17β-[4-(1,3-THIAZOLYL)]-ANDROST-5-ENE DERIVATIVES*

Pavel DRAŠAR[a], František TUREČEK[b] and Miroslav HAVEL[a]

[a] *Institute of Organic Chemistry and Biochemistry, Czechoslovak Academy of Sciences, 166 10 Prague 6 and*
[b] *The J. Heyrovský Institute of Physical Chemistry and Electrochemistry, Czechoslovak Academy of Sciences, 121 38 Prague 2*

Received March 16th, 1981

17β-[4-(1,3-Thiazolyl)]-androst-5-ene with a formamide and ethoxycarbonyl group in position 2′ of the 1,3-thiazole ring were synthesized and their structures studied by means of hydrogenolytic degradation and spectroscopic methods.

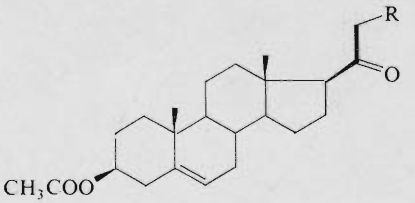
The literature reports a series of steroidal 1,3-thiazoles bearing the heterocyclic moiety in position 17β. Examples of such compounds are 2′-hydroxy-4′-thiazolyl[1], 2′-amino-4′-thiazolyl[1-3] and N-substituted 2′-amino-4′-thiazolyl[3-7] derivatives or (20*R*)-(2,4-disubstituted 5-thiazolyl)-pregnane[8] and 23-(2-amino-4-thiazolyl)--24-norcholane[9]. Contrasting with the formation of the above derivatives is a report[10] on a synthesis of 17β-[2-formamido-5-(1,3-thiazolyl)]-3β-hydroxyandrost-5-en (in original communication 2′-formamidothiazolo[5′,4′:20,21]pregna-5,20-dien-3β-ol) the formation of which would require unusual course of the Hantzsch reaction[11].

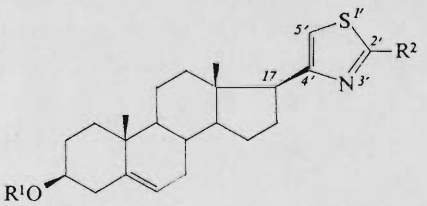
Derivatives of the above series were shown[1,3-7] to possess cardiotonic activity that is accompanied by lower toxicity than is that of the cardiotonic steroid lactones used as standards[4,5]. It appeared therefore of interest to compare them with the derivatives of the 5′-thiazolylandrostane type[10]. For all these reasons we decided to re-investigate this application of the Hantzsch reaction in some detail, particularly with respect to the attachment of the thiazole ring to the steroid skeleton. To this purpose, we have chosen two ways, both starting from the bromo ketone *III*. The first route was the reaction of *III* with thiourea in dimethylformamide which was also applied in the above cited paper[10]. An alternative approach, leading however to a thiazole ring substituted at position 2 in a different manner, was reaction of the compound *III* with ethyl thioxamate; this route parallels the synthesis of the thiazole analog ribavirin[12,13].

The starting ketone *III* was prepared from 21-methanesulfonyloxy derivative[14] *II* by substitution of the methanesulfonyl group by bromine. Although successful application of the tosylates of the type *II* to direct condensation with thiourea had

* Part CCLII in the series On Steroids; Part CCLI: This Journal *46*, 2758 (1981).

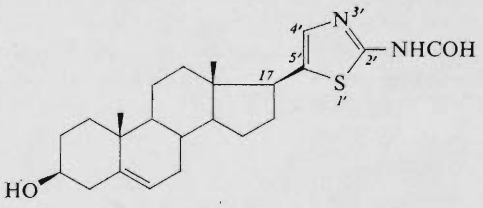
been reported[1,2], we only obtained an untractable reaction mixture from *II*. The substitution of the methanesulfonyl group in the derivative *II* was carried out using tetrabutylammonium bromide in dimethylformamide or lithium bromide in acetone. From preparative point of view, the second method is preferable due to a more easy workup of the reaction mixture. It is also noteworthy that prolonged heating of the compound *II* with tetrabutylammonium bromide in dimethylformamide led to formation of some 3β-acetoxypregna-5,16-dien-20-one. The ketone *III* was condensed with ethyl thioxamate in acetonitrile to furnish the thiazole *V* in 62% yield or with thiourea in dimethylformamide to give *IV* in 83% yield. The derivative *IV* was deacetylated by alkaline hydrolysis to provide the compound *VI* which was compared with a compound described earlier[10] and considered to possess the structure *VIII*. Both the melting point and mass spectrum agree with the data published[10]. This finding and the mode of formation prove identity of both products. It thus appeared necessary to establish whether the product reported in the present paper bears a 4′ or 5′(1,3-thiazolyl) substituent. We have chosen reductive cleavage of the thiazole ring despite its known chemical resistance generally[15] and towards Raney nickel reduction specifically[16].





I, R = —H
II, R = —OSO_2CH_3
III, R = —Br

IV, R^1 = —$COCH_3$; R^2 = —NHCOH
V, R^1 = —$COCH_3$; R^2 = —$COOC_2H_5$
VI, R^1 = —H; R^2 = —NHCOH
VII, R^1 = —$COCH_3$; R^2 = —NH_2



VIII

Both thiazoles *IV* and *V* were cleaved by desulfuration with Raney nickel in boiling ethanol. 3β-Acetoxypregn-5-en-20-one (*I*) was isolated from the thiazole *IV* in 30% yield. Formation of the steroid *I* was also demonstrated after desulfuration cleavage with Raney nickel of the thiazole *V*. We assume that it arises by desulfuration cleavage

of the thiazole ring followed by hydrolysis. Replacing the Raney nickel by P-2 nickel[17] leads to a considerably lower yield of thiazole cleavage (about 3%). The above experiments make it clear that the thiazole ring is attached to the steroid molecule by 17,4'-bond, *i.e.* the sulfur atom is adjacent to $C_{(21)}$-atom of the pregnane skeleton.

The above presented chemical proof was further supported by a study of ^{13}C-NMR spectra of both thiazoles *IV* and *V* whereas ^{1}H-NMR, mass or IR spectra gave no unequivocal answer regarding steroid-thiazole attachment. ^{13}C-chemical shifts of the carbon atoms in 1,3-thiazole are known from the literature[18]: $C_{(2)}$ δ = 152·2, $C_{(4)}$ δ = 142·45, $C_{(5)}$ δ = 118·5. Substitution on the thiazole ring leads to a downfield shift of the signal associated with the carbon bearing the substituent[18] (Table I). A comparison of the values in the Table I leads to the conclusion that compounds *IV* and *V* possess a thiazole ring that is attached to the steroid skeleton by the 17,4'--bond. This finding was confirmed by determining the tertiary and quaternary carbon atoms of the thiazole ring in *IV* and *V* (Table I).

It can be concluded that the modified Hantzsch reaction[11] of the halo ketones (type *III*) with substituted thioamides, as *e.g.* ethyl thioxamate or thiourea, gives rise solely to substituted 1,3-thiazoles (type *VI*) bearing the rest of the halo ketone in the position 4 of the thiazole ring. Formation of thiazoles substituted in position 5 (type *VIII*), claimed in the cited work[10], could not be confirmed.

EXPERIMENTAL

Melting points were determined on a Boetius block. Optical rotations were measured on a polarimeter Opton, type VDRNA. The infrared spectra were recorded on a Zeiss UR-20 instrument, the NMR spectra on instruments indicated below, using tetramethylsilane as internal reference. Chemical shifts are given in ppm. (δ-scale), coupling constants in Hz. The number of hydrogen atoms, attached to the carbons of the thiazole ring, was determined by off-resonance multiplicities (compound *IV*) or by changes of signal intensities due to different weighting factors in Fourier transformation of the free induction decay signal (compound *V*). The mass spectra were recorded on a AEI MS 901 instrument. Analytical samples were dried at 50°C/25 Pa. Column chromatographies were carried out on silica gel with size of particles 60—120 μm, prepared according to Pitra in the Service laboratories of this Institute. Silica gel G (according to Stahl, Woelm) was used for thin layer chromatography. Depending on the solvent used solutions were evaporated on a vacuum rotary evaporator at 30—50°C/2·5 kPa. Solutions were dried by anhydrous magnesium sulfate.

3β-Acetoxy-21-bromopregn-5-en-20-one (*III*)

a) A mixture of the mesylate *II* (ref.[14], 453 mg, 1 mmol) and tetrabutylammonium bromide (1·6 g, 5 mmol) in dimethylformamide (15 ml) was stirred at 25° for 2 h. The mixture was poured on ice, the precipitate filtered, washed with water, dried, dissolved in benzene, filtered through a column of aluminum oxide (150 ml) and the column eluted with benzene. The solvent was evaporated and the residue crystallized from benzene–acetone to yield the bromo ketone *III* (370 mg, 84·6%) with physical constants identical with those reported in the literature[19]:

b) A mixture of the mesylate *II* (ref.[14], 325 mg, 0·72 mmol), lithium bromide (634 mg) and acetone was heated with stirring at 50°C for 1 h, passed through a column of aluminum oxide (150 ml) and worked up as in *a*) to yield the bromo ketone *III* (289 mg, 92%) of the same properties as above.

3β-Acetoxy-17β-[2-formamido-4-(1,3-thiazolyl)]androst-5-ene (*IV*)

The bromo ketone *III* (300 mg, 0·68 mmol) and thiourea (114 mg, 1·5 mmol) in dimethylformamide (10 ml) were heated with stirring at 150°C for 2 h. The mixture was poured on ice, the precipitate washed with water and dried. The material was dissolved in benzene, dried and the solvent evaporated to give the formamido derivative *IV* as a foam (250 mg, 83·1%); $[\alpha]_D^{25}$ —120° (*c* 1·6, chloroform). IR spectrum (chloroform): 1 726, 1 258, 1 036 cm^{-1} (CH_3COO), 3 410, 3 185, 1 704, 1 693, 1 570, 1 553 cm^{-1} (RNHCHO), 1 635 cm^{-1} (C=N). ^{1}H-NMR spectrum (Varian HA-100, 100 MHz, deuteriochloroform): 8·54 br s (1 H, CHO), 6·57 s (1 H, $C_{(5')}$—H), 5·35 m (1 H, $C_{(6)}$—H), 4·56 m ($C_{(3)}$—H), 2·00 s (3 H, CH_3COO), 1·14 s (3 H, CH_3), 0·48 s (3 H, CH_3). ^{13}C-NMR spectrum (Jeol FX-60, 15·036 MHz, FT-mode, 25°C, deuteriochloroform): 152·02 d (NHCHO), 170·49 s ($OCOCH_3$), 158·02 s ($C_{(2')}$), 155·95 s ($C_{(4')}$), 139·69 s ($C_{(5')}$), 122·41 d ($C_{(6)}$), 108·25 d ($C_{(5')}$), 73·94 ($C_{(3)}$), 56·10, 52·76, 50·03, 44·05, 38·87 (2 C), 36·90, 36·64, 32·23, 31·84, 27·68, 26·38, 24·43, 21·31 (carbons of the steroid skeleton), 20·74 (CH_3COO), 19·23, 12·86 (angular methyls). Mass spectrum (*m/z*): 442 ($M^{+\cdot}$), 427 $(M-CH_3)^+$, 414 $(M-CO)^{+\cdot}$, 399 $(M-CH_3CO)^+$, 382 $(M-CH_3COOH)^{+\cdot}$, 367 (382—CH_3; ($C_{22}H_{27}N_2OS$)), 155 ($C_6H_7N_2OS$), 128 (formamidothiazole; $C_4H_4N_2OS$), 127 (128 — H). For $C_{25}H_{34}N_2O_3S$ (442·6) calculated: 67·84% C, 7·74% H, 6·33% N, 7·24% S; found: 67·74% C, 7·54% H, 6·14% N, 7·13% S.

3β-Acetoxy-17β-[2-ethoxycarbonyl-4-(1,3-thiazolyl)]-androst-5-ene (*V*)

A mixture of the bromo ketone *III* (300 mg, 0·68 mmol), ethyl thioxamate (90 mg, 0·68 mmol) and acetonitrile (10 ml) was refluxed for 24 h, diluted with ice water and the product taken up in benzene, the extract treated with charcoal, dried and filtered. Evaporation gave a residue which was crystallized from benzene–light petroleum to yield the thiazole *V* (200 mg, 62·5%), m.p. 168—172°C; $[\alpha]_D^{25}$ —84° (*c* 1·0, chloroform). IR spectrum (chloroform): 1 728, 1 258, 1 033 cm^{-1} (CH_3COO), 1 504 cm^{-1} (thiazole), 1 714, 1 305, 1 096 cm^{-1} (ethoxycarbonylthiazole). ^{1}H-NMR spectrum (Tesla B 476, 60 MHz, deuteriochloroform): 7·18 s (1 H, $C_{(5')}$—H), 5·37 m (1 H,

TABLE I

Comparison of ^{13}C-chemical shifts (ppm, δ scale) of substituted and unsubstituted carbon atoms in 1,3-thiazole and thiazoles *IV* and *V*

Carbon	Substituted[18] 1,3-thiazole	Unsubstituted[18] 1,3-thiazole	*IV*[a]	*V*
$C_{(2)}$	158—164	148—152	158·02 s	160·85[b]
$C_{(4)}$	149—165	141—142	155·95 s	156·70[b]
$C_{(5)}$	122—125	108—118	108·25 d	119·44[c]

[a] s singlet, d doublet; [b] quaternary carbon atom; [c] tertiary carbon atom.

$C_{(6)}$—H), 4·55 m (1 H, $C_{(3)}$—H), 4·42 q (2 H, —$COCH_2CH_3$, J_{HH} = 7·5), 1·99 s (3 H, CH_3.COO), 1·40t (3 H, —CH_2CH_3, J_{HH} = 7·5), 1·00 s (3 H, CH_3), 0·48 s (3 H, CH_3). ^{13}C-NMR spectrum (Varian XL-200, 50·3 MHz, FT-.mode, 23°, deuteriochloroform): 170·54 ($OCOCH_3$), 165·80 ($COOC_2H_5$), 160·85 ($C_{(2')}$), 156·70 ($C_{(4')}$), 139·83 ($C_{(5)}$), 122·42 ($C_{(6)}$), 119·44 ($C_{(5')}$), 73·94 ($C_{(3)}$), 62·34 ($COOCH_2CH_3$), 56·09, 52·76, 50·18, 44·28, 38·14, 37·77, 37·04, 36·74, 32·30, 31·90, 27·77, 26·75, 24·64, 21·43 (carbons of the steroid skeleton), 20·74 (CH_3COO), 19·35 (angular methyl), 14·29 (CH_3CH_2O), 13·02 (angular methyl). Mass spectrum (*m/z*): 471 ($M^{+\cdot}$), 411 ($M—CH_3COOH)^{+\cdot}$, 396 (411 — $CH_3)^+$, 382 (411 — $C_2H_5)^+$, 338 (411 — $COOC_2H_5)^+$. For $C_{27}H_{37}NO_4S$ (471·7) calculated: 68·76% C, 7·91% H, 2·97% N, 6·80% S; found: 68·63% C, 7·70% H, 2·85% N, 6·93% S.

17β-[2-Formamido-4-(1,3-thiazolyl)]-3β-hydroxyandrost-5-ene (*VI*)

A mixture of the thiazole *IV* (50 mg, 0·11 mmol), potassium carbonate (100 mg), water (one drop) and methanol (5 ml) was heated at 80°C (bath) for 15 min, diluted with chloroform (100 ml) and the solvent evaporated. Water and ether were added to the residue and the mixture shaken, the ether layer dried and evaporated. Crystallization of the residue from chloroform–methanol gave the hydroxy derivative *VI* (35 mg, 77%), m.p. 229—231°C, mass spectrum identical with the published data[10]. IR-spectrum (KBr): 1 700, 1 685, 1 675, 1 542, 1 533 cm^{-1} (CONH), 1 055 cm^{-1} (OH), 1 619 cm^{-1} (thiazole).

Reduction of the Thiazole *IV* on Raney Nickel

The thiazole *IV* (100 mg, 0·23 mmol) was refluxed with excess Raney nickel in ethanol for 40 h. Fresh Raney nickel was added every 8 h, the mixture was filtered and evaporated. The residue was chromatographed on 20 × 20 cm plates of silica gel to give 3β-acetoxypregn-5-en-20-one (*I*) (20 mg, 30%) identified by its IR and mass spectra. The chromatography also furnished a mixture (50 mg) of the starting thiazole *IV* and 3β-acetoxy-17β-[2-amino-4-(1,3-thiazolyl)]androst-5-ene (*VII*) characterized by mass spectrometry (molecular peak of *VII* (*m/z*): 414).

Reduction of the Thiazole *V* on Raney Nickel

The thiazole *V* (30 mg, 0·06 mmol) was refluxed with excess Raney nickel in ethanol (4 ml) for 40 h. Fresh Raney nickel was added every 8 h, the mixture was filtered and evaporated. Since the starting thiazole *V* and the reaction product *I* showed identical R_F values and could not be separated either by column or by thin layer chromatography, the mixture was analyzed by gas chromatography using a Labora (ČSSR) Chrom-4 instrument (phase OV-17-3F, 220°C, inlet chamber 280°C, pressure of the carrier gas 60 kPa). Along with the starting thiazole *V* the presence of pregnenolone acetate (*I*) was proved. Presence of *I* was also demonstrated by mass spectrometry (the spectrum is identical with that of *I*).

The authors thank Dr P. Sedmera, Microbiological Institute, Czechoslovak Academy of Sciences, Prague, for measurement of the ^{13}C-NMR *spectrum of the thiazole* VI. *Mass spectra were recorded by Dr A. Trka, the IR spectra by Mrs J. Pospíšilová and interpreted by Dr J. Smolíková.* ^{1}H-NMR *spectra were measured by Mrs J. Jelínková.*

REFERENCES

1. Ralls J. W., Bergstrom C. G.: U.S. 2, 793, 207 (1957); Chem. Abstr. *52*, 464 g (1958).
2. Schaub R. E., Weiss M. J.: J. Org. Chem. *26*, 1223 (1961).

3. Takamura K., Takama S.: Japan. 22, 578 (1963); Chem. Abstr. *60*, 4214b (1964).
4. Takamura K., Isono C., Takaku S., Nitta Y.: Chem. Pharm. Bull. *11*, 604 (1963).
5. Takamura K., Isono C., Takama S., Nitta Y.: Chem. Pharm. Bull. *11*, 613 (1963).
6. Takamura K., Takahisa S.: Japan. 18, 690 (1963); Chem. Abstr. *60*, 4217d (1964).
7. Takamura K., Takama S.: Japan. 22, 282 (1963); Chem. Abstr. *60*, 3058e (1964).
8. Dodson R. M.: U.S. 2, 705, 232 (1955); Chem. Abstr. *50*, 5793b (1956).
9. Hadáček J., Klímek M.: Publs. Fac. Sci. Masaryk Univ. *349*, 217 (1953); Chem. Abstr. *49*, 6288e (1955).
10. Catsoulacos P., Kallias D.: J. Heterocycl. Chem. *16*, 763 (1979).
11. Hantzsch A.: Justus Liebigs Ann. Chem. *249*, 1 (1888).
12. Vaněk T., Farkaš J., Gut J.: This Journal *44*, 1339 (1979).
13. Erlenmeyer H., Marbet R., Schenkel H.: Helv. Chim. Acta *28*, 922 (1945).
14. von Tuinman A., Iwasaki S., Schaffner K., Jeger O.: Helv. Chim. Acta *51*, 1778 (1968).
15. Sprague J. M., Land A. H. in the book: *Heterocyclic Compounds*, Vol. 5, (R. C. Elderfield, Ed.), p. 484. Wiley, New York 1957.
16. Burger A., Ullyot G. E.: J. Org. Chem. *12*, 342 (1947).
17. Brown C. A., Ahuja V. K.: J. Org. Chem. *38*, 2226 (1973).
18. Garnier R., Faure R., Babadjamian A., Vincent E. J.: Bull. Soc. Chim. Fr. *1972*, 1040.
19. Marquet A., Kagan H. B., Dvolaitzky M., Mamlok L., Weidmann C., Jacques J.: C. R., Acad. Sci. *248*, 984 (1959).

Translated by V. Černý.