The Synthesis of some 4-Nitroisothiazoles as Potential Antifungal Agents

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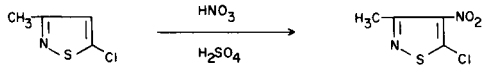
Received June 29, 1979

5-Chloro-3-methyl-4-nitroisothiazole (III) was prepared by nitration of 5-chloro-3-methylisothiazole. Compound III was found to exhibit significant antifungal activity *in vitro* against a wide spectrum of fungi. The synthesis of 3-methyl-4-nitro-5-nitroamino, 5-carboxamido, 5-*N,N*-dimethylamino and 5-β-hydroxyethylaminoisothiazole are here reported. The synthesis of 3-methyl-4-nitroso-5-ethylthioisothiazole (IX) is reported *via* an unusual reaction of 5-bromo-3-methyl-4-nitroisothiazole (I) and sodium ethyl mercaptide. 5-Bromo-4-nitroisothiazole was prepared by nitration of 5-bromoisothiazole. The nitro group was shown to be essential for antifungal activity.

J. Heterocyclic Chem., **17**, 385 (1980).

Due to the broad spectrum of pharmacological activity shown by certain nitrothiazoles (1-3), we chose to synthesize the closely related nitroisothiazoles to study their biological properties. Although several 4-nitroisothiazoles have been previously described (4-6), relatively little has been reported on their biological activity.

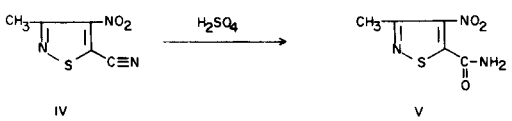
5-Bromo-3-methyl-4-nitroisothiazole (4), (I) was found in our Laboratory to possess broad spectrum antifungal activity *in vitro* (7). These results prompted a synthetic study of the 5-halogen and related 4-nitroisothiazoles.



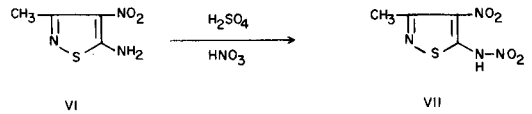
II III

The previously unknown 5-chloro-3-methyl-4-nitroisothiazole (III) was synthesized from 5-chloro-3-methylisothiazole (4) (II) and 90% nitric acid in concentrated sulfuric. III was found to possess antimycotic activity essentially equal to I.

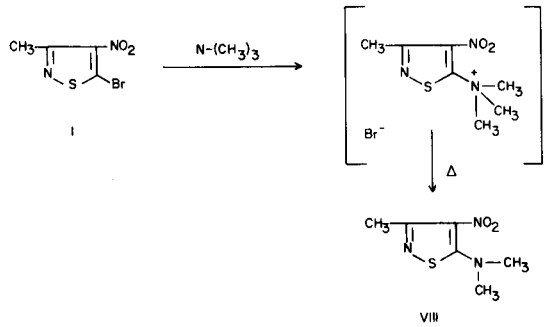
In order to evaluate the importance of the electron withdrawing 5-halogeno groups of these nitroisothiazoles in relation to their antifungal properties, we prepared other isothiazoles possessing electron withdrawing substituents in the 5-position. 5-Cyano-3-methyl-4-nitroisothiazole (6) (IV) was treated with concentrated sulfuric acid to yield the corresponding 3-methyl-4 nitroisothiazole-5-carboxamide (V). 5-Amino-3-methyl-4-nitroisothiazole (4) (VI) was nitrated to give 3-methyl-5-nitramino-4-nitroisothiazole (VII). An attempt to prepare 3-methyl-4-nitro-5-trimethylammonium bromide from I gave instead 5-*N,N*-dimethylamino-3-methyl-4-nitroisothiazole VIII, when the crude product was sublimed in an attempted effort at purification. Compounds IV, V and VIII were devoid of any significant antifungal activity. When compound I was treated with sodium ethyl mercaptide in refluxing ethanol, an unexpected 4-nitroso derivative (IX) was obtained. Elemental analysis indicated that the product



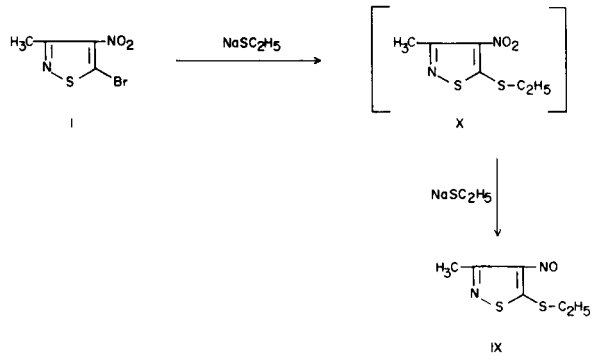
IV V



VI VII



I VIII



I X IX

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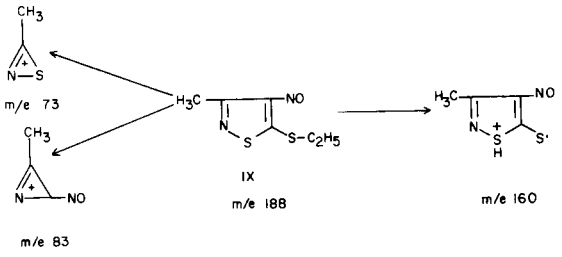
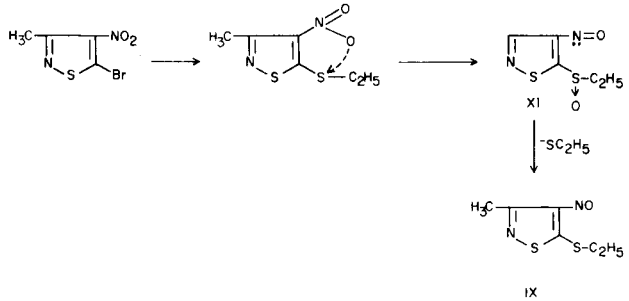
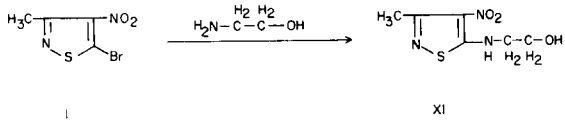
(IX) had the empirical formula $C_6H_8N_2OS_2$. The product IX, showed N=O stretching in the ir at 1550 cm^{-1} and also gave a positive nitroso test (7), which indicated that the product was 5-ethylthio-3-methyl-4-nitroisothiazole (IX). The mass spectrum of IX showed the molecular ion and the major fragments (m/e of 188, 160, 83, and 73) which confirm this structure.



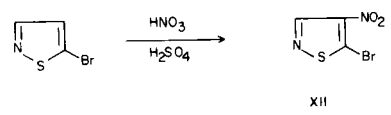
It is quite possible that an intramolecular oxidation reduction occurs with X to give the sulfoxide IX which then undergoes nucleophilic displacement of the sulfoxide group to give IX.



Further nucleophilic desplacement reactions of 5-bromo-3-methyl-4-nitroisothiazole (I) were studied. These reactions might be considered similar to the reactions of the 2-halo nitrothiazoles (3). Treatment of I with 2-aminoethanol gave 5-β-hydroxyethylamino-3-methyl-4-nitroisothiazole in good yield.



It is interesting to note that 4-amino-5-bromo-3-methylisothiazole (6) which might be considered a metabolic product of I, showed no antifungal properties *in vitro*. The effect upon antifungal activity of replacing the 3-methyl group of I with a hydrogen atom was next considered. This was accomplished by nitration of 5-bromoisothiazole (5) to give 5-bromo-4-nitroisothiazole (XII). 5-Bromo-4-nitroisothiazole possessed essentially the same antifungal spectrum *in vitro* as I.



Finally it is of interest to note that 4-nitroisothiazole (5) itself shows significant antifungal activity (7) although of a lower order than I or XII.

Thus the presence of the 4-nitro group is essential to antifungal activity which is also enhanced by a halogen at position 5.

EXPERIMENTAL (9)

5-Chloro-3-methyl-4-nitroisothiazole (III).

A mixture of 5-chloro-3-methylisothiazole (4) (0.90 g.), concentrated sulfuric acid (4 ml.), and 90% nitric acid (0.9 ml.) was heated at *ca.* 120° for 5 hours with stirring. After cooling to room temperature the reaction mixture was allowed to sit for 15 hours, then it was poured over ice. Filtration produced 0.70 g. (58%) of colorless solid, m.p. 54-6.5°. Recrystallization from absolute ethanol followed by sublimation produced an analytical sample, m.p. 57-58°

Anal. Calcd. for $C_4H_3ClN_2O_2S$: C, 26.87; H, 1.69; N, 15.69. Found: C, 27.21; H, 1.81; N, 15.38.

5-*N*,*N*-Dimethylamino-3-methyl-4-nitroisothiazole (VIII).

A mixture of 5-bromo-3-methyl-4-nitroisothiazole (4) (I, 1.0 g.) and trimethylamine (5 ml.) was allowed to stand for 30 minutes, then the excess amine was allowed to evaporate. The resulting yellow solid was sublimed to give a bright yellow solid, 0.7 g. (83%), m.p. 69-76°. Recrystallization of the yellow solid from petroleum ether (60-90°) gave an analytically pure sample, m.p. 79-80°.

Anal. Calcd. for $C_6H_9N_3O_2S$: C, 38.49; H, 4.84; N, 22.44. Found: C, 38.59; H, 5.04; N, 22.18.

3-Methyl-5-nitramino-4-nitroisothiazole (VII).

To concentrated sulfuric acid (4 ml.) cooled in an ice bath was slowly added 5-amino-3-methyl-4-nitroisothiazole (4) (VI 1.0 g.), followed by the slow addition of 90% nitric acid (1 ml.). The reaction mixture was then stirred for 25 hours at 30-35°, poured onto ice, and the colorless solid removed by filtration and dried, yield 1.2 g. (93%), m.p. 193° dec. Recrystallization from ethanol produced an analytically pure solid m.p. 193° dec.

Anal. Calcd. for $C_4H_4N_4O_4S$: C, 23.53; H, 1.97; N, 27.44. Found: C, 23.29; H, 1.78; N, 27.23.

5-Ethylthio-3-methyl-4-nitrosoisothiazole (IX).

A mixture of 5.0 g. (0.022 mole) of 5-bromo-3-methyl-4-nitroisothiazole (4) and 2.0 g. (0.024 mole) of sodium ethylmercaptide was heated in refluxing ethanol (100 ml.) for 2 hours. Cooling the solution in the freezer (-20°) overnight produced a yellow solid (m.p. 56-61°). Sublimation of this product produced 0.4 g. of a light yellow solid melting at 59-62.5°. Recrystallization of IX from ethanol-water followed by sublimation gave a light yellow solid, m.p. 60.5-2.5°; ir (potassium bromide): 1550 cm^{-1} (N=O); ms: (70 eV) m/e (relative intensity) 188 (29, M+), 160 (46), 83 (44), 73 (100).

Anal. Calcd. for $C_6H_8N_2OS_2$: C, 38.28; H, 4.28; N, 14.88; S, 30.21. Found: C, 38.22; H, 4.06; N, 15.09; S, 30.24.

5-β-Hydroxyethylamino-3-methyl-4-nitroisothiazole (XI).

A mixture of 5-bromo-3-methyl-4-nitroisothiazole (4) (I, 1.0 g.) and ethanolamine (10 ml.) was heated at 75° for 30 minutes and then stirred for 3 days at room temperature. This mixture was poured into a crystallizing dish and allowed to sit until yellow needles had formed. The solid was removed by filtration,

washed with water, and dried; m.p. 101.5-104°. Recrystallization from water produced 0.6 g. (69%) yellow solid, m.p. 102.4-105°

Anal. Calcd. for $C_6H_9N_3O_3S$: C, 35.46; H, 4.46; N, 20.67. Found: C, 35.49; H, 4.52; N, 20.48.

5-Bromo-4-nitroisothiazole (XII).

To 10 g. of redistilled 5-bromoisothiazole (5) was slowly (20 minutes) dropped a mixture of concentrated sulfuric acid (60 ml.) and 90% nitric acid (23 ml.), while the reaction was stirred and cooled in an ice bath. The temperature of the reaction mixture was slowly raised to 110° where it was maintained for 5 hours, then allowed to cool to room temperature and stirred for 16 hours. After pouring the reaction onto ice the resulting light yellow solid was collected, washed with water, and dried over sodium hydroxide under a slight vacuum. This product was then chromatographed on 300 g. silica gel. Elution with petroleum ether (60-90°) gave a small fore run which was discarded. This was followed by further elution with chloroform-petroleum ether (1:2) which gave the crude material 5-bromo-4-nitroisothiazole (m.p. 32-35°) which was further sublimed to give a pure product; 5.5 g., m.p. 37-38°.

Anal. Calcd. for $C_3HBrN_2O_2S$: C, 17.20; H, 0.48; N, 13.40. Found: C, 16.99; H, 0.66; N, 13.23.

REFERENCES AND NOTES

(1) G. Asato, G. Berkelhammer, and E. L. Moon, *J. Med. Chem.*, **12**, 374 (1969).

(2) E. F. Elslager, Abstracts of papers, 12th Nat. Med. Chem. Symp. Seattle, WA., June 22-25, 1970, p. 9a.

(3) A. Silberg, A. Ursu-Donea, and L. Gazakas, *Stud. Univ. Babes-Bolzai, Ser. Chem.*, **11**, 29 (1966), and references cited therein.

(4) A. Adams and R. Slack, *J. Chem. Soc.*, 3061 (1959).

(5) M. P. L. Caton, D. H. Jones, R. Slack and K. R. H. Wooldridge, *ibid.*, 446 (1964).

(6) A. Holland, R. Slack, R. F. Warren, and D. Buttimore, *ibid.*, 7277 (1965).

(7) D. E. O'Brien, R. K. Robins and A. H. Albert, U. S. Patent 3,840,665 (1974).

(8) F. Feigl, "Spot Tests in Organic Analysis", Elsevier Publishing Co.

(9) Melting points were taken on a Thomas-Hoover melting point apparatus and are uncorrected. Infrared and mass spectra were determined on a Perkin-Elmer 257 grating infrared spectrophotometer and on a Perkin-Elmer 270 double focusing mass spectrophotometer respectively. All samples displayed a single spot on thin layer chromatography. Elemental analyses were performed by the Heterocyclic Chemical Corporation of Harrisonville, Missouri and M-H-W Laboratories of Garden City, Michigan.