[CONTRIBUTION FROM THE SQUIBB INSTITUTE FOR MEDICAL RESEARCH]

Analogs of Aspergillic Acid. IV. Substituted 2-Bromopyridine-N-oxides and Their Conversion to Cyclic Thiohydroxamic Acids[1]

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The antibiotic aspergillic acid (X) contains a cyclic hydroxamic acid functional group in a pyrazine nucleus.[2] The synthesis of a pyridine analog (V) was accomplished using steps leading to the tautomeric 2-hydroxypyridine-N-oxide (IV),[3] that is, by oxidation of 2-pyridyl ethers followed by cleavage of the resultant 2-pyridyl ether N-oxides. Since the desired sulfur analog could not be obtained by oxidation of a 2-pyridyl thioether due to the ease of oxidation of the divalent sulfur, the nitrogen was oxidized before the introduction of the sulfur into the molecule. In the preparation of the parent compound, N-hydroxy-2-pyridinethione (III), 2-bromopyridine (I) was converted to its N-oxide (II) by oxidation with perbenzoic acid or peracetic acid. Treatment of II with sodium sulfide or sodium hydrosulfide under rather mild conditions gave the thiohydroxamic acid (III), since the bromine atom in 2-bromopyridine-N-oxide is considerably more reactive than in 2-bromopyridine itself. Further indication of the lability of the bromine atom in II is shown by its reaction in aqueous alkali to form N-hydroxy-2-pyridone (V). These reactions were achieved by brief heating at steam-bath temperature. By contrast, 2-chloropyridine and 2-iodopyridine were heated for several hours with potassium hydrosulfide in sealed tubes at 140 and 200°, respectively, in the preparation of 2-mercaptopyridine.[4,5] Similarly, the conversion of 2-chloropyridine to 2-hydroxypyridine was carried out at 175°.[6]

The synthesis of N-hydroxy-2-pyridinethione (III) was also accomplished by the reaction of 2-bromopyridine-N-oxide (II) with thiourea to form 2-pyridyl-N-oxide-isothiourea hydrobromide (IX), followed by treatment of (IX) with aqueous sodium carbonate.[7]

It was shown that a divalent sulfur atom is oxidized before the nitrogen in the pyridine ring, since the oxidation of 2-benzylmercaptopyridine (VII) gave a compound (VIII) which melted at 87-88°; while the reaction of 2-bromopyridine-N-oxide (II) with sodium benzylmercaptide gave the expected 2-benzylmercaptopyridine-N-oxide (VI), m. p. 168–169°. Compounds VI and VIII are isomeric so it may be assumed that VIII is 2-pyridyl benzyl sulfoxide.

In addition to N-hydroxy-2-pyridinethione, several ring-substituted derivatives were also prepared. Oxidation of substituted 2-bromopyridines with perbenzoic acid gave the corresponding 2-bromopyridine-N-oxides (Table I). These were then converted to the corresponding substituted cyclothiohydroxamic acids by reaction with aqueous sodium hydrosulfide (Table II).

The N-hydroxy-2-pyridinethiones represent a new class of compounds, being the first cyclic thiohydroxamic acids synthesized. The compounds, which are sternutators, show high *in vitro* antibacterial activity against several different organisms. These results are given in Table III. For purposes of comparison, the activities of aspergillic acid and N-hydroxy-2-pyridone are also included. More complete pharmacological testing of these compounds is being carried out and will be reported subsequently.

(1) For the previous paper in this series see Shaw and McDowell, THIS JOURNAL, **71**, 1691 (1949).

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(2) Dutcher, *J. Biol. Chem.*, **171**, 321 (1947).

(3) Shaw, THIS JOURNAL, **71**, 67 (1949).

(4) Marckwald, Klemm and Trahert, *Ber.*, **33**, 1556 (1900).

(5) van Gastel and Wihaut, *Rec. trav. chim.*, **53**, 1031 (1934).

(6) British Patent 288,628.

(7) Surrey and Lindwall, THIS JOURNAL, **62**, 1696 (1940).

TABLE I

2-BROMOPYRIDINE-N-OXIDE HYDROCHLORIDES

Substituent[a]	Yield, %	M. p., °C.	Empirical formula	Nitrogen, % Calcd.	Nitrogen, % Found	Chlorine, % Calcd.	Chlorine, % Found
.....	60[b]	135–136	$C_5H_5BrClNO$	6.65	6.86	16.85	16.80
3-CH_3	67[c]	179–180	$C_6H_7BrClNO$	6.24	6.07	15.83	15.99
4-CH_3	59[c]	147–148	$C_6H_7BrClNO$	6.24	6.38	15.83	15.55
5-CH_3[d]	64[c]	141–142	$C_6H_7BrClNO$	6.24	5.63	15.83	15.67
6-CH_3[e]	61[c]	185–186	$C_6H_7BrClNO$	6.24	5.97	15.83	15.52
3-OC_2H_5	52[b]	159–160	$C_7H_9BrClNO_2$	5.53	6.01	13.93	13.55
5-Br	6.5[c]	165–166	$C_5H_4Br_2ClNO$	4.83	4.75	12.25	11.97

[a] The substituted 2-bromopyridine N-oxide hydrochlorides were prepared by Procedure A, Experimental Section. [b] Crystallized from absolute alcohol and ether. [c] Crystallized from absolute alcohol. [d] Prepared similarly to 2-bromo-4-methylpyridine, Lott and Shaw, THIS JOURNAL, 71, 70 (1949), in 77% yield; b. p., 97° at 14 mm.; m. p. 42–43° (crystallized from hexane). *Anal.* Calcd. for C_6H_6BrN: Br, 46.45. Found: Br, 46.84. [e] Prepared similarly to 2-bromo-4-methylpyridine, see (*d*); b. p. 77° at 9 mm. *Anal.* Calcd. for C_6H_6BrN: Br, 46.45. Found: Br, 46.35.

TABLE II

N-HYDROXY-2-PYRIDINETHIONES

Substituent[a]	Yield,[b] %	M. p., °C.	Empirical formula	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found	Nitrogen, % Calcd.	Nitrogen, % Found
.....	61[c]	68–70	C_5H_5NOS	47.21	46.94	3.96	3.98	11.01	10.97
3-CH_3	52[d]	74–75	C_6H_7NOS	51.02	50.75	4.99	5.24	9.92	9.54
4-CH_3	60	59–61	C_6H_7NOS	51.02	50.87	4.99	5.31	9.92	9.80
5-CH_3	53	106–107	C_6H_7NOS	51.02	51.12	4.99	5.17	9.92	9.77
6-CH_3	50	52–54	C_6H_7NOS	51.02	51.16	4.99	4.82	9.92	9.92
3-OC_2H_5	85	101–103	$C_7H_9NO_2S$	49.11	48.87	5.29	5.57	8.18	8.54
5-Br	40	130–131	C_5H_4BrNOS	29.14	29.43	1.95	2.19	6.79	6.74

[a] The substituted N-hydroxy-2-pyridinethiones were prepared by Procedure C, Experimental Section. [b] Crystallized from aqueous alcohol. The yields varied appreciably with the purity of the sodium hydrosulfide used, maximum yields being obtained with fresh samples supplied by the Hooker Electrochemical Co., Buffalo, N. Y. [c] Also prepared in 56% yield by Procedure D, Experimental Section. [d] Also prepared in 15% yield by Procedure D, Experimental Section.

TABLE III

THE *in Vitro* ANTIBACTERIAL ACTIVITIES[a]

	Minimal inhibiting concentration microgram/ml. *Staphylococcus aureus* P209[b]	*Klebsiella pneumoniae*[b]	Bacillus of Calmette and Guerin[c]
Aspergillic acid	20	30	4
N-Hydroxy-2-pyridone	3	40	2
N-Hydroxy-2-pyridinethione	0.06	1.5	0.006
N-Hydroxy-3-methyl-2-pyridinethione	.06	0.6	.004
N-Hydroxy-4-methyl-2-pyridinethione	.08	1.5	.001
N-Hydroxy-5-methyl-2-pyridinethione	.07	1.5	.005
N-Hydroxy-6-methyl-2-pyridinethione	.1	3.5	.003
N-Hydroxy-3-ethoxy-2-pyridinethione	.08	1.5	.03
N-Hydroxy-5-bromo-2-pyridinethione	.1	2.0	.008

[a] Activities determined by the Division of Microbiology, Research and Development Laboratories, E. R. Squibb & Sons, New Brunswick, N.J. [b] Tested in yeast beef broth. [c] Tested in modified Kirchner's medium; Rake, Jambor, McKee, Pansy, Wiselogle and Donovick, *Am. Rev. Tuber.*, 60, 121 (1949).

Experimental[8]

2-Bromopyridine-N-oxide Hydrochloride. A. By Perbenzoic Acid Oxidation.—Thirty-five grams (0.22 mole) of 2-bromopyridine was added to a chloroform solution of perbenzoic acid containing a 20% excess of the oxidizing agent. After four days at room temperature, the chloroform solution was extracted with three 150-cc. portions of 20% hydrochloric acid. The acidic extracts were concentrated to dryness under reduced pressure and the residue crystallized from an alcohol solution by the addition of ether, yielding 28 g. (60%), of colorless crystals, m. p. 135–136°.

B. By Peracetic Acid Oxidation.—One hundred fifty grams (0.75 mole) of 40% peracetic acid was added slowly to 79 g. (0.5 mole) of 2-bromopyridine at 10–15°. The temperature rose rapidly but was kept below 40° by external cooling until the initial reaction had subsided; then the solution was heated at 45–50° for 24 hours. The solution was concentrated to one-half its volume at 2–3 mm. (bath temperature, 30°). The residue was added to cracked ice and made strongly alkaline with 40% potassium hydroxide solution, keeping the temperature at 5°. The product was extracted with three 300-cc. portions of chloroform, and the combined chloroform solutions then extracted with three 300-cc. portions of 20% hydrochloric acid. The aqueous layer was concentrated to dryness under reduced pressure and the residue treated as in **A** to give 61 g. (70%), m. p. 130–135°. There was no depression with the sample of 2-bromopyridine-N-oxide hydrochloride prepared by method **A**.

N-Hydroxy-2-pyridinethione. C. By Reaction with Sodium Hydrosulfide.—Sixty grams (0.31 mole) of 2-bromopyridine-N-oxide hydrochloride in 75 cc. of water was neutralized with 25% sodium hydroxide solution. A solution of 32 g. of sodium hydrosulfite in 150 cc. of water was added portionwise with heating (steam-bath) during one hour. After an additional one-half hour heating, the solution was cooled, filtered and acidified with 6 *N* hydrochloric acid. The crystalline precipitate weighed 24 g. (61%), m. p. 65–68°. After recrystallization from aqueous alcohol the sample melted at 68–70°. N-Hydroxy-2-pyridinethione gives a deep blue color with ferric chloride solution. A similar result was obtained when sodium sulfide was added to the N-oxide hydrochloride in an equimolar amount.

D. By Reaction with Thiourea.—A solution of 19.4 g. (0.1 mole) of 2-bromopyridine-N-oxide hydrochloride and 9.7 g. (0.1 mole) of thiourea in 300 cc. of absolute alcohol was refluxed for one hour. The precipitate of 2-pyridyl-N-

(8) All melting points are uncorrected. Microanalyses were carried out by Mr. J. F. Alicino.

oxide-isothiourea hydrobromide which formed almost immediately was filtered and weighed 18 g. (72%), m. p. 160–160.5° (dec.). Crystallization from absolute alcohol did not change the melting point.

Anal. Calcd. for $C_6H_8BrN_3OS$: C, 28.89; H, 3.22. Found: C, 28.68; H, 3.54.

A solution of 12.5 g. (0.05 mole) of the above thiourea addition compound and 10 g. of sodium carbonate in 125 cc. of water was allowed to stand four hours at room temperature. The solution was acidified with 20% hydrochloric acid and yielded 5 g. (78%) of product, m. p. 65–67°. There was no depression with an authentic sample of N-hydroxy-2-pyridinethione.

N-Hydroxy-2-pyridone.—Seven grams (0.031 mole) of 2-bromopyridine-N-oxide hydrochloride was heated on a steam-bath with 50 cc. of 10% sodium hydroxide solution for one and one-half hours. The cooled solution, after acidification with concentrated hydrochloric acid, was concentrated to dryness under reduced pressure. The residue was dissolved in water, neutralized and filtered. The addition of aqueous cupric acetate to the filtrate precipitated the copper salt of N-hydroxy-2-pyridone, 2.5 g. (53%), m. p. 283–284°, undepressed when mixed with an authentic sample.[8]

2-Benzylmercaptopyridine-N-oxide.—To a solution of 1 g. (0.043 mole) of sodium in 30 cc. of absolute alcohol were added 4.5 cc. (0.036 mole) of benzyl mercaptan and 2.5 g. (0.012 mole) of 2-bromopyridine-N-oxide hydrochloride. The mixture was warmed at 50° for one hour, then left at room temperature for two hours. After the addition of excess 10% sodium hydroxide, the solution was extracted with ethyl acetate. The organic layer was concentrated to yield 1.2 g. (46%) of product, m. p. 167–169°. After recrystallization from ethyl acetate the compound melted at 168–169°. *Anal.* Calcd. for $C_{12}H_{11}NOS$: C, 66.32; H, 5.10; N, 6.45. Found: C, 66.42; H, 5.14; N, 6.25.

2-Pyridyl Benzyl Sulfoxide.—Ten grams (0.05 mole) of 2-benzylmercaptopyridine was added to a chloroform solution containing 0.05 mole of perbenzoic acid. Since the reaction was exothermic, the reaction mixture was cooled in running water. Titration, after a few minutes, indicated that all the perbenzoic acid had reacted. The chloroform solution was washed with aqueous sodium carbonate and dried. Removal of the solvent left a crystalline residue which was recrystallized from ethyl acetate and hexane, yielding 7 g. (68%), m. p. 87–88°. *Anal.* Calcd. for $C_{12}H_{11}NOS$: C, 66.32; H, 5.10; N, 6.45. Found: C, 66.17; H, 5.10; N, 6.28.

Summary

Substituted 2-bromopyridines have been oxidized to the corresponding 2-bromopyridine-N-oxides by perbenzoic acid or peracetic acid. The labilized bromine atom was readily replaced by alkaline reagents; treatment with sodium hydroxide yielded the cyclic hydroxamic acid, while treatment with sodium hydrosulfide or sodium sulfide gave the thio analog.

The cyclic thiohydroxamic acid was also obtained by reaction of the 2-bromopyridine-N-oxide with thiourea, followed by decomposition of the addition compound with sodium carbonate.

The cyclic thiohydroxamic acids show high *in vitro* antibacterial activity against a variety or organisms.

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[CONTRIBUTION FROM THE LILLY RESEARCH LABORATORIES]

Preparation of Some Homologs of Papaverine

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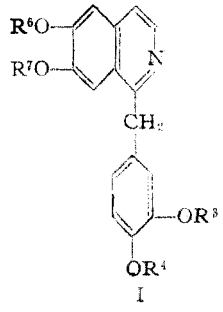
Although there have been a multiplicity of synthetic papaverine-like isoquinolines reported in the chemical literature, most have been 3-methyl or 1-phenyl isoquinolines. We found that of the sixteen possible methoxy-ethoxy homologs (I) of papaverine ($R^{3,4,6,7} = CH_3$),



R = CH_3, C_2H_5

I

only three besides papaverine are known: I, $R^{3,4,6,7} = C_2H_5$ ("Perparin"); $R^{6,7} = C_2H_5$ and $R^{3,4} = CH_3$; $R^{6,7} = CH_3$ and $R^{3,4} = C_2H_5$. Since the patents[1] revealing these three compounds and subsequent pharmacological literature[2] report varying activities and toxicities, it became of interest to prepare the entire group of homologs (I) for pharmacological evaluation.

Since in our hands the Bischler–Napieralski closure and subsequent catalytic dehydrogenation have proven somewhat difficult when operating with much less than twenty to twenty-five grams of phenethylamide (A), the recorded preparative methods for several of the prerequisite amines and acids were felt to be less desirable for the larger amounts needed in this and subsequent work. For the preparation of the required benzyl alcohols from the corresponding aldehydes we preferred catalytic hydrogenation over copper chromite insofar as reliability of results and convenience of handling were concerned.

In attempting to apply the simple preparation of the benzyl chlorides by the use of anhydrous hydrogen chloride[3] excellent results were obtained

(1) German Patent 574,656; also French Patent 719,638 and U. S. Patent 1,962,224.

(2) Issekutz, Leinzinger and Divner, *Arch. exptl. Path. Pharmakol.*, 164, 158, 173 (1932); Longecker and Starkenstein, *Klin. Wchschr.*, 10, 2257 (1931).

(3) Cannizzaro and Bertagnini, *Ann.*, **98**, 191 (1856).