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Note

Steroids and related studies

XXXII*. Thin-layer chromatography of some lactam, tetrazole, basic and quaternary azasteroids

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Our laboratory is active in the synthesis of azasteroids, and recently several lactams[2,3], tetrazoles[1,4], and basic and quaternary compounds[2,3] have been prepared. The purity of these compounds was ascertained by TLC, and in this paper we report the results of the chromatographic investigations; other communications on the TLC patterns of steroidal lactams have been published[5,6].

EXPERIMENTAL

Reagents

All the reagents were of analytical grade and were used as such.

Azasteroids

The lactams (Table I), tetrazoles (Table II) and basic (Table III) and quaternary (Table IV) azasteroids used were synthesized in this laboratory.

Method

The plates (200 × 200 × 3.5 mm) were coated with silica gel G (Merck) to a thickness of 0.25 mm and were activated at 110° for 1 h, then stored in a drying cabinet over calcium chloride.

Solutions of the azasteroids were prepared in methanol or chloroform and were applied to give 50–100 μg of the material as a spot on the plate.

The nine solvent systems used were chloroform, methanol, chloroform–methanol, chloroform–ethyl acetate, ethyl acetate–*n*-hexane, methanol–ammonia, benzene–methanol–ethyl acetate, ethanol–chloroform–ethyl acetate–water–hydrochloric acid, and *n*-butanol–acetic acid–water.

The following three systems proved to be of particular interest for steroidal lactams and tetrazoles:

(1) chloroform–methanol (19:1),

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* For the previous paper in this series, see ref. 1.

(2) benzene–methanol–ethyl acetate (17:2:1), and
(3) ethyl acetate–*n*-hexane (3:2).

The systems found to be appropriate for basic and quaternary azasteroids were:

(4) methanol–30% (w/w) ammonia solution (9:1),
(5) ethanol–chloroform–ethyl acetate–water–concentrated hydrochloric acid (12:12:12:2:1), and
(6) *n*-butanol–glacial acetic acid–water (5:3:2).

The chromatograms were run at room temperature (≃35°), and spots were detected with use of ceric sulphate solution (2 g in 100 ml of 10% (v/v) sulphuric acid), followed by heating at 150°, and by exposure to iodine vapour; permanent black spots were obtained with the ceric sulphate spray reagent. The development of spots with iodine vapour took only 2 to 4 min, and the spots were brown.

RESULTS

Tables I, II, III and IV list the R_F values of lactams, tetrazoles and basic and quaternary azasteroids, respectively. For the lactams, solvent systems 1 and 2 gave consistent results. For the tetrazoles, solvent system 3 was also found to be of interest. Solvent systems 4, 5 and 6 gave consistent results for basic azasteroids, and systems 5 and 6 were useful for quaternary compounds.

TABLE I

THIN-LAYER CHROMATOGRAPHY OF STEROIDAL LACTAMS IN SOLVENT SYSTEMS 1 AND 2

Compound	R_F *value in solvent*	
	1	*2*
4-Aza-5α-cholestan-3-one	0.56	0.43
17-Oxo-17a-aza-D-homoandrost-5-en-3β-yl acetate	0.43	0.34
3β-Hydroxy-17a-aza-D-homoandrost-5-en-17-one	0.22	0.19
17a-Aza-D-homoandrost-4-ene-3,17-dione	0.32	0.26
3-Hydroxyimino-17a-aza-D-homoandrost-4-en-17-one	0.20	0.26
4,17a-Diaza-D-homo-5α-androstane-3,17-dione	0.06	0.08
4-Hydroxy-4,17a-diaza-D-homo-5α-androstane-3,17-dione	0.10	0.10
4-Benzyl-4,17a-diaza-D-homoandrost-5-ene-3,17-dione	0.31	0.29
3,5-Seco-5,17-dioxo-17a-aza-D-homo-4-norandrostan-3-oic acid	—	0.03
3,5-Seco-5-hydroxyimino-17-oxo-17a-aza-D-homo-4-norandrostan-3-oic acid	—	0.03

The results suggest that solvent system 2 is useful for the TLC of steroidal lactams; this observation is in agreement with the work of Doorenbos and Sharma[5]. For steroidal tetrazoles, systems 1 and 3 can also be used with good results. System 1 may be particularly useful for detecting lactams and tetrazoles in mixtures resulting from the Schmidt reaction with steroidal ketones.

The quaternary compounds generally move slower than the related tertiary amines in systems 5 and 6, so that quaternisation experiments can be monitored, as was done by Bamford *et al.*[7], who used system 5.

TABLE II

THIN-LAYER CHROMATOGRAPHY OF STEROIDAL TETRAZOLES IN SOLVENT SYSTEMS 1, 2 AND 3

Compound	R_F value in solvent		
	1	*2*	*3*
3-Aza-A-homo-4a-cholesteno[3,4-*d*]tetrazole	0.73	0.73	0.86
(25*R*)-3-Aza-A-homo-4a-spirosteno[3,4-*d*]tetrazole	0.72	0.72	0.75
3-Aza-A-homo-4a,16-pregnadieno[3,4-*d*]tetrazol-20-one	0.66	0.55	0.47
3-Aza-A-homo-4a,16-pregnadieno[3,4-*d*]tetrazol-20-one oxime	0.52	0.53	0.68
17β-Acetamido-3-aza-A-homo-4a-androsteno[3,4-*d*]tetrazole	0.37	0.25	0.02
17β-(5-Methyltetrazol-1-yl)-3-aza-A-homo-4a-androsteno[3,4-*d*]tetrazole	0.42	0.32	0.05
3-Aza-A-homo-4a-androsteno[3,4-*d*]tetrazol-17-one	0.59	0.51	0.29
3-Aza-A-homo-4a-androsteno[3,4-*d*]tetrazol-17β-ol	0.38	0.37	0.27
3-Aza-A-homo-4a-androsteno[3,4-*d*]tetrazol-17β-yl acetate	0.73	0.65	0.61
3-Aza-A-homo-4a-androsteno[3,4-*d*]tetrazol-17-one oxime	0.40	0.37	0.24
3,17a-Diaza-A,D-bishomo-4a-androsteno[3,4-*d*]tetrazol-17-one	—	0.13	—
3,17a-Diaza-A,D-bishomo-4a-androsteno[3,4-*d*][17a,17-*d*]bistetrazole	0.65	0.43	0.28
13,17-Seco-13α-azido-A-homo-4a-androsteno[3,4-*d*]tetrazole 17-nitrile	0.66	0.43	0.26
3-Methoxy-17a-aza-D-homo-oestra-1,3,5(10)-trieno[17a,17-*d*]tetrazole	0.67	0.41	0.28
7a-Aza-B-homocholest-5-eno[7a,7-*d*]tetrazol-3β-yl acetate	0.75	0.69	0.77
7a-Aza-B-homocholest-4-eno[7a,7-*d*]tetrazol-3β-yl acetate	0.77	0.70	0.71
7a-Aza-B-homocholest-4-eno[7a,7-*d*]tetrazol-3β-ol	0.29	0.26	—
7a-Aza-B-homocholest-5-eno[7a,7-*d*]tetrazol-3β-ol	0.38	0.32	0.17
7a-Aza-B-homocholest-4-eno[7a,7-*d*]tetrazol-3-one	0.71	0.47	0.40

TABLE III

THIN-LAYER CHROMATOGRAPHY OF BASIC AZASTEROIDS IN SOLVENT SYSTEMS 4, 5 AND 6

Compound	R_F value in solvent		
	4	*5*	*6*
4-Aza-5α-cholestane	0.42	0.49	0.67
4-Methyl-4-aza-5α-cholestane	—	0.38	0.40
4-Aza-5α-androstan-17β-ol	0.41	0.54	0.57
4-Methyl-4-aza-5α-androstan-17β-ol	0.66	0.40	0.46
4-Methyl-4-aza-5α-androstan-17β-ylacetate	0.68	0.46	0.47
3β-Hydroxy-17a-aza-D-homoandrost-5-ene	0.25	0.49	0.55
17a-Methyl-17a-aza-D-homoandrost-5-en-3β-ol	0.45	0.40	0.42
17a-Methyl-17a-aza-D-homoandrost-4-en-3-one	0.45	0.32	0.35
17a-Methyl-17a-aza-D-homoandrost-5-en-3β-yl acetate	0.47	0.49	0.47
17a-Methyl-17a-aza-D-homo-4a-androsteno[3,4-*d*]tetrazole	0.42	0.25	0.25
4,17a-Diaza-D-homo-5α-androstane	0.08	0.13	0.17
4,17a-Dimethyl-4,17a-diaza-D-homo-5α-androstane	0.32	0.03	0.08
3β-Pyrrolidino-17a-aza-D-homoandrost-5-ene	0.19	0.19	0.23
3β-Pyrrolidino-17a-methyl-17a-aza-D-homoandrost-5-ene	0.34	0.10	0.13
3β-Pyrrolidino-17a-aza-D-homoandrost-5-en-17-one	0.62	0.27	0.36
17β-Amino-4-aza-5α-androstane	0.28	0.18	0.30
4-Methyl-17β-dimethylamino-4-aza-5α-androstane	0.61	0.06	0.13

TABLE IV

THIN-LAYER CHROMATOGRAPHY OF QUATERNARY AZASTEROIDS IN SOLVENT SYSTEMS 5 AND 6

Compound	*R_F value in solvent*	
	5	*6*
4-Methyl-4-aza-5α-cholestane methiodide	0.47	0.56
4-Methyl-4-aza-5α-androstan-17β-ol methiodide	0.16	0.26
4-Methyl-4-aza-5α-androstan-17β-yl acetate methiodide	0.29	0.28
17a-Methyl-17a-aza-D-homoandrost-5-en-3β-ol methiodide	0.17	0.24
17a-Methyl-17a-aza-D-homoandrost-4-en-3-one methiodide	0.11	0.16
4,17a-Dimethyl-4,17a-diaza-D-homo-5α-androstane dimethiodide	—	0.02
17a-Methyl-3β-pyrrolidino-17a-aza-D-homoandrost-5-ene dimethiodide	—	0.05
3β-Pyrrolidino-17a-aza-D-homoandrost-5-en-17-one methiodide	0.10	0.26
4-Methyl-17β-dimethylamino-4-aza-5α-androstane dimethiodide	—	0.08

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