2\)O: C, 72.08; H, 5.85; N, 8.13. Found: C, 72.39; H, 5.60; N, 8.25.

4-[2]-[5-(2-Pyridyl)methyl-10,11-dihydropyridin-2-yl]azepin-2-ylcarboxamidophenoyl]butyric Acid (25) 25: As amorphous. 'H-NMR (CDCl\(_3\)) δ: 1.27—1.29 (2H, m), 2.59 (2H, J = 7.1 Hz), 2.35 (4H, brs), 4.14 (1H, d, J = 8.8 Hz), 6.89 (1H, d, J = 8.9 Hz), 6.89—7.07 (3H, m), 7.12—7.18 (4H, m), 7.34 (1H, d, J = 8.3 Hz), 7.52—7.60 (2H, m), 6.79 (1H, d, J = 1.7 Hz), 8.45—8.48 (2H, m), 8.52 (1H, s). IR (KBr) cm\(^{-1}\): 3416, 3493, 1620, 1508, 1543, 1451, 1250. Anal. Calcd for C\(_{34}\)H\(_{33}\)N\(_5\)O\(_5\): C, 72.08; H, 5.85; N, 8.13. Found: C, 72.24; H, 6.05; N, 8.03.

4-[2]-[5-Benzyl-10,11-dihydropyridin-2-yl]azepin-2-ylcarboxamidophenoyl]butyric Acid (26) 26: As amorphous. 'H-NMR (CDCl\(_3\)) δ: 6.86 (4H, d, J = 6.4 Hz), 1.77—1.82 (1H, m), 2.13—2.22 (2H, m), 2.36 (2H, d, J = 6.4 Hz), 2.52 (2H, t, J = 7.1 Hz), 2.30—3.30 (4H, m), 4.11 (2H, t, J = 6.1 Hz), 5.00 (2H, s), 6.85—7.30 (1H, m), 7.36 (2H, d, J = 8.3 Hz), 7.51 (1H, d, J = 8.6 Hz), 6.74 (1H, s), 8.44 (1H, d, J = 2.3, 7.3 Hz). IR (KBr) cm\(^{-1}\): 3345, 2950, 1706, 1610, 1530, 1493, 1452, 1334, 1250, 747. Anal. Calcd for C\(_{36}\)H\(_{39}\)N\(_5\)O\(_5\): C, 76.23; H, 6.84; N, 8.45. Found: C, 76.12; H, 7.10; N, 8.48.

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References and Notes

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16) a) Leo A., J. Chem. Rev., 93, 1281 (1993); b) The values were calculated by Cligno for Windows Version 1.0 BioByte Corp.).