N-BENZOYL-*N*-METHYLANDROSTAN-17β-AMINES; 20-AZA ANALOGUES OF BRASSINOLIDE*

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3β-Hydroxyandrost-5-en-17-one (**1**) was converted into 17β-(*N*-methylamino)androst-5-en-3β-ol (**4**). In the corresponding *N*-benzamide **5**, structural features characteristic of brassinolide were produced in a standard way, *i.e. via* 3α,5α-cyclo derivatives **7** and **8**, Δ^2-olefin **9** and 2α,3α-diol **10**. Baeyer–Villiger oxidation yielded two products: 2α,3α-dihydroxy-17β-*N*-(methylbenzamido)-7-oxa-7a-homo-5α-androstan-6-one (**11**) and 2α,3α-dihydroxy-17β-(*N*-methylbenzamido)-6-oxa-7a-homo-5α-androstan-7-one (**12**).
Key words: Steroids; Brassinosteroids; Leuckart reaction; Aza steroids; ^{1}H NMR spectroscopy; Baeyer–Villiger oxidation; *N*-Methylmorpholine *N*-oxide.

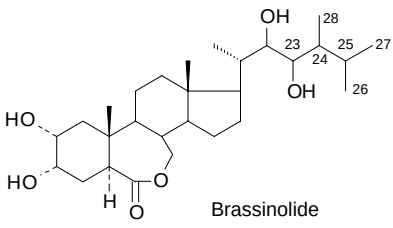
In the past, we have prepared and tested 21-nor-20-oxa[2,3] analogues of brassinolide and corresponding 23-oxa and 23-aza analogues[4,5]. 22-Oxa analogues have so far been prepared[6] with the reversed configuration at carbon C(20) though the synthesis of isomers with the natural C(20)-configuration is under way. Preparations of all these analogues with a hetero atom in the side chain have been prompted by two motives: by the desire to find a more available analogue applicable in agriculture and by structure activity considerations.

Here we present a synthesis of 20-aza analogue that was undertaken for the same two reasons: first, an aza derivative is readily available[7] from androstane starting material and unlike the oxa analogue, it offers the option to include a methyl group (*i.e.* carbon C(21)). And secondly, the aromatic ring represents a conformationally fixed part of the brassinolide side chain (compare carbons C(23) to C(26) of brassinolide and its analogue). The effect of rigidity of the brassinolide side chain has already been discussed[8].

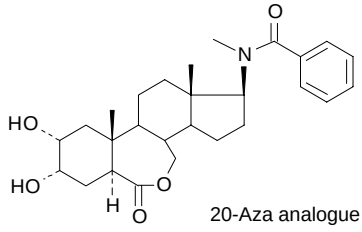
The starting material, 3β-hydroxyandrost-5-en-17-one (**1**), was converted to a mixture of amide **2** and its formate **3** by Leuckart reaction (see Scheme 1). Analytical samples were separated and identified by comparison with authentic samples. The *N*-formylamino

* Part CCCXCVI in the series On Steroids; Part CCCXCV see ref.[1].

group was not stable enough to survive further modifications of the A ring (*e.g.* tosylation of the 3β-hydroxyl group[9]). Therefore we converted the mixture of compounds **2** and **3** into 17β-methylamino derivative **4** by lithium aluminium hydride reduction[10]. This compound turned out to be a useful intermediate for the synthesis of 20-aza steroids. This time we used it for the synthesis of an analogue with the aromatic ring in the side chain.



Brassinolide



20-Aza analogue **11**

To this effect, a 2-methyl-2-propanol solution of amine **4** was treated with benzoyl chloride in the presence of aqueous potassium hydroxide according to Schotten–Baumann which afforded benzamide **5**.

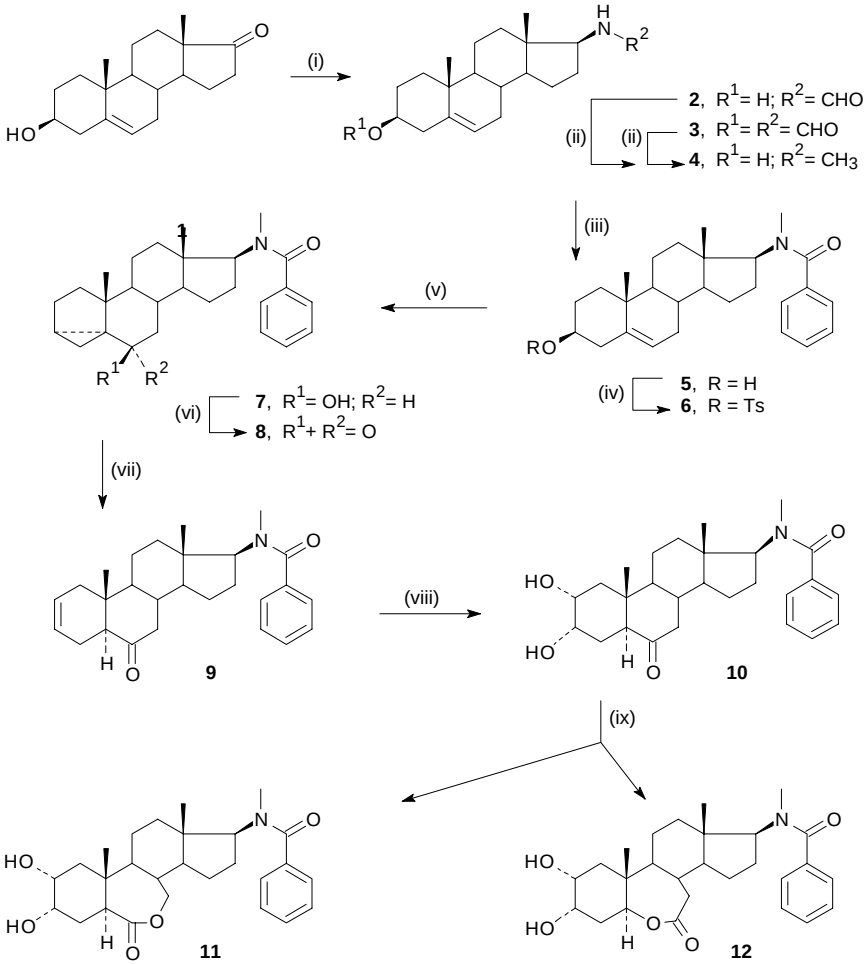
Further transformations of substituents in the A and B rings follow the general pattern of our previous syntheses[1,11]. It consists in a sequence of tosylation of the 3β-hydroxy group, solvolysis of tosylate **6** with isosteroid rearrangement under the formation of 3α,5α-cyclosteroid alcohol **7**, its oxidation to 3α,5α-cyclosteroid ketone **8** and isomerization to oxoalkene **9**. Osmium tetroxide catalyzed oxidation with *N*-methylmorpholine *N*-oxide resulted in the formation of 2α,3α-diol **10** and, finally, oxidation with trifluoroperoxyacetic acid yielded products of the Baeyer–Villiger oxidation, δ-lactones **11** and **12** (mixture 4 : 1). Careful chromatography on silica gel led to their separation: the desired lactone with the natural structure (compound **11**) was the less polar isomer. The use ^{1}H NMR spectroscopy made identification of both isomers possible: the desired lactone **11** shows signals of a $CHCOOCH_2$ group (one-proton doublet of doublet at δ 3.09 and two-proton multiplet at δ 4.02, respectively) while a signal of a CH_2COOCH group characterizes isomeric lactone **12** (one-proton doublet of doublet at δ 4.56; signals of 7a-protons were not observed).

The brassinolide-type activity of analogues of castasterone and brassinolide, compounds **10–12**, will be discussed elsewhere.

EXPERIMENTAL

Melting points were determined on a micro melting point apparatus Boetius and are uncorrected. Analytical samples were dried over phosphorus pentoxide at 50 °C/100 Pa. Optical rotations were measured in chloroform and $[\alpha]_D$ values are given in ° (10^{-1} rad cm^2 g^{-1}). IR spectra of chloroform solutions were recorded on a Bruker IFS 88 spectrometer. Wavenumbers are given in cm^{-1}. ^{1}H NMR spectra were measured on a Varian UNITY-200 (at 200.04 MHz) spectrometer at 23 °C in deuterio-

chloroform with tetramethylsilane as internal standard. Chemical shifts are given in ppm (δ-scale), coupling constants (*J*) and width of multiplets (*W*) in Hz. Thin-layer chromatography (TLC) was done on silica gel (ICN Biochemicals). Characteristic ^{1}H NMR parameters of compounds **2–12** are given in Table I. Preparative thin-layer chromatography (PLC) was carried out on 200 × 200 mm plates coated with a 0.7 mm layer of the same material. Whenever aqueous solutions of hydrochloric



(i) $HCONH_2$, HCOOH; (ii) $LiAlH_4$, dioxane; (iii) C_6H_5COCl, aq. KOH; (iv) TsCl, pyridine; (v) AcOK, water; (vi) Jones reagent, acetone; (vii) pyridinium 4-toluenesulfonate, LiBr, $CH_3CON(CH_3)_2$; (viii) OsO_4, *N*-methylmorpholine *N*-oxide, acetone, water; (ix) CF_3CO_3H, CH_2Cl_2

SCHEME 1

acid and potassium carbonate were used, their concentration is always 5%. Sodium chloride and potassium hydrogen carbonate were used in saturated aqueous solutions.

17β-(Formylamino)androst-5-en-3β-ol (**2**) and 17β-(Formylamino)androst-5-en-3β-yl Formate (**3**)

Formamide (8.0 g, 177.6 mmol) was added to a solution of 3β-hydroxyandrost-5-en-17-one (**1**, 10.0 g, 34.7 mmol) in formic acid (80%, 12 ml), and the mixture was stirred and heated to 175 °C. After 5 h, the mixture was cooled and diluted with water (80 ml). The precipitate formed a mixture of amides **2** and **3** was filtered off, washed with methanol and dried. Analytical samples of the two major components were obtained by PLC: 100 mg of the mixture was chromatographed on four preparative plates in chloroform–methanol (9 : 1). The polar zones yielded 43 mg (39%) of compound **2**, which was crystallized from dioxane; m.p. 258–263 °C, ref.[7] gives m.p. 260–265 °C. The lipophilic zones yielded 53 mg (44%) of compound **3**, which was crystallized from ethanol, m.p. 251–253 °C, ref.[10] gives m.p. 252–253 °C.

17β-(Methylamino)androst-5-en-3β-ol (**4**)

A mixture of compounds **2** and **3** (7 g) was placed in a Soxhlet extractor attached to a flask with lithium aluminum hydride (1.8 g, 47.2 mmol) in dioxane (134 ml). The mixture was heated to reflux under nitrogen until all the starting compound was dissolved (*ca* 1 h). The mixture was cooled and

TABLE I
Characteristic ^{1}H NMR parameters (δ, ppm) of compounds **2–12** in $CDCl_3$

Compound	H-18[a]	H-19[a]	NCH_3[a]	H-3[b]	Other signals[c]
2	0.76	1.05	–	3.52	5.38[d], 8.03[e]
3	0.71	1.03	–	4.68	5.36[d], 8.03[e], 8.21[f]
4	0.72	1.02	2.44	3.52	5.36[d]
5	0.82	1.00	3.00	3.50	5.31[d]
6	0.80	0.96	3.00	4.30	5.28[d], 2.44[g]
7	0.87	1.05	3.00	–	3.25[h], 0.28 and 0.52[i]
8	0.86	1.00	3.00	–	0.74[j]
9	0.71	0.82	2.99	5.62[k]	
10	0.74	0.80	2.98	4.02[l]	3.71[m], 2.66[n]
11	0.83	0.90	2.98	4.02[o]	3.09[n], 3.62[m]
12	0.82	0.92	2.98	3.96[p]	3.63[m], 4.56[n]

[a] Singlet, 3 H; [b] multiplet, 1 H (W = 40), unless stated otherwise; [c] compounds with an aromatic ring (all except **2–4**) exert multiplets (5 H) around 7.37 and 8.05; [d] doublet, 1 H (J = 6), H-6; [e] singlet, 1 H, N-CHO; [f] singlet, 1 H, O-CHO; [g] singlet, 3 H, tosylate-CH_3; [h] multiplet, 1 H (W = 13), H-6α; [i] multiplets, 2 × 1 H (W = 18 and W = 14), two cyclopropane protons; [j] multiplet, 1 H (W = 18), cyclopropane proton; [k] overlapped signals of H-2 and H-3 (5.48–5.71), 2 H; [l] multiplet, 1 H (W = 16), H-3β; [m] multiplet, 1 H (W = 24), H-2β; [n] doublet of doublet, 1 H (J = 5, J' = 12), H-5α; [o] multiplet, 3 H (W = 30), H-3β and 2 × H-7a; [p] multiplet, 1 H (W = 16), H-3β.

an excess reagent decomposed with a saturated aqueous solution of sodium sulfate. The inorganic material was filtered out and washed with chloroform. The filtrate was dried over anhydrous sodium sulfate, evaporated *in vacuo* and the residue was crystallized from acetone (4.42 g, 69%); m.p. 209–211 °C, ref.[10] gives m.p. 210.5–212.5 °C.

17β-(*N*-Methylbenzamido)androst-5-en-3β-ol (**5**)

Potassium hydroxide (0.34 g, 6.0 mmol) in minimum amount of water was added to a solution of compound **4** (0.7 g, 2.31 mmol) in 2-methyl-2-propanol (20 ml) and benzoyl chloride (0.7 g, 5.0 mmol) was added under rapid stirring. After 5 min, the mixture was cooled by ice, a precipitate was filtered off, washed with the sodium chloride solution, dissolved in chloroform and dried over anhydrous sodium sulfate. Evaporation of solvent gave compound **5** (0.92 g, 97%). Analytical sample was obtained by crystallization from acetone; m.p. 258–261 °C, $[\alpha]_D$ –103 (*c* 1.8). IR spectrum: 3 609 (O–H); 1 665 (C=C); 1 617 (amide); 1 059 (N–CH_3). For $C_{27}H_{37}NO_2$ (407.6) calculated: 79.56% C, 9.15% H, 3.44% N; found: 79.37% C, 8.90% H, 3.27% N.

17β-(*N*-Methylbenzamido)androst-5-en-3β-yl Tosylate (**6**)

4-Toluenesulfonyl chloride (100 mg, 0.52 mmol) was added to a solution of 3β-hydroxy compound **5** (100 mg, 0.25 mmol) in pyridine (1 ml). After 48 h, the mixture was diluted with the sodium chloride solution. The precipitate was extracted with chloroform and the extract washed with water, dilute hydrochloric acid, the potassium hydrogen carbonate solution and dried over anhydrous sodium sulfate. Evaporation of the solvent *in vacuo* to dryness gave compound **6**, which crystallized from acetone (110 mg, 80%); m.p. 173–174 °C, $[\alpha]_D$ –103 (*c* 1.0). IR spectrum: 1 666 (C=C); 1 637 (amide); 1 496, 1 306, 1 189 (tosylate). For $C_{34}H_{43}NO_4S$ (561.8) calculated: 72.69% C, 7.72% H, 2.49% N, 5.71% S; found: 72.91% C, 7.92% H, 2.62% N, 5.21% S.

17β-(*N*-Methylbenzamido)-3α,5-cyclo-5α-androstan-6β-ol (**7**)

Potassium acetate (6.5 g, 66.2 mmol) and water (31 ml) were added to a solution of tosylate **6** (3.28 g, 5.84 mmol) in acetone (95 ml). The reaction mixture was refluxed for 5 h, cooled, poured into water and extracted with ether. The ethereal phase was washed with water, dried over anhydrous magnesium sulfate and the solvent was evaporated to dryness. The residue (2.3 g) crystallized from toluene (2.1 g, 88%); m.p. 182–184 °C, $[\alpha]_D$ –24 (*c* 1.3). IR spectrum: 3 605 (O–H); 1 617 (amide). For $C_{27}H_{37}NO_2$ (407.6) calculated: 79.56% C, 9.15% H, 3.44% N; found: 79.41% C, 8.97% H, 3.31% N.

17β-(*N*-Methylbenzamido)-3α,5-cyclo-5α-androstan-6-one (**8**)

Alcohol **7** (2.83 g, 6.94 mmol) was oxidized with Jones reagent in acetone (50 ml) until purple colour of the mixture persisted for 2 min. After standing for 5 min at room temperature, 2-propanol (2 ml) was added and after 5 min the mixture was poured into water. The precipitate was extracted with chloroform, washed with water, the potassium hydrogen carbonate solution, water and dried over anhydrous sodium sulfate. Evaporation of the solvent gave 2.45 g (87%) of compound **8** which crystallized from acetone–heptane; m.p. 212–213 °C, $[\alpha]_D$ –23 (*c* 1.2). IR spectrum: 1 690 (C=O); 1 638 (amide). For $C_{27}H_{35}NO_2$ (405.6) calculated: 79.96% C, 8.70% H, 3.45% N; found: 79.82% C, 8.68% H, 3.35% N.

17β-(*N*-Methylbenzamido)-5α-androst-2-en-6-one (**9**)

Pyridinium 4-toluenesulfonate (18.8 mg, 0.10 mmol) and lithium bromide (21.2 mg, 0.24 mmol) were added to a solution of compound **8** (200 mg, 0.49 mmol) in *N*,*N*-dimethylacetamide (4 ml). The reaction mixture was heated at 160 °C under nitrogen for 6 h. After cooling, the mixture was poured into water and the product was extracted with chloroform, washed with dilute hydrochloric acid, water, the potassium hydrogen carbonate solution, water, and dried over anhydrous sodium sulfate. After evaporation of the solvent, the residue was crystallized from acetone–heptane (116 mg, 58%); m.p. 257–266 °C, $[\alpha]_D$ –54 (*c* 1.2). IR spectrum: 1 704 (C=O); 1 657 (C=C); 1 620 (amide). For $C_{27}H_{35}NO_2$ (405.6) calculated: 79.96% C, 8.70% H, 3.45% N; found: 79.88% C, 8.78% H, 3.52% N.

2α,3α-Dihydroxy-17β-(*N*-methylbenzamido)-5α-androstan-6-one (**10**)

Alkene **9** (130 mg, 0.32 mmol) in acetone (6.5 ml) was treated with a solution of OsO_4 (13 mg, 0.05 mmol) in 2-methyl-2-propanol (0.13 ml) and *N*-methylmorpholine *N*-oxide (180 mg, 1.54 mmol) in water (0.18 ml) at room temperature. After 5 h, an aqueous solution of sodium sulfite (10%, 7 ml) was added and stirring continued for 1 h. The mixture was poured into water and the product was taken up in chloroform. The chlorofom extract was washed with water, dilute hydrochloric acid, water, the potassium hydrogen carbonate solution, water, and dried over anhydrous sodium sulfate. Evaporation of the solvent gave compound **10** which was crystallized from acetone–water (82 mg, 58%); m.p. 277–279 °C, $[\alpha]_D$ –61 (*c* 1.3). IR spectrum: 3 613, 3 567, 3 399 (O–H); 1 705 (C=O); 1 617 (amide). For $C_{27}H_{37}NO_4$ (439.6) calculated: 73.77% C, 8.48% H, 3.19% N; found: 73.79% C, 8.65% H, 3.09% N.

2α,3α-Dihydroxy-17β-(*N*-methylbenzamido)-7-oxa-7a-homo-5α-androstan-6-one (**11**) and
2α,3α-Dihydroxy-17β-(*N*-methylbenzamido)-6-oxa-7a-homo-5α-androstan-7-one (**12**)

Ketone **10** (200 mg, 0.44 mmol) in dichloromethane (3 ml) was mixed with a solution of trifluoroperoxyacetic acid, freshly prepared from trifluoroacetic anhydride (646 mg, 3.08 mmol) and hydrogen peroxide (30%, 105 mg) in dichloromethane (3 ml). After standing for 3 h at room temperature, the reaction mixture was poured into water and a product was extracted with chloroform. The extract was washed with water, the potassium hydrogen carbonate solution (briefly), water, and dried over anhydrous sodium sulfate. After evaporation of solvent, the residue was chromatographed on 10 preparative plates in 2-propanol–chloroform (1 : 9). The lipophilic zones yielded 95 mg (46%) of lactone **11**; m.p. 245–249 °C (MeOH–ether), $[\alpha]_D$ –17 (*c* 1.3). IR spectrum: 3 608, 3 576, 3 424 (O–H); 1 722 (lactone); 1 619 (amide). For $C_{27}H_{37}NO_5$ (455.6) calculated: 71.18% C, 8.19% H, 3.07% N; found: 71.12% C, 8.03% H, 2.86% N.

The polar zones from the preparative plates gave 22 mg (11%) of oily lactone **12**, $[\alpha]_D$ –20 (*c* 1.3). IR spectrum: 3 609, 3 568, 3 399 (O–H); 1 721 (lactone); 1 619 (amid). For $C_{27}H_{37}NO_5$ (455.6) calculated: 71.18% C, 8.19% H, 3.07% N; found: 71.91% C, 8.04% H, 2.98% N.

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