Synthesis of Novel 5*H*,11*H*-Pyrido[2',3':2,3]thiopyrano[4,3-*b*]-indoles by Fischer-Indole Cyclization

Antonio Da Settimo, Anna Maria Marini*, Giampaolo Primofiore, Federico Da Settimo, Silvia Salerno, Concettina La Motta, Gianluca Pardi, Pier Luigi Ferrarini and Claudio Mori

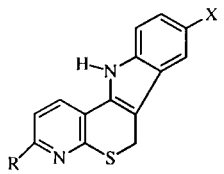
Dipartimento di Scienze Farmaceutiche, Via Bonanno 6, I-56126 Pisa, Italy
Received August 10, 1999

The synthesis of the title compounds 5*H*,11*H*-pyrido[2',3':2,3]thiopyrano[4,3-*b*]indoles was accomplished by the Fischer indole cyclization of some 2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one phenylhydrazones and 7-methyl-2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one phenylhydrazones. The synthesis of the new 2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one, which was used as one of the starting compounds, is also described.

J. Heterocyclic Chem., **37**, 379 (2000).

In recent years several papers dealing with the mechanism by which anticancer drugs exert their action at the molecular level have aroused considerable interest. Many different targets have been identified and characterized, including DNA and enzymes involved in processing nucleic acids, such as topoisomerases [1-3]. Intercalation of planar aromatic or heteroaromatic compounds with DNA is one of the important modes of actions in DNA-drug interaction [4,5], based on the ability of the chromophore to bind strongly between the base pairs. In this field, the promising antitumor properties shown by ellipticine [6,7], a tetracyclic indole alkaloid, have attracted considerable interest towards the synthesis and biological evaluation of a number of other related compounds [8].

As a part of our studies concerning the synthesis of biologically active heterocycles [9,10], we were especially interested in the field of indole derivatives and we had previously described a number of [1]benzoxepinoindoles [11] and indolo[1,8]naphthyridines [12,13]. In this paper we wish now to report the synthesis of some substituted 5*H*,11*H*-pyrido[2',3':2,3]thiopyrano[4,3-*b*]indoles **1** and **2**, which represent a new heterocyclic ring system.



1: R=H
2: R=CH_3

The key intermediates for the synthesis of the desired indole derivatives **1** and **2** were the 7-methyl-2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one **4** [14], and the new unsubstituted compound **3**, which was prepared by the route summarized in Scheme 1.

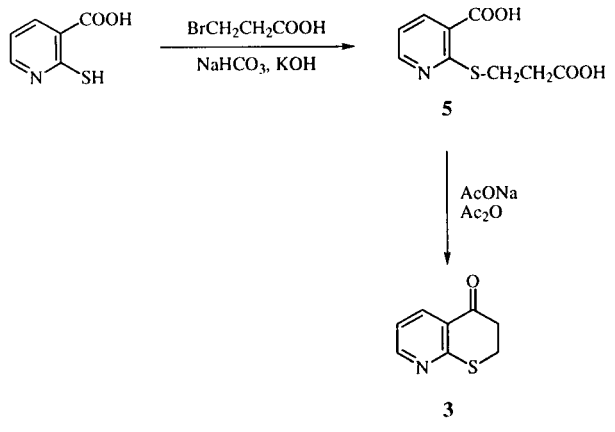
Reaction of 2-mercaptopyridin-3-carboxylic acid with 3-bromopropionic acid in aqueous potassium hydroxide solution gave the 3-(3-carboxy-2-pyridylthio)propionic acid **5** in a very good yield (98%), which by treatment with anhydrous sodium acetate and acetic anhydride, afforded the desired 2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one **3**.

The 2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one **3** and 7-methyl-2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one **4** were then converted to the corresponding phenylhydrazones **6a-d** and **7a-d**, respectively, by treatment with the appropriately substituted phenylhydrazine hydrochlorides in ethanolic solution.

The title compounds **1a-d** and **2a-d** were obtained by Fischer indole cyclization on the phenylhydrazones **6a-d** and **7a-d**, respectively, in the presence of 12% hydrochloric acid, with yields ranging from 40-70% (Scheme 2).

The proposed structures of all products were confirmed by analytical, ir, ^{1}H nmr and mass spectral data. Evidence for the structure of indole derivatives **1** and **2** derived from an examination of their ^{1}H nmr spectra, in which the disappearance was observed of the signals at $\delta \cong 3$ assigned to the methylene groups in the 3 position of the starting phenylhydrazones **6** and **7**. The broad exchangeable singlet due to the NH, resonating generally at $\delta \cong 11$, confirmed the proposed indole structure. Moreover, in the mass spectra of **1** and **2**, the base peaks were those corresponding to the values of the molecular weight of the cyclized compounds, suggesting a very good stability for them (Tables I and II).

Scheme 1



Scheme 2

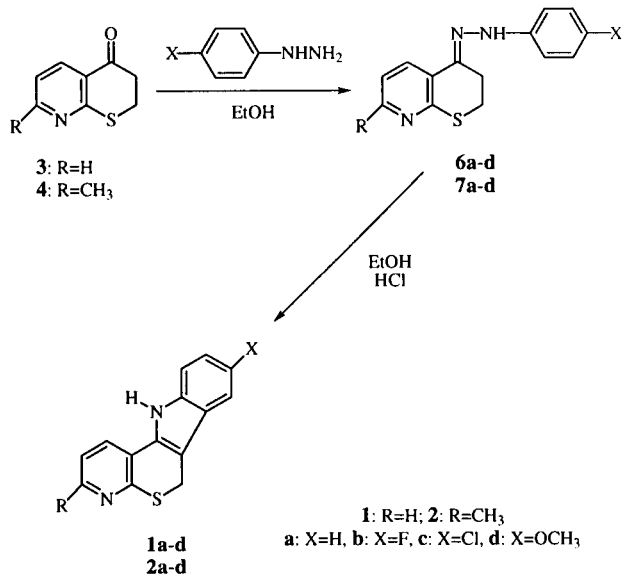
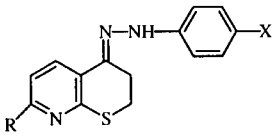
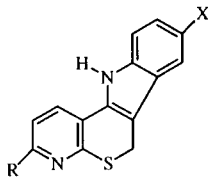


Table I
Physical and Spectral Data of Compounds **6a-d** and **7a-d**



No.	R	X	Yield (%)	Mp °C (Recrystallization solvent)	^{1}H NMR (δ ppm)	MS m/z	Molecular Formula	Analysis (%) C	H	N
6a	H	H	97	172-175 (Methanol)	3.03 (d, 2H, 2-CH_2), 3.17 (d, 2H, 3-CH_2), 6.91-7.65 (m, 6H, Ar-H), 8.24-8.64 (m, 2H, ArH), 9.81 (br s, 1H, NH exch.)	255	$C_{14}H_{13}N_3S$	65.85 65.58	5.13 5.15	16.46 16.33
6b	H	F	95	148-150 (Methanol)	3.02 (d, 2H, 2-CH_2), 3.11 (d, 2H, 3-CH_2), 6.01-7.26 (m, 5H, Ar-H), 8.18-8.40 (m, 2H, ArH), 9.45 (br s, 1H, NH exch.)	273	$C_{14}H_{12}FN_3S$	61.52 61.27	4.42 4.06	15.37 15.19
6c	H	Cl	89	190-195 (Methanol)	3.02 (d, 2H, 2-CH_2), 3.10 (d, 2H, 3-CH_2), 7.03-7.27 (m, 5H, Ar-H), 8.18-8.40 (m, 2H, ArH), 9.58 (br s, 1H, NH exch.)	289	$C_{14}H_{12}ClN_3S$	58.03 58.15	4.17 3.83	14.50 14.23
6d	H	OCH_3	93	144-147 (Methanol)	3.02 (d, 2H, 2-CH_2), 3.08 (d, 2H, 3-CH_2), 4.41 (s, 3H, OCH_3), 6.70-7.31 (m, 5H, Ar-H), 7.84-8.17 (m, 2H, ArH), 9.28 (br s, 1H, NH exch.)	285	$C_{15}H_{15}N_3OS$	63.13 63.56	5.30 5.33	14.72 14.38
7a	CH_3	H	93	121-125 (Methanol)	2.52 (s, 3H, 7-CH_3), 3.02 (d, 2H, 2-CH_2), 3.32 (d, 2H, 3-CH_2), 6.77-7.26 (m, 6H, Ar-H), 8.50 (d, 1H, ArH), 9.58 (br s, 1H, NH exch.)	269	$C_{15}H_{15}N_3S$	66.88 66.78	5.61 5.35	14.62 14.93
7b	CH_3	F	92	181-184 (Methanol)	2.48 (s, 3H, 7-CH_3), 3.02 (d, 2H, 2-CH_2), 3.28 (d, 2H, 3-CH_2), 6.93-7.37 (m, 5H, Ar-H), 8.52 (d, 1H, ArH), 9.58 (br s, 1H, NH exch.)	287	$C_{15}H_{14}FN_3S$	62.70 62.52	4.91 4.62	12.99 13.37
7c	CH_3	Cl	54	97-100 (Methanol)	2.39 (s, 3H, 7-CH_3), 3.02 (d, 2H, 2-CH_2), 3.15 (d, 2H, 3-CH_2), 6.92-7.27 (m, 5H, Ar-H), 8.23 (d, 1H, ArH), 9.56 (br s, 1H, NH exch).	303	$C_{15}H_{14}ClN_3S$	59.30 59.50	4.64 4.31	13.83 13.48
7d	CH_3	OCH_3	30	132-135 (Methanol)	2.38 (s, 3H, 7-CH_3), 3.20 (d, 2H, 2-CH_2), 3.28 (d, 2H, 3-CH_2), 3.76 (s, 3H, OCH_3), 6.68-7.29 (m, 5H, Ar-H), 7.78 (d, 1H, ArH), 9.29 (br s, 1H, NH exch.)	299	$C_{16}H_{17}N_3OS$	64.19 64.60	5.72 5.77	14.03 13.96

Table II
Physical and Spectral Data of Compounds **1a-d** and **2a-d**



No.	R	X	Yield (%)	Mp °C (Recrystallization solvent)	^{1}H NMR (δ ppm)	MS m/z	Molecular Formula	Analysis (%) C	H	N
1a	H	H	41	215-217 (Methanol)	4.45 (s, 2H, 5-CH_2), 6.99-7.55 (m, 5H, Ar-H), 7.88-8.19 (dd, 2H, ArH), 11.55 (br s, 1H, 11-NH exch.)	238	$C_{14}H_{10}N_2S$	70.56 70.32	4.23 4.44	11.75 11.96
1b	H	F	44	252-256 (Methanol)	4.40 (s, 2H, 5-CH_2), 6.80-7.43 (m, 4H, Ar-H), 7.86-8.20 (dd, 2H, ArH), 11.58 (br s, 1H, 11-NH exch.)	256	$C_{14}H_9FN_2S$	65.61 65.32	3.54 3.17	10.93 10.68
1c	H	Cl	80	258-262 (Methanol)	4.46 (s, 2H, 5-CH_2), 7.17-7.62 (m, 4H, Ar-H), 7.89-8.16 (dd, 2H, ArH), 11.82 (br s, 1H, 11-NH exch.)	272	$C_{14}H_9ClN_2S$	61.65 61.58	3.32 3.10	10.27 9.95
1d	H	OCH_3	38	255-260 (Methanol)	3.76 (s, 3H, 3-OCH_3), 4.42 (s, 2H, 5-CH_2), 6.70-7.31 (m, 4H, Ar-H), 7.84-8.18 (dd, 2H, ArH), 11.39 (br s, 1H, 11-NH exch.)	268	$C_{15}H_{12}N_2OS$	67.14 67.28	4.51 4.17	10.44 10.12
2a	CH_3	H	53	192-196 (Methanol)	2.49 (s, 3H, 8-CH_3), 4.52 (s, 2H, 5-CH_2), 6.99-7.44 (m, 5H, Ar-H), 8.11 (d, 1H, ArH), 11.82 (br s, 1H, 11-NH exch.)	252	$C_{15}H_{12}N_2S$	71.40 71.08	4.79 4.49	11.10 11.38
2b	CH_3	F	78	207-210 (Methanol)	2.45 (s, 3H, 8-CH_3), 4.44 (s, 2H, 5-CH_2), 7.07-7.33 (m, 4H, Ar-H), 7.94-8.04 (d, 1H, ArH), 11.88 (br s, 1H, 11-NH exch.)	270	$C_{15}H_{11}FN_2S$	66.65 66.34	4.10 3.91	10.36 10.03
2c	CH_3	Cl	71	218-222 (Methanol)	2.45 (s, 3H, 8-CH_3), 4.45 (s, 2H, 5-CH_2), 7.07-7.57 (m, 4H, Ar-H), 7.92-8.03 (d, 1H, ArH), 11.83 (br s, 1H, 11-NH exch.)	286	$C_{15}H_{11}ClN_2S$	62.82 62.53	3.87 4.02	9.77 9.42
2d	CH_3	OCH_3	49	221-224 (Methanol)	2.38 (s, 3H, 8-CH_3), 3.75 (s, 3H, 3-OCH_3), 4.38 (s, 2H, 5-CH_2), 6.67-7.29 (m, 4H, Ar-H), 7.78 (d, 1H, ArH), 11.27 (br s, 1H, 11-NH exch.)	282	$C_{16}H_{14}N_2OS$	68.06 67.99	4.99 4.98	9.92 9.65

EXPERIMENTAL

Melting points were determined using a Reichert Köfler hot-stage apparatus and are uncorrected. Infrared spectra were obtained on a PYE/UNICAM Model PU 9561 spectrophotometer as Nujol mulls. Nuclear magnetic resonance spectra were recorded on a Varian CFT-20 spectrometer, in dimethyl-d_6 sulfoxide solution, using tetramethylsilane as the internal standard. Mass spectra were obtained on a Hewlett-Packard 5988 A spectrometer using a direct injection probe and an electron beam energy of 70 eV. Magnesium sulfate was always used as the drying agent. Evaporations were performed *in vacuo* (rotating evaporator). Analytical tlc was carried out on Merck 0.2 mm precoated silica gel aluminium sheets (60 F-254). Elemental analyses were performed by our Analytical Laboratory.

3-(3-Carboxy-2-pyridylthio)propionic Acid **5**.

3-Bromopropionic acid (11.83 g, 77.3 mmoles) in 50 ml of water and 6.5 g (72.0 mmoles) of sodium hydrogen carbonate were added to a solution of 2-mercaptopyridine-3-carboxylic acid (10 g, 64.5 mmoles) in 90 ml of 10% potassium hydroxide aqueous solution. The reaction mixture was stirred at 60 °C for 4 hours, cooled and acidified with concentrated hydrochloric acid to *p*H 2. The solid precipitate product was collected and washed with water to give 14.52 g (98% yield) of pure **5**, mp 205-208 °C (dec) (ethyl acetate); ^{1}H nmr (dimethyl-d_6 sulfoxide) δ: 2.61 (t, 2H, CH_2COOH), 3.26 (t, 2H, SCH_2), 7.17 (dd, 1H, 5-H), 8.16 (dd, 1H, 4-H), 8.57 (dd, 1H, 6-H), 12.6 (bs, 1H, COOH); ms: m/z 227 (M^+), 45 (100).

Anal. Calcd. for $C_9H_9NO_4S$: C, 47.58; H, 3.96; N, 6.17; Found: C, 47.50; H, 3.77; N, 5.71.

2,3-Dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one **3**.

A solution of 3-(3-carboxy-2-pyridylthio)propionic acid **5** (8.0 g, 35.2 mmoles) and anhydrous sodium acetate (5.376 g, 64 mmoles) in 72 ml of acetic anhydride was refluxed at 160 °C for 1.5 hours. After cooling, the reaction mixture was diluted with water, basified with 30% ammonium hydroxide solution to *p*H 8-9 and, after filtering off the dark material, extracted with ethyl acetate. The combined extracts were washed with water, dried and evaporated to give 4.6 g of crude **3** as an orange oil. Purification was made by filtration on a silica gel (60/0.040-0.063 mm) chromatographic column, using petroleum ether 60-80 °C/ethyl acetate 7:3 as the eluting system. The product recovered from the less mobile fractions (tlc) gave 0.87 g (25% yield) of pure **3**, mp 66-67 °C (petroleum ether 60-80 °C); ^{1}H nmr

(dimethyl-d_6 sulfoxide) δ: 2.81-3.01 (m, 2H, 3-CH_2), 3.24-3.43 (m, 2H, 2-CH_2), 7.20 (dd, 1H, 6-H), 8.16 (dd, 1H, 5-H), 8.48 (dd, 1H, 7-H); ms: m/z 165 (M^+), 137 (100).

Anal. Calcd. for C_8H_7NOS: C, 58.18; H, 4.24; N, 8.48; Found: C, 58.00; H, 3.92; N, 8.10.

2,3-Dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one Phenylhydrazones **6a-d** and 7-methyl-2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one Phenylhydrazones **7a-d**.

General Procedure.

A suspension of 2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one **3** or 7-methyl-2,3-dihydrothiopyrano[2,3-*b*]pyridin-4(4*H*)-one **4** (1.2 mmoles) and of 1.4 mmoles of the appropriately substituted phenylhydrazine hydrochloride in 5 ml of ethanol was refluxed until disappearance of the starting reagents (1-8 hours, tlc analysis). After cooling, the precipitate obtained was treated with 10% sodium hydroxide aqueous solution to give compounds **6a-d** and **7a-d**, which were purified by recrystallization from methanol (Table I).

5*H*,11*H*-Pyrido[2',3':2,3]thiopyrano[4,3-*b*]indoles **1a-d** and 8-Methyl-5*H*,11*H*-pyrido[2',3':2,3]thiopyrano[4,3-*b*]indoles **2a-d**.

General Procedure.

A solution of the appropriate phenylhydrazone **6a-d** or **7a-d** (1 mmole) in 3 ml of 12% hydrochloric acid and 5 ml of ethanol was refluxed for 12 hours. After cooling, the indole derivatives **1a-d** and **2a-d** were obtained as crude hydrochlorides. Treatment with 10% sodium hydroxide aqueous solution and recrystallization from methanol afforded pure compounds **1a-d** and **2a-d** (Table II).

Acknowledgments.

This work was supported by grants from the Ministry of University and Scientific and Technological Research (MURST) (Research fund 60%).

REFERENCES AND NOTES

[1] L. F. Liu, *Ann. Rev. Biochem.*, **58**, 351-375 (1989).

[2] G. Capranico, M. Binaschi, M. E. Borghetto, F. Zunino, and M. Palumbo, *Trends Pharm. Sci.*, **18**, 323-329 (1997).

[3] Y. Pommier, G. Capranico, A. Orr, and K. W. Kohn, *J. Mol. Biol.*, **222**, 909 (1991).

[4] W. A. Denny, Cancer Chemotherapeutic Agents, W. Foye, ed, ACS, Washington D.C, pp 218-239 (1995).

[5] U. Pindur and G. Fisher, *Curr. Med. Chem*, **3**, 379 (1996).

[6] G. W. Gribble, The Alkaloids, **39**, A. Brossi, ed, Academic Press, New York, p 239 (1990).

[7] M. Ohashi and T. Oki, *Exp. Opin. Ther. Patents*, **6**, 1285 (1996).

[8] M. Y. Vilarem, I. Y. Charcosset, F. Primaux, M. P. Gras, and F. Calvo, *Cancer Res.*, **45**, 3906 (1985).

[9] A. Da Settimo, G. Primofiore, M. C. Bertacchi, F. Da Settimo, and A. M. Marini, *J. Heterocyclic Chem.*, **32**, 941 (1995).

[10] A. Da Settimo, A. M. Marini, G. Primofiore, F. Da Settimo, S. Salerno, G. Viola, L. Dalla Via, and S. Marciani Magno, *Eur. J. Med. Chem.*, **33**, 685 (1998).

[11] F. Chetoni, F. Da Settimo, A. M. Marini, and G. Primofiore, *J. Heterocyclic Chem.*, **30**, 1481 (1993).

[12] A. Da Settimo, G. Primofiore, G. Biagi, and V. Santerini, *J. Heterocyclic Chem.*, **13**, 97 (1976).

[13] A. Da Settimo, G. Primofiore, G. Biagi, V. Santerini, and L. D'Amico, *J. Org. Chem.*, **42**, 1725 (1977).

[14] P. L. Ferrarini, C. Mori, M. Badawneh, V. Calderone, R. Greco, C. Manera, A. Martinelli, P. Nieri, and G. Saccomanni, *Eur. J. Med. Chem.*, in press.