Eur J Med Chem (1996) 31, 241–247

Short communication

Synthesis and biological activity of azasteroidal [3,2-*c*]- and [17,16-*c*]pyrazoles

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(Received 6 June 1995; accepted 8 November 1995)

Summary — Cholesterol, testosterone acetate and dehydroepiandrosterone acetate were used as starting materials for the preparation of azasteroidal [3,2-*c*]- and [17,16-*c*]pyrazole derivatives. In case of the 4-aza androstane series, a mixture of 5α/5β epimers **8** was obtained, which were separated by chemical methods. The compounds **4**, **5**, **10**, **12**, **13**, **15**, **16** and **18**–**22** were screened for antiinflammatory activity using the carrageenan rat paw oedema model. Oxirane **22** was found to be around ten times more potent than hydrocortisone. Evaluation of compounds **14**, **18** and **19** for their antineoplastic activity was also carried out at the National Cancer Institute, Bethesda, MD, USA, using standard procedures.

azasteroid / pyrazole derivative / antiinflammatory activity / antineoplastic activity / carrageenan test

Introduction

There has been considerable interest in the synthesis and biological study of several heterocyclic steroids as extremely potent antiinflammatory agents [1–4]. It has been reported that 2'-phenyl-11β, 17α, 21-trihydroxy-16α-methyl-4-pregneno[3,2-*c*]pyrazol-20-one-21-acetate and its *p*-fluorophenyl analogue were 60 and 100 times as active as hydrocortisone, respectively [1, 5]. The importance of the [3,2-*c*]pyrazole function has been demonstrated by a number of investigators [1, 2, 6]. Both cortivazol and nivazol have the [3,2-*c*]-pyrazole structural component, and the 3-keto function is absent; both have been described as potent anti-inflammatory steroids [2, 7–9]. The chemistry and pharmacology of several heterocyclic steroids have been reported from this laboratory [10–12]. The observation that [3,2-*c*]pyrazoles bring about notable changes in pharmacological activity and our previous experience with related studies led us to further study azasteroidal [3,2-*c*]- and [17,16-*c*]pyrazole derivatives in cholestane and androstane series, which we report herein.

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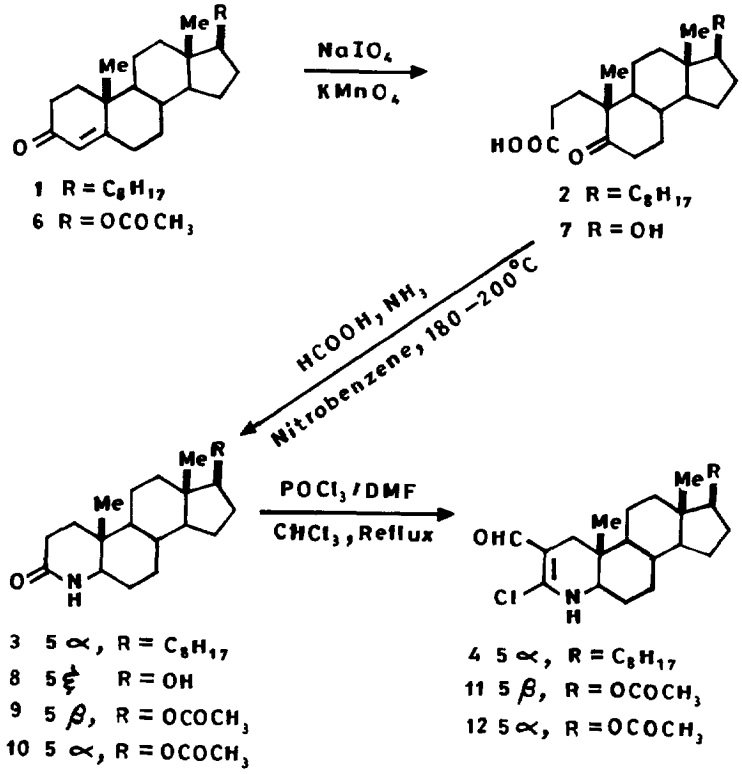
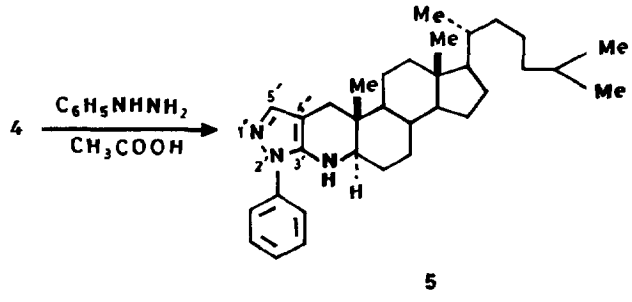
Chemistry

The exploratory work was first carried out in the cholestane series. 3-Chloro-4-aza-5α–cholest-2-en-2-aldehyde **4** was prepared via known procedures [13–16] as shown in scheme 1. It showed a proton singlet at δ 9.80 in the NMR spectrum. Refluxing **4** with phenylhydrazine gave product **5** (scheme 2). The NMR spectrum showed a multiplet at δ 7.23–7.26 for aromatic protons and 5'-C*H*. The phenyl group is at position 2' and not 1'. This is expected as phenylhydrazine has its more nucleophilic nitrogen β to the phenyl group.

A similar sequence of reactions was carried out for the synthesis of 4-aza-5α-androstano [3,2-*c*]pyrazoles. Testosterone acetate **6** was oxidized with permanganate/periodate solution to obtain the seco-keto acid **7** [17]. The acetoxy function was hydrolysed under these reaction conditions. Compound **7** was subjected to the Leukart reaction resulting in the formation of a mixture of epimers **8**, whose NMR spectrum had a singlet at δ 0.76 for 18-methyl (3H) and two singlets for 19-methyl protons at δ 0.93 (α-isomer) and 1.02 (β-isomer) (3H) in a 60:40 ratio. Upon acetylation with an acetic anhydride/pyridine mixture at room temperature and crytallization, the mixture **8** resolved into the β-epimer **9** (mp 219–220 °C; 19-methyl singlet at δ 1.01) and the α-epimer **10** [14] (mp 260–262 °C; 19-methyl singlet at δ 0.93). The



Scheme 1.

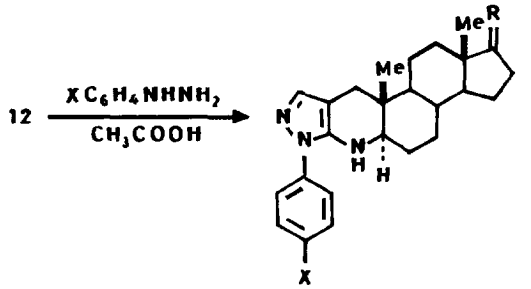


Scheme 2.

elemental composition of the two compounds was identical. The lactams **9** and **10** were treated separately with Vilsmeier–Haack reagent [16] to obtain 17β-acetoxy-3-chloro-4-aza-5β-androst-2-en-2-aldehyde **11** and 17β-acetoxy-3-chloro-4-aza-5α-androst-2-en-2-aldehyde **12** [18], respectively. Compounds **11** and **12** both showed a UV maximum at 306 nm.

Treatment of **12** with phenylhydrazine gave **13** (scheme 3), which hydrolysed to yield 2'-phenyl-4-aza-5α-androstano [3,2-*c*]pyrazol-17β-ol **14**. A multiplet appeared in the range of δ 7.45–7.80 (6H, aromatic protons and 5'-C*H*) in the NMR spectrum.

Similarly, on treatment with *p*-fluorophenylhydrazine hydrochloride product **12** gave **15** (scheme 3),



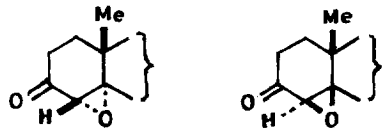
13 R = H,OCOCH3; X = H
14 R = H,OH; X = H
15 R = H,OCOCH3; X = F
16 R = H,OH; X = F
17 R = O; X = F

Scheme 3.

which on hydrolysis and Jones's oxidation [19] gave 17-oxo derivative **17**. An infrared band appeared at 1739 cm^{-1} indicating the presence of a 17-keto function. An 18-methyl singlet appeared at δ 1.06 in the NMR spectrum, which is slightly downfield due to the 17-oxo function.

Unsuccessful attempts were made to prepare pyrazole derivatives with 5β-3-chloro-2-aldehyde epimer **11**.

The synthesis of 17a-aza-D-homo steroidal pyrazoles was then carried out. 3β-Acetoxy-17-chloro-17a-aza-D-homo-5,16-androstadien-16-aldehyde **18** was prepared as reported previously [10]. Treatment of **18** with *p*-fluorophenylhydrazine hydrochloride gave the 2'-*p*-fluorophenyl derivative **19** (UV maxima at 244 and 274.4 nm; NMR signals at δ 1.10 (18-C*H*$_3$), 7.06–7.30 (m, 2H, aromatic protons), 7.40 (s, 1H, 5'-C*H*) and 7.78 (m, 2H; aromatic protons *ortho* to pyrazole ring) (scheme 4). Alkaline hydrolysis of **19** gave the 3β-hydroxy derivative **20**, which was subjected to Oppenauer oxidation [13] using the cyclohexanone/toluene system to get α,β-unsaturated ketone **21** (UV maximum at 242.4 nm; NMR singlet at δ 5.80 (4-C*H*). Epoxidation [20] of **21** with alkaline hydrogen peroxide in methanol gave a 30:70 mixture of 4α,5α-oxido **22A** and 4β,5β-oxido **22B** isomers as reported previously [21]. In the NMR spectrum, a singlet appeared at δ 3.03 for 4α-*H* (19-methyl singlet at δ 1.16) and another at δ 3.08 for 4β-*H* (19-methyl singlet at δ 1.08) in a 70:30 ratio; together these signals integrated for one proton.

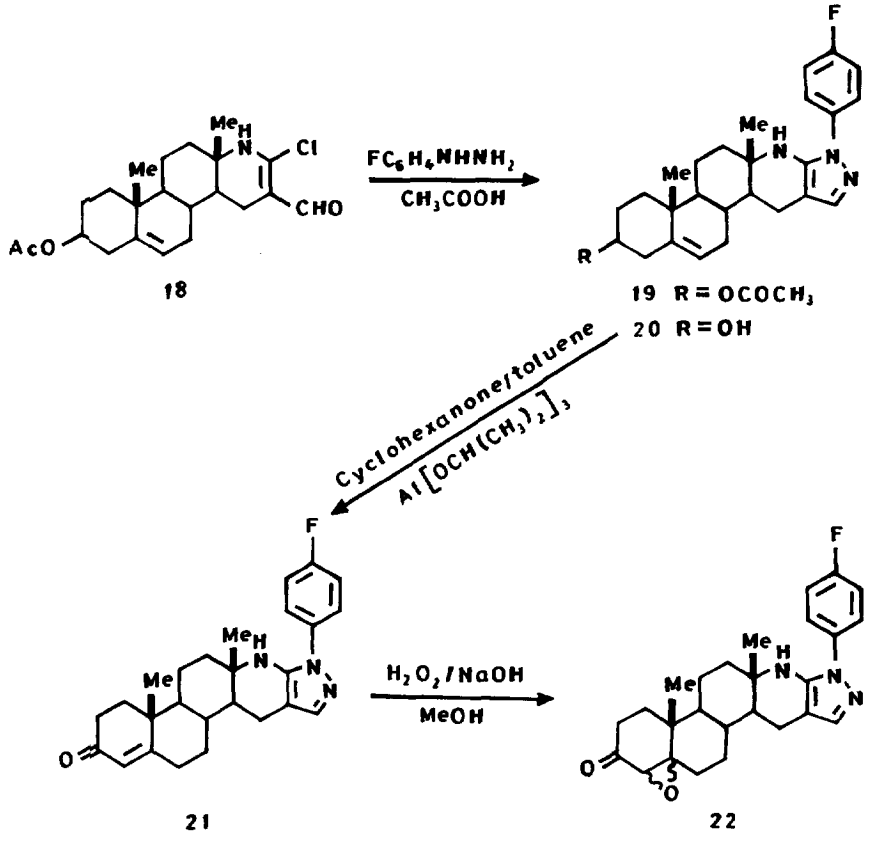


22 A 22 B

Biological activity

Antiinflammatory activity

The screening was carried out using the carrageenan rat paw oedema model [22]. The compounds were



Scheme 4.

given orally as suspensions. The extent of inhibition of oedema was related to the dose administered. For comparison, hydrocortisone was used as the standard. The results obtained are summarized in table I.

Compounds **4**, **10**, **12** and **18**, which do not have heterocycles fused to the azasteroid skeleton, were inactive in the dose range of 4–32 mg/kg. In cholestane series, steroidal pyrazole **5** did not show activity whereas 4-aza-androstano[3,2-*c*]pyrazoles **13** (ED_{50} = 11.00 mg/kg), **15** (ED_{50} = 7.60 mg/kg), **16** (ED_{50} = 2.93 mg/kg) showed good activity and were found more active than hydrocortisone (ED_{50} = 13.49 mg/kg). The acetoxy derivative **15** was found to be less active than the hydroxy derivative **16**.

In the case of the 17-aza series, the α,β-unsaturated ketone **21** (ED_{50} = 3.67 mg/kg) and oxirane **22** (ED_{50} = 1.41 mg/kg) showed good activity. Hydroxy derivative **20** (ED_{50} not determined) and acetoxy derivative **19** (ED_{50} not determined) were not found active at doses up to 16 mg/kg. 2'-*p*-Fluorophenyl-17a-aza-4ξ, 5-oxido-D-homo-5ξ-androstano[17,16-*c*]pyrazol-3-one **22** was found to be the most active compound in this study. In conclusion, this study shows that fusion of pyrazole ring systems at the 2,3- and 16,17-positions of azasteroids afforded compounds with enhanced antiinflammatory activity compared with standard drugs in the androstane series.

Antineoplastic activity

The compounds were also tested at National Cancer Institute, Bethesda, MD, USA, in vitro against a cell panel consisting of 60 lines. A 48 h continuous exposure protocol was used and a sulforhodamin B (SRB) protein assay was used to estimate cell viability or growth. A selectivity analysis of the compounds for differential cellular sensitivity based on the response parameters GI50, TGI and LC50 is shown in table II. It is suggested that a value of D_{GI50}, D_{TGI} or $D_{LC50} > 50$ is statistically significant. Figures within parentheses indicate the concentrations at which the maximum selective effect occurred. These values indicate that the compounds have statistically insignificant antineoplastic activity for further study.

Experimental protocols

Chemistry

Melting points reported are uncorrected. NMR spectra were recorded on Varian EM-390, 90 MHz and EM-360, 60 MHz for solutions in deuteriochloroform using tetramethylsilane (TMS) as the internal standard. IR and UV spectra were obtained using Perkin-Elmer 881 and Lambda-15 spectrophotometers, respectively. Mass spectra were recorded on a VG-11-250 J 70S. The purity of the compounds was examined by thin-layer chromatography. Elemental analysis was carried out on a Perkin-Elmer-2400. Ultraviolet spectra were recorded in methanol (λ_{max} in nm, figures within parentheses refer to log ε values) and IR spectra were obtained in potassium bromide pellets (ν_{max} in cm^{-1}).

*2'-Phenyl-5α-cholestano[3,2-c]pyrazole **5***

Glacial acetic acid (0.25 mL) was added to a refluxing solution of 3-chloro-4-aza-5α-cholest-2-en-2-aldehyde **4** (0.25 g) in aldehyde-free ethanol (100 mL). Phenylhdrazine (0.5 mL) was added to the refluxing solution and refluxed for 10 h. The reaction mixture was concentrated to 10 mL and poured into ice-cold water (200 mL). The precipitate obtained was filtered, washed and crystallized from acetone. Yield: 0.08 g (28.5%); mp: 174–177 °C; $[\alpha]_D^{20}$ +97.00° (*c* = 1.00 in $CHCl_3$); λ_{max}^{MeOH} 247 nm (log ε 4.15); IR (Br): 3260 (N-H stretch), 2930 (C-H stretch); NMR ($CDCl_3$): δ 0.70 (3H, s, 18-C*H*$_3$), 0.93 (3H, s, 19-C*H*$_3$), 2.87 (H, m, 5α-*H*), 3.56 (H, exchangeable, -N*H*), 7.23–7.76 (6H, m, 5 aromatic protons and 5'-C*H*); MS: *m/z* 487. Anal for $C_{33}H_{49}N_3$: C, 81.26; H, 10.15; N, 8.61. Found: C, 81.34; H, 10.58; N, 8.24.

*17β-Hydroxy-4-aza-5ξ-androstan-3-one **8***

A concentrated solution of ammonia was added dropwise to formic acid (3 mL, 98%) till the mixture became alkaline. A suspension of 17β-hydroxy-5-oxo-3,5-seco-4-norandrostan-3-oic acid **7** (2.0 g) in nitrobenzene (10 mL) was added to an ammonium formate solution which had been brought to 165 °C in an oil bath. The reaction mixture was stirred for 20 h at 180–200 °C, cooled, and washed gently with water. Ethanol (20 mL) and concentrated hydrochloric acid (5 mL) were added and the mixture was refluxed for 2 h. It was steam distilled to remove nitrobenzene. The residue was extracted with chloroform (5 x 30 mL). The combined organic layer was washed with 2% sodium carbonate solution, water, and dried. The solvent was removed to leave an amorphous residue, which was crystallized from methanol to obtain a mixture of epimers **8**. Yield: 1.0 g (53.0%); mp: 278–280 °C; IR (KBr): 3381 (-OH), 3175 (NH), 1660 (amide); NMR ($CDCl_3$): δ 0.76 (3H, s, 18-C*H*$_3$), 0.93 (s) and 1.03 (s) (60:40, 3H, 19-C*H*$_3$; α,β-epimers), 3.10 (m) and 3.30 (s) (H, 5α-*H* and 5β-*H*), 3.70 (H, m, 17α-*H*), 6.26 (br, H, exchangeable, -N*H*).

*3-Oxo-4-aza-5α/5β-androstan-17β-yl acetates **9** and **10***

Acetic anhydride (5 mL) was added to a solution of 17β-hydroxy-4-aza-5-androstan-3-one **8** (0.5 g) in pyridine (10 mL) with continuous stirring. The reaction mixture was stirred at room temperature for 20 h. Solvents were removed under reduced pressure and ice-cold water was added to the residue. The precipitated material was filtered, dried and crytallized from acetone to afford the 5α-epimer **10**. Yield: 0.25 g (43.5%); mp: 260–262 °C; $[\alpha]_D^{20}$ +74.0° (*c* = 0.50 in $CHCl_3$); IR (KBr): 3189 (NH), 1734 (ester), 1664 (amide); NMR ($CDCl_3$): δ 0.83 (3H, s, 18-C*H*$_3$), 0.93 (3H, s, 19-C*H*$_3$), 2.03 (3H, s, -OCOC*H*$_3$), 3.01 (H, m, 5α-*H*), 4.60 (H, t, 17α-*H*), 6.90 (H, s, exchangeable, -N*H*). Anal for $C_{20}H_{31}NO_3$: C, 72.03; H, 9.37; N, 4 20. Found: C, 71.88; H, 9.14; N, 4.12.

Concentration of the mother liquor gave a product which on recrystallization yielded 5β-epimer **9**. Yield: 0.2 g (35%); mp 219–220 °C; $[\alpha]_D^{20}$ +45.16° (*c* = 0.043 in $CHCl_3$); IR (KBr): 3269 (-NH), 1733 (ester), 1667 (amide); NMR ($CDCl_3$): δ 0.80 (3H, s, 18-C*H*$_3$), 1.00 (3H, s, 19-C*H*$_3$), 2.03 (3H, s, -OCOC*H*$_3$), 3.30 (H, m, 5β-*H*), 4.60 (H, t, 17α-*H*), 6.00 (H, s, exchangeable, -N*H*). Anal for $C_{20}H_{31}NO_3$: C, 72.03; H, 9.37; N, 4.20. Found: C, 71.60; H, 9.12; N,4.07.

Table I. Antiinflammatory activity of various compounds and hydrocortisone on carrageenan-induced paw oedema in rats (observed 180 min after the administration of carrageenan).

Compound	*Dose (mg/kg)*	*Number of rats*	*Inhibition of oedema (%) ± SEM*	*ED_{50} (mg/kg)*
12	8.00	6	9.28 ± 1.28	
	16.00	6	12.36 ± 0.96	ND
	32.00	6	16.00 ± 7.31	
10	8.00	6	1.36 ± 0.18	
	16.00	6	6.64 ± 1.12	ND
	32.00	6	9.25 ± 1.32	
4	4.00	6	5.36 ± 1.44	
	8.00	6	6.50 ± 1.20	ND
	16.00	6	12.00 ± 1.44	
18	8.00	6	5.36 ± 1.68	
	16.00	6	8.00 ± 1.44	ND
	32.00	6	11.24 ± 1.78	
5	2.00	6	9.28 ± 1.28	
	4.00	6	12.36 ± 0.96	ND
	8.00	6	16.00 ± 7.39	
	16.00	6	21.51 ± 2.12	
13	2.00	6	8.20 ± 1.44*	
	4.00	6	21.40 ± 1.47*	
	8.00	6	42.00 ± 1.92*	11.00
	16.00	6	55.03 ± 2.02*	
15	2.00	6	10.12 ± 1.32*	
	4.00	6	30.22 ± 1.40*	7.60
	8.00	6	52.01 ± 5.40*	
	16.00	6	62.00 ± 0.68*	
16	1.00	6	8.38 ± 4.06*	
	2.00	6	43.20 ± 0.68*	
	4.00	6	64.19 ± 5.80*	2.93
	8.00	6	79.43 ± 2.74*	
19	2.00	6	15.60 ± 1.20	
	4.00	6	22.53 ± 3.60	
	8.00	6	25.57 ± 2.20	ND
	16.00	6	24.42 ± 1.03	
20	2.00	6	4.32 ± 3.20	
	4.00	6	5.40 ± 1.90	
	8.00	6	13.54 ± 2.60	ND
	16.00	6	20.62 ± 1.32	
21	1.00	6	15.46 ± 2.40*	
	2.00	6	25.29 ± 2.99*	
	4.00	6	57.97 ± 2.42*	3.67
	8.00	6	71.16 ± 3.78*	
22	0.50	6	19.36 ± 2.99*	
	1.00	6	42.82 ± 3.50*	
	2.00	6	66.29 ± 1.00*	1.41
	4.00	6	72.03 ± 2.13*	
Hydrocortisone	4.00	6	18.80 ± 1.65*	
	8.00	6	43.07 ± 4.75*	
	16.00	6	59.24 ± 5.84*	13.49
	32.00	6	63.60 ± 2.96*	

Significance relative to control group data: *$P < 0.01$; ND: not determinable.

Table II.

Compound	*GI50*	*TGI*	*LC50*
14	43.0 (– 5.0)	29.0 (– 5.0)	44.0 (– 4.0)
18	62.0 (– 5.0)	43.0 (– 4.0)	23.0 (– 4.01)
19	58.0 (– 5.0)	48.0 (– 4.0)	31.0 (– 4.0)

Values in parentheses are the concentrations at which the maximum selective effect occurred.

2'-Phenyl-4-aza-5α-androstano[3,2-c]pyrazol-17β-yl acetate ***13***
To a refluxing solution of 17β-acetoxy-3-chloro-4-aza-5α-androst-2-en-2-aldehyde **12** (0.2 g) in aldehyde-free ethanol (100 mL), glacial acetic acid (0.5 mL) was added dropwise. The solution was refluxed for 10 min, phenylhydrazine (0.5 mL) was added and the mixture was refluxed for 5 h. The reaction mixture was concentrated to about 20 mL and then poured into ice-cold water. The precipitated product was filtered, washed, dried and crystallized from acetone/hexane. Yield: 0.16 g (70.1%); mp: 205–206 °C; λ_{max}^{MeOH} 246 nm (log ε 4.01); IR (KBr): 3335, 2920, 1730 (ester), 1600; NMR ($CDCl_3$): δ 0.80 (6H, s, 18-C*H*$_3$ and 19-C*H*$_3$), 2.03 (3H, s, -OCOC*H*$_3$), 3.53 (H, s, -N*H*, exchangeable), 4.60 (H, m, 17α-*H*), 7.28–7.80 (6H, m, 5 aromatic protons and 5'-C*H*); MS: *m/z* 433. Anal for $C_{27}H_{35}N_3O_2$: C, 74.79; H, 8.13; N, 9.69. Found: C, 74.57; H, 8.02; N, 9.46.

2'-Phenyl-4-aza-5α-androtano[3,2-c]pyrazol-17β-ol ***14***
A mixture of 2'-phenyl-4-aza-5α-androstano[3,2-*c*]pyrazol-17β-yl acetate **13** (0.2 g) and potassium carbonate (0.7 g) in 10% aqueous methanol (20 mL) was stirred at room temperature for 3 h. The slurry obtained was poured into ice-cold water. The precipitated product was filtered, washed and dried. Yield: 0.125 g (69.19%); mp: 198–200 °C; λ_{max}^{MeOH} 242.8 nm (log ε 3.97); IR (KBr): 3399 (OH), 1601, 1531; NMR ($CDCl_3$): δ 0.80 (3H, s, 18-C*H*$_3$), 0.83 (3H, s, 19-C*H*$_3$), 3.03 (H, m, 5α-*H*), 3.70 (H, m, 17α-*H*), 6.60 (2H, br, -N*H* and -O*H*, exchangeable), 7.45–7.80 (6H, m, 5 aromatic protons and 5'-CH). Anal for $C_{25}H_{33}N_3O$: C, 76.68 ; H, 8.49; N, 10.73. Found: C, 76.28; H, 8.63; N, 10.50.

2'-p-Fluorophenyl-4-aza-5α-androstano[3,2-c]pyrazol-17β-yl acetate ***15***
To a refluxing solution of 17β-acetoxy-3-chloro-4-aza-5α-androst-2-en-2-aldehyde **12** (0.5 g) in aldehyde-free ethanol (250 mL), glacial acetic acid (1.5 mL) was added dropwise. The solution was refluxed for 10 min and then *p*-fluorophenylhydrazine hydrochloride (0.25 g) was added and the mixture was refluxed for 5 h. The solution was concentrated to about 20 mL and poured into ice-cold water. The precipitated product was filtered, washed, dried and crystallized from acetone. Yield: 0.25 g (41.9%); mp: 238–240 °C; λ_{max}^{MeOH} 244.4 nm (log ε 4.13); IR (KBr): 3289, 1715 (ester), 1591; NMR ($CDCl_3$): δ 0.86 (6H, s, 18-C*H*$_3$ and 19-C*H*$_3$), 2.06 (3H, s, -OCOC*H*$_3$), 2.90 (H, m, 5α-H,) 3.46 (H, br, -N*H*, exchangeable), 4.66 (H, t, 17α-*H*), 7.06–7.30 (2H, m, aromatic protons), 7.40 (H, s, 5'-C*H*), 7.56–7.80 (2H, m, aromatic protons *ortho* to pyrazole ring); MS: *m/z* 451. Anal for $C_{27}H_{34}N_3O_2F$: C,71.80; H, 7.59; N, 9.30. Found: C, 71.59; H, 7.95; N, 9.67.

2'-p-Fluorophenyl-4-aza-5α-androstano[3,2-c]pyrazol-17β-ol ***16***
A mixture of 2'-*p*-fluorophenyl-4-aza-5α-androstano[3,2-*c*]-pyrazol-17β-yl acetate **15** (0.2 g) and potassium carbonate (0.5 g) in 10% aqueous methanol (50 mL) was stirred at room temperature for 3 h. The slurry obtained was poured into ice-cold water. The precipitated product was filtered, washed, dried and crystallized from acetone. Yield: 0.125 g (68.9%); mp: 240–242 °C; λ_{max}^{MeOH} 224.2 nm (log ε 3.95); IR (KBr): 3440 (-OH), 3251 (-NH), 2932, 1591; NMR ($CDCl_3$): δ 0.76 (3H, s, 18-C*H*$_3$), 0.80 (3H, s, 19-C*H*$_3$), 3.63 (H, t, 17α-*H*), 7.03–7.43 (3H, m, aromatic protons and 5'-C*H*), 7.60 (2H, m, aromatic protons *ortho* to pyrazole ring); MS: *m/z* 409. Anal for $C_{25}H_{32}N_3OF$: C, 73.30; H, 7.88; N, 10.20. Found: C, 72.68; H, 8.13; N, 10.46.

2'-p-Fluorophenyl-4-aza-5α-androstano[3,2-c]pyrazol-17-one ***17***
To a solution of 2'-*p*-fluorophenyl-4-aza-5α-androstano[3,2-*c*]-pyrazol-17β-ol **16** (0.15 g) in aldehyde-free acetone (15 mL) was added Jones's reagent [14] (prepared by dissolving chromium trioxide (2.5 g) in water (7 mL) and then adding concentrated sulphuric acid (2.5 mL)) dropwise with stirring and maintaining the temperature around 15 °C till the colour of Jones's reagent persisted. The reaction mixture was diluted with water and extracted with chloroform. The chloroform extract was washed with water, dried and the solvent removed. The product obtained was crystallized from acetone/hexane. Yield: 0.08 g (53%); mp 188–189 °C; λ_{max}^{MeOH} 226.8 nm (log ε 4.05), 275 nm (log ε 3.88); IR (KBr): 2938, 1739 (C=O), 1586; NMR ($CDCl_3$): δ 0.91 (3H, s, 19-C*H*$_3$), 1.06 (3H, s, 18-C*H*$_3$), 7.05– 7.11 (2H, m, aromatic protons), 7.19 (H, s, N*H*, exchangeable), 7.39 (H, m, 5'-C*H*), 7.63–7.84 (2H, m, aromatic protons *ortho* to pyrazole ring). MS: *m/z* 407. Anal for $C_{25}H_{30}N_3OF{\cdot}H_2O$: C, 70.56; H, 7.57; N, 9.88. Found: C, 70.20; H, 7.15; N, 9.43.

2'-p-Fluorophenyl-17a-aza-D-homo-5-androsteno[17,16-c]-pyrazol-3β-yl acetate ***19***
To a refluxing solution of 3β-acetoxy-17-chloro-17a-aza-D-homo-5,16-androstadien-16-aldehyde **18** (0.5 g) in aldehyde-free ethanol (50 mL) was added glacial acetic acid (2 mL). The solution was refluxed for 10 min and *p*-fluorophenylhydrazine hydrochloride (0.3 g) was added. The solution was further refluxed for 6 h. The reaction mixture was concentrated and poured into ice-cold water. The precipitated product was filtered, washed, dried and crystallized from methanol. Yield: 0.48 (63.49%); mp: 244–246 °C; λ_{max}^{MeOH} 244 nm (log ε 4.08), 274.4 nm (log ε 3.63); IR (KBr): 2943, 1720 (ester), 1591, 1511; NMR ($CDCl_3$): δ 1.06 (3H, s, 18-C*H*$_3$), 1.10 (3H, s, 19-C*H*$_3$), 2.06 (3H, s, -OCOC*H*$_3$), 4.63 (H, m, 3α-*H*), 5.47 (H, m, 6-C*H*), 7.06–7.30 (2H, m, aromatic protons), 7.40 (H, s, 5'-C*H*), 7.53–7.78 (2H, m, aromatic protons *ortho* to pyrazole ring). Anal for $C_{28}H_{34}N_3O_2F$: C, 72.55; H, 7.38; N, 9.06 Found: C, 72.11; H, 7.28; N, 8.92.

2'-p-Fluorophenyl-17a-aza-D-homo-5-androsteno[17,16-c]-pyrazol-3β-ol ***20***
A solution of 2'-*p*-fluorophenyl-17a-aza-D-homo-5-androsteno-[17,16-*c*]pyrazol-3β-yl acetate (0.5 g) **19** in methanol (50 mL) containing potassium hydroxide (0.2 g) was refluxed for 45 min. The reaction mixture was concentrated, acidified with glacial acetic acid and poured into ice-cold water. The precipitated product was filtered, washed, dried and crytallized from methanol. Yield: 0.45 g (99.11%); mp: 266–268 °C; λ_{max}^{MeOH} 241.2 nm (log ε 3.94); IR (KBr): 3439 (OH), 3286, 1591, 1535; NMR ($CDCl_3$/DMSO-d_6): δ 1.00 (6H, s, 18-C*H*$_3$ and 19-C*H*$_3$),

3.00 (H, m, -OH, exchangeable), 3.40 (H, m, 3α-*H*), 4.10 (H, m, -NH, exchangeable), 5.40 (H, m, 6-C*H*), 7.13–7.33 (2H, m, aromatic protons), 7.40 (H, s, 5'-C*H*), 7.63–7.93 (2H, m, aromatic protons *ortho* to pyrazole ring). Anal for $C_{26}H_{32}N_3O$ F: C, 74.08; H, 7.64; N, 9.96. Found: C, 73.93; H, 7.61; N, 9.95.

2'-p-Fluorophenyl-17a-aza-D-homo-4-androsteno[17,16-c]-pyrazol-3-one ***21***

A solution of 2'-*p*-fluorophenyl-17a-aza-D-homo-5-androsteno-[17,16-*c*]pyrazol-3β-ol **20** (0.5 g) in cyclohexanone (5 mL) and toluene (100 mL) was slowly distilled while aluminium isopropoxide (1.0 g) in toluene (10 mL) was added to remove moisture. The distillation was continued for 30 min. The reaction mixture was refluxed for 4 h and allowed to stand overnight. The solution was filtered, the filtrate was steam distilled and the residue obtained was crytallized from ethyl acetate. Yield: 0.35 g (70.42%); mp: 226–228 °C; λ_{max}^{MeOH} 241.4 nm (log ε 4.39); IR (KBr): 3269, 2940, 1672, 1612, 1509; NMR ($CDCl_3$): δ 1.13 (3H, s, 18-CH_3), 1.23 (3H, s, 19-CH_3), 3.63 (H, br, -N*H*, exchangeable), 5.80 (H, s, 4-C*H*), 7.10–7.36 (2H, t, aromatic protons), 7.40 (H, s, 5'-C*H*), 7.56–7.83 (2H, m, aromatic protons *ortho* to pyrazole ring). Anal for $C_{26}H_{30}N_3OF$: C, 74.43; H, 7.21; N, 10.02. Found C, 74.25; H, 7.34; N, 10.09.

2'-p-Fluorophenyl-17a-aza-4ξ,5-oxido-D-homo-5ξ-androstano[17,16-c]pyrazol-3-one ***22***

To a stirred solution of 2'-*p*-fluorophenyl-17a-aza-D-homo-4-androsteno[17,16-*c*]pyrazol-3-one (0.2 g) **21** in methanol (20 mL), hydrogen peroxide (30% v/v, 3 mL) and aqueous sodium hydroxide (4 N, 1 mL) were added simultaneously, maintaining the temperature at 0 °C. The reaction mixture was kept for 24 h at 0–5 °C, diluted with water and extracted with ethyl acetate. The combined extract was washed with water, dried and the solvent was removed. Upon crystallization from ethanol, the product gave oxirane **20**. Yield: 0.14 g (67.42%), mp: 205–207 °C; λ_{max}^{MeOH} 243.2 nm (log ε 4.09); IR (KBr): 3314, 2944, 1707, 1534, 1512; NMR ($CDCl_3$): δ 1.07 (3H, s, 18-CH_3), 1.08 (s) and 1.16 (s) (3H, 19-CH_3, 30:70, α and β-isomers), 3.03 (s) and 3.08 (s) (70:30, 1H, 4-C*H*), 3.40 (H, s, -NH, exchangeable), 7.10–7.33 (2H, m, aromatic protons), 7.40 (H, s, 5'-C*H*), 7.56–7.80 (2H, m, aromatic protons *ortho* to pyrazole ring). Anal for $C_{26}H_{30}N_3O_2F$: C, 71.69; H, 6.94; N, 9.65. Found: C, 71.31; H, 7.18; N, 9.36.

Pharmacology

The antiinflammatory study was conducted on male albino rats (Porton strain) weighing 100–150 g against carrageenan-induced rat pedal oedema [22]. The rats were randomly divided into groups of a minimum of six animals. Suspensions of different doses of the relative compounds, uniformly dispersed in distilled water by adding 0.1 mL of Tween 80 were given to test animals orally 1 h prior to the administration of carrageenan. The control group received the same experimental handling as the test groups except that equivalent doses of vehicle alone were administered by the same route in place of the test compounds. The rats were fasted 20 h before the start of the study. Hydrocortisone was used as the standard antiinflammatory drug.

Carrageenan-induced oedema

Acute oedema in the hind paws of the rats was induced by injecting 0.1 mL of freshly prepared 1% solution of carrageenan (type IV, Sigma) in distilled water under the plantar aponeurosis of the right hind paw. The paw volumes were measured immediately using plethysmometer 7150, and again 30, 60, 120 and 180 min after the injection of carrageenan.

The percent inhibition of inflammation after 30, 60, 120 and 180 min was calculated after the method of Newbould [23] using the following formula: percent inhibition = $100\ (1 - (a - x)/(b - y))$, where x and a are the mean foot volumes of the rats, before and after the administration of carrageenan injection, respectively, treated with test compounds or standard drug; y and b are the mean foot volumes of the rats before and after the administration of carrageenan, respectively, in the control group.

Acknowledgments

We are grateful to the University Grant Commission, New Delhi and Panjab University for providing financial support. We thank VL Narayanan, NCI, USA for screening the compounds for antineoplastic activity.

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