the control interferes with the determination. Some uninvestigated materials are eluted earlier than the esters of GGE and mephenesin. Quantitation is made comparing peak heights of the internal standard with that of the GGE ester.

A large amount of the ester was prepared by the procedure described to determine whether reaction had occurred with both or only one of the hydroxyl groups. NMR spectra showed that only the primary hydroxyl had reacted.

The results from the blood level determinations following the oral dose are shown in Fig. 2. GGE is readily absorbed, with the maximum amount determined occurring in the 0.25-hr. sample. The half-life was 1.00 hr. Detectable amounts of the drug were no longer present in the 8-hr. samples of any of the subjects, indicating rapid metabolism and excretion.

REFERENCES

(1) H. Hafner, *Wien. Med. Wochenschr.*, **114**, 523(1964).

(2) R. D. Eastham and E. P. Griffiths, *Lancet*, **7441**, 795(1966).

(3) J. L. Silverman and H. A. Wurzel, *Amer. J. Med. Sci.*, **254**, 491(1967).

(4) H. R. Baumgartner, M. Cronquist, P. Wobman, and F. Streuli, *Schweiz. Med. Swchr.*, **97**, 1674(1967).

(5) J. Franco and L. R. Grams, *Clin. Res.*, **16**, 41(1968).

(6) S. Naito and M. Mizutani, *Chem. Pharm. Bull.*, **17**, 822 (1969).

(7) A. M. Morgan, E. B. Truitt, Jr., and J. M. Little, *J. Amer. Pharm. Ass., Sci. Ed.*, **46**, 374(1957).

(8) M. Mizutani and S. Naito, *Chem. Pharm. Bull.*, **15**, 1422 (1967).

(9) B. R. Rades and E. S. Aranda, *J. Pharm. Sci.*, **57**, 847(1968).

(10) M. Koibuchi, T. Shibazaki, T. Minamakawa, and Y. Nishimura, *J. Pharm. Soc. Jap.*, **88**, 877(1968).

ACKNOWLEDGMENTS AND ADDRESSES

Received February 10, 1970, from the *Research Laboratories, A. H. Robins Company, Inc., Richmond, VA 23220*

Accepted for publication March 18, 1970.

The authors thank Dr. Russell C. Seckendorf, Clinical Pharmacology, A. H. Robins Company, for the administration of the drug and collection of blood samples.

Antiradiation Compounds XIV: Dithiocarbamates of Aminothiophenes

WILLIAM O. FOYE, JAMES MICKLES, and GERARD M. BOYCE

Abstract □ Dithiocarbamates of 2-amino-3-cyano(or carbethoxy)-4,5-dialkylthiophenes and a corresponding furan have been obtained. Dithiocarbamate formation of 2-amino-3-cyano-4,5-diphenylfuran resulted in a conversion to the corresponding thiophene dithiocarbamate. A dithiocarbamate trithiocarbonate of 2-amino-3-carbethoxy-4-mercaptomethylthiophene was also synthesized, and ring closure of the 2-amino-3-cyanothiophenes to thiopheno[2,3-*d*]pyrimidines was observed. None of the compounds tested showed radiation-protective or antimalarial properties.

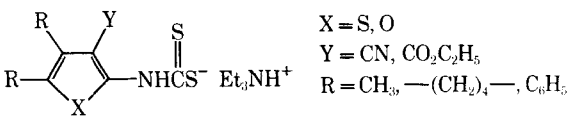
Keyphrases □ Antiradiation compounds—dithiocarbamates of aminothiophenes □ Aminothiophenes, dithiocarbamate derivatives—radiation-protective capacity, antimalarial properties □ IR spectrophotometry—structure

Although thiophene derivatives have not appeared frequently with radiation-protective properties, a basic derivative, *N*-phenyl-2-thiophenecarboxamidine, has been reported to have appreciable protection in rats (1). Since several heterocyclic dithiocarbamates in the pyridine, pyrimidine, and acridine series (2) are radiation protective, dithiocarbamates of thiophenes and furans having basic functions appeared to be logical candidates as radiation-protective compounds. Methods for obtaining thiophenes and furans having primary amino substituents in the ring have recently been announced (3), and the conversion of compounds of this type to dithiocarbamates has been attempted. Inclusion of this sulfur-containing function provides a thiol anion capable of undergoing rapid hydrogen-atom exchange reactions (4), which could account for radiation protection.

PROCEDURE

Preparation of 2-aminothiophenes was carried out by the method of Gewald *et al.* (5) with modifications. This procedure involved the base-catalyzed condensation of a carbonyl compound with an active methylene nitrile and sulfur. Using methyl ethyl ketone and malononitrile, the reaction was found to be best catalyzed with morpholine, with excess ketone as the solvent. Using methyl ethyl ketone and malononitrile, the product was 2-amino-3-cyano-4,5-dimethylthiophene; with ethanol as the solvent, the product was 2-butylidenemalononitrile. By the same procedure, but with ethanol as the solvent, the following were obtained: 2-amino-3-carbethoxy-4,5-dimethylthiophene, 2-amino-3-cyano-4,5-tetramethylenothiophene, and 2-amino-3-carbethoxy-4,5-tetramethylenothiophene. Also obtained by the same general procedure, without sulfur, were 2-amino-3-cyano-4,5-dimethylfuran and the corresponding 4,5-diphenyl compound.

Attempts to form the dithiocarbamates of the 2-aminothiophenes previously mentioned, using carbon disulfide and ethanol as the solvent, gave only small yields over a period of 24–72 hr. In the case of 2-amino-3-cyano-4,5-dimethylthiophene, the thiourea was formed instead. By using the procedure of Fairfull and Peak (6), however, triethylammonium salts of the dithiocarbamates (I) of the aminothiophenes and one of the aminofurans were obtained in good yield and sufficiently pure for analysis.



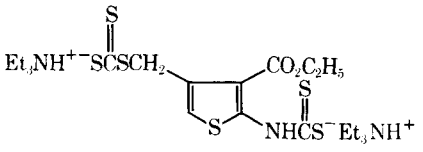
I

The attempted conversion of 2-amino-3-cyano-4,5-diphenylfuran to the dithiocarbamate gave a product having a poor analysis for the ethyl ester of the dithiocarbamate. By allowing the reaction to take place during a much longer time (2 weeks), a product was obtained for which the analysis indicated formation of the dithiocarbamate salt of 2-amino-3-cyano-4,5-diphenylthio-

phene (I, R = C_6H_5, Y = CN). During this prolonged reaction, the furan ring apparently opened and reacted with carbon disulfide to give the corresponding thiophene.

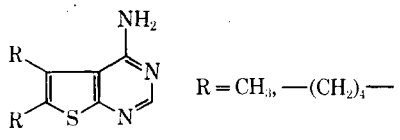
IR absorption of the dithiocarbamates containing cyano groups showed either very weak or no peaks for cyano absorption. This may be attributed to interaction with the adjacent, negatively charged dithiocarbamate group. In addition, absorption due to C=S occurred at lower wavenumbers than in the case of the 3-carbethoxy compounds, again indicating interaction with cyano groups.

2-Amino-3-ethoxycarbonyl-4-mercaptomethylthiophene was obtained by a modification of the method of Gewald and Schinke (7) for the disulfide. The mercaptan was not isolated but was treated with carbon disulfide and triethylamine to give the dithiocarbamate trithiocarbonate (II) from condensation at both the amino and thiol functions. A previous dithiocarbamate trithiocarbonate, prepared from cysteine, was found to be strongly radiation protective in mice (8). A similar attempt to obtain 2-amino-3-cyano-4-methyl-5-mercaptothiophene gave the sulfide.



II

Thiopheno[2,3-*d*]pyrimidines (III) were also obtained using the method of Gewald (9) for synthesis of furo[2,3-*d*]pyrimidines. These compounds resulted on refluxing the appropriate 3-cyanothiophene with formamide and acetic anhydride.



III

BIOLOGICAL TESTING RESULTS

Antiradiation screening of several of the compounds was carried out.[1] Tests were carried out in mice *versus* 825 r (X-rays) or 950 r (γ-rays) with an observation period of 30 days. The dithiocarbamate of 2-amino-3-carbethoxy-4,5-dimethylthiophene (I) was inactive against 950 r (γ-rays), and the dithiocarbamate trithiocarbonate (II) and the two thiopheno[2,3-*d*]-pyrimidines (III) were inactive against 825 r (X-rays).

The three compounds (II and III) were also screened for antimalarial activity in mice infected with *Plasmodium berghei* and in *Aedes aegypti* infected with *Plasmodium gallinaceum;* they were inactive in both tests. Also, the dithiocarbamates of the 2-amino-3-cyano(or carbethoxy)-4,5-dialkylthiophenes (I) were inactive in the mouse test.

It may be concluded from the lack of radiation-protective activity for the dithiocarbamate trithiocarbonate (II) reported here and the powerful protective effect of the corresponding cysteine derivative (8) that a rigid structure connecting these functions is detrimental to radiation protection.

EXPERIMENTAL

Analyses for carbon, hydrogen, and nitrogen were performed.[2] Sulfur analyses were done by Parr bomb peroxide fusion. Melting points were taken on a Mel-Temp apparatus and are corrected. IR absorption spectra were obtained with a Perkin-Elmer model 137B spectrometer.

Triethylammonium 3-Cyano-4,5-dimethylthiophene-2-dithiocarbamate—A mixture of 5.3 g. (0.035 mole) of 2-amino-3-cyano-4,5-dimethylthiophene (5), 42 ml. (0.600 mole) of carbon disulfide, 76.5 ml. (0.550 mole) of triethylamine, and 2.5 ml. of absolute ethanol formed a red-orange solution. The solution was stirred at room temperature for 48 hr. and yielded 3.3 g. (28%) of orange-yellow product; m.p. 241–242°; IR (KBr) 880 (β-ring), 1135 (C=S), 2200 (C≡N, weak) $cm.^{-1}$

Anal.—Calcd. for $C_{14}H_{23}N_3S_3$: C, 51.02; H, 7.03; N, 12.75; S, 29.19. Found: C, 51.37; H, 6.97; N, 12.29; S, 29.63.

Triethylammonium 3-Carbethoxy-4,5-dimethylthiophene-2-dithiocarbamate—A mixture of 7.0 g. (0.035 mole) of 2-amino-3-carbethoxy-4,5-dimethylthiophene (5), 42 ml. (0.600 mole) of carbon disulfide, 76.5 ml. (0.550 mole) of triethylamine, and 2.5 ml. of absolute ethanol formed a red-orange solution, which was allowed to stand at room temperature for 2 days, giving 7.5 g. (56%) of bright-yellow crystals; m.p. 105–107°; IR (KBr) 990 (C=S), 1650 (C=O) $cm.^{-1}$.

Anal.—Calcd. for $C_{16}H_{28}N_2O_2S_3$: C, 51.02; H, 7.49; N, 7.44; S, 25.54. Found: C, 50.86; H, 7.11; N, 7.15; S, 25.10.

Triethylammonium 3-Cyano-4,5-tetramethylenothiophene-2-dithiocarbamate—A mixture of 8.9 g. (0.05 mole) of 2-amino-3-cyano-4,5-tetramethylenothiophene (5), 60 ml. (0.86 mole) of carbon disulfide, 109 ml. (0.78 mole) of triethylamine, and 3.5 ml. of absolute ethanol formed a yellow solution, which was allowed to stand for 3 days at room temperature, giving 6.5 g. (36%) of rust-colored product; m.p. 210–212°; IR (KBr) 875 (β-ring), 1115 (C=S) $cm.^{-1}$.

Anal.—Calcd. for $C_{16}H_{25}N_3S_3$: C, 54.04; H, 7.09; N, 11.82; S, 27.05. Found: C, 54.11; H, 6.82; N, 11.70; S, 27.50.

Triethylammonium 3-Carbethoxy-4,5-tetramethylenothiophene-2-dithiocarbamate—A mixture of 11.25 g. (0.05 mole) of 2-amino-3-carbethoxy-4,5-tetramethylenothiophene (5), 60 ml. (0.86 mole) of carbon disulfide, 109 ml. (0.78 mole) of triethylamine, and 3.5 ml. of absolute ethanol formed a yellow solution, which was allowed to stand at room temperature for 3 days, giving 11.3 g. (56%) of bright-yellow product; m.p. 101–103°; IR (KBr) 965 (C=S), 1660 (C=O) $cm.^{-1}$.

Anal.—Calcd. for $C_{18}H_{30}N_2O_2S_3$: C, 53.69; H, 7.51; N, 6.96; S, 24.89. Found: C, 53.97; H, 7.36; N, 6.75; S, 24.76.

***N,N'*-Bis(3-cyano-4,5-dimethyl-2-thienyl)thiourea**—A solution of 2-amino-3-cyano-4,5-dimethylthiophene (5) (3.04 g., 0.02 mole) in 50 ml. of absolute ethanol was treated with carbon disulfide (42 ml., 0.06 mole). The yellow solution was stirred at room temperature for 2 days, and 0.95 g. (27%) of yellow compound was obtained which did not melt below 300°; IR (KBr) 1290 (C=S, thioamide), 2200 (C≡N) $cm.^{-1}$.

Anal.—Calcd. for $C_{15}H_{14}N_4S_3$: C, 52.0; H, 4.1; S, 27.7. Found: C, 51.7; H, 4.5; S, 27.5.

Triethylammonium 3-Cyano-4,5-dimethylfuran-2-dithiocarbamate—A solution of 3.1 g. (0.02 mole) of 2-amino-3-cyano-4,5-dimethylfuran (9), 60 ml. (0.44 mole) of triethylamine, and 32.5 ml. (0.44 mole) of carbon disulfide was stirred at room temperature for 2 weeks. The orange solid was triturated with absolute ethanol, giving 0.64 g. (9%) of compound which slowly decomposed on standing; m.p. 195–200° dec.; IR (KBr) 880 (β-ring), 1145 (C=S) $cm.^{-1}$.

Anal.—Calcd. for $C_{14}H_{23}N_3OS_2$: C, 53.7; H, 7.3; N, 13.3; S, 20.4. Found: C, 53.3; H, 7.4; N, 12.7; S, 20.5.

Triethylammonium 3-Cyano-4,5-diphenylthiophene-2-dithiocarbamate—A solution of 2.28 g. (0.009 mole) of 2-amino-3-cyano-4,5-diphenylfuran (9), 10.5 ml. (0.150 mole) of carbon disulfide, 19.1 ml. (0.138 mole) of triethylamine, and 6 ml. of absolute ethanol was allowed to stand at room temperature for 2 weeks. A yield of 0.61 g. (15%) of orange crystals was obtained; m.p. 256–258°; IR (KBr) 987 (C=S), 2050 (C≡N) $cm.^{-1}$.

Anal.—Calcd. for $C_{24}H_{27}N_3S_3$: C, 63.5; H, 6.0; N, 9.2; S, 21.2. Found: C, 63.4; H, 5.8; N, 9.0; S, 20.7.

Bis(triethylammonium)-3-carbethoxy-2-dithiocarbamate-4-thienyltrithiocarbonate—A solution of 7.3 g. (0.06 mole) of 1,3-dimercaptoacetone and 6.8 g. (0.06 mole) of ethyl cyanoacetate in 30 ml. of absolute ethanol was treated with 3 ml. of triethylamine. After 15 min., 35 ml. (0.47 mole) of carbon disulfide and 45 ml. (0.30 mole) of triethylamine were added; the solution was stirred for 1 hr. and allowed to stand overnight at room temperature. A red oil was separated and crystallized by the addition of 200 ml. of absolute ethanol. After being stirred for 2 hr., the yellow product (10.8 g., 31%) was isolated; m.p. 106–108°; IR (KBr) 1010 (C=S), 1125 (C=S), 1670 (C=O) $cm.^{-1}$.

[1] At the Walter Reed Army Institute of Research; results reported through the courtesy of Dr. D. P. Jacobus.

[2] By Weiler and Strauss, Oxford, England, or by Carol Fitz, Needham, Mass.

Anal.—Calcd. for $C_{22}H_{41}N_3O_3S_6$: C, 46.20; H, 7.23; N, 7.35; S, 33.64. Found: C, 45.97; H, 7.05; N, 7.05; S, 33.62.

Bis(2-amino-3-cyano-4-methyl-5-thienyl)sulfide—A mixture of 58 g. (1 mole) of acetone, 13.2 g. (0.2 mole) of malononitrile, and 6.4 g. (0.2 g. atom) of sulfur was treated dropwise with 20 ml. of triethylamine. The mixture was stirred at 30–35° for 7 hr. and allowed to stand at room temperature for 36 hr. After addition of 400 ml. of aqueous ethanol (1:1) and vigorous stirring, a tan, crystalline product was obtained which was extracted with boiling ethanol, giving 1.85 g. (3%) of product; m.p. 255–257°.

Anal.—Calcd. for $C_{12}H_{10}N_4S_3$: C, 47.03; H, 3.29; N, 18.28; S, 31.39. Found: C, 47.14; H, 3.38; N, 18.15; S, 31.28.

4-Amino-5,6-dimethylthiopheno[2,3-*d*]pyrimidine—A mixture of 3.04 g. (0.02 mole) of 2-amino-3-cyano-4,5-dimethylthiophene (5), 30 ml. of formamide, and two drops of acetic anhydride was refluxed at 160–165° for 2 hr. After being cooled, a solid product was isolated and recrystallized from dioxane, giving 1.48 g. (39%) of white crystals; m.p. 261–263°; IR (KBr) 1650 (NH_2), 3400 (NH_2) $cm.^{-1}$.

Anal.—Calcd. for $C_8H_9N_3S$: C, 53.47; H, 5.21; N, 23.22; S, 18.25. Found: C, 53.61; H, 5.01; N, 23.44; S, 17.94.

4-Amino-5,6-tetramethylenothiopheno[2,3-*d*]pyrimidine—A mixture of 3.56 g. (0.02 mole) of 2-amino-3-cyano-4,5-tetramethylenothiophene (5), 30 ml. of formamide, and two drops of acetic anhydride was refluxed at 165–170° for 2 hr. After being cooled, a solid product was isolated and recrystallized from dioxane, giving 0.7 g. (17%) of white product; m.p. 261–263°; IR (KBr) 1635 (NH_2), 3350 (NH_2) $cm.^{-1}$.

Anal.—Calcd. for $C_{10}H_{11}N_3S$: C, 58.23; H, 6.32; N, 21.15; S, 15.62. Found: C, 58.51; H, 5.94; N, 20.67; S, 16.01.

REFERENCES

(1) I. Baev and S. Robev, *C. R. Acad. Bulg. Sci.*, **13**, 733(1960).

(2) W. O. Foye and D. H. Kay, *J. Pharm. Sci.*, **57**, 345(1968); W. O. Foye, D. H. Kay, and P. R. Amin, *ibid.*, **57**, 1793(1968).

(3) K. Gewald, *Chem. Ber.*, **98**, 3571(1965); V. I. Shvedov, V. K. Ryzhkova, and A. N. Grimev, *Khim. Getero. Soedin.*, **1967**, 239.

(4) S. G. Cohen, in "Organosulfur Chemistry," M. J. Janssen, Ed., Interscience, New York, N. Y., 1967, chap. 3.

(5) K. Gewald, E. Schinke, and H. Bottcher, *Chem. Ber.*, **99**, 94 (1966).

(6) A. E. S. Fairfull and D. A. Peak, *J. Chem. Soc.*, **1955**, 796.

(7) K. Gewald and E. Schinke, *Chem. Ber.*, **99**, 2712(1966).

(8) R. I. H. Wang, W. Dooley, Jr., W. O. Foye, and J. Mickles, *J. Med. Chem.*, **9**, 394(1966).

(9) K. Gewald, *Chem. Ber.*, **99**, 1002(1966).

ACKNOWLEDGMENTS AND ADDRESSES

Received January 1, 1970, from the *Department of Chemistry, Massachusetts College of Pharmacy, Boston, MA 02115*

Accepted for publication March 18, 1970.

Presented to the Medicinal Chemistry Section, APhA Academy of Pharmaceutical Sciences, Washington, D. C. meeting, April 1970.

This investigation was supported by Research Grant RH00297 from the National Center for Radiological Health, U. S. Public Health Service, Bethesda, Md.

N-Substituted Aminoethanethiols and *N*-Substituted Aminoethanethiol *S*-Sulfonic Acids as Radioprotective Agents

F. I. CARROLL and MONROE E. WALL

Abstract □ Several *N*-substituted aminoethanethiols and *N*-substituted aminoethanethiol *S*-sulfonic acids were tested as potential radioprotective agents. 2-(2′-Carbamidoethylamino)-ethanethiol (I*a*), 2-(2′-carboxyethylamino)-ethanethiol (I*b*), 2-(2′-cyanoethylamino)-ethanethiol (I*c*), and 2-(2′-carbamidoethylamino)-ethanethiol *S*-sulfonic acid (II*a*) exhibited significant protective effects against ionizing radiation. Compound I*a* showed the highest activity and was selected for further radiation-protection test studies. The structure–activity relationships of this class of compounds are discussed.

Keyphrases □ Aminoethanethiols, *N*-substituted—radioprotective capacity, structure–activity relationships □ Aminoethanethiol *S*-sulfonic acids, *N*-substituted—radioprotective capacity, structure–activity relationships □ Radioprotective agents—aminoethanethiols, aminoethanethiol *S*-sulfonic acids, *N*-substituted

A recent list of the various types of compounds that show radioprotective properties has appeared and their structure–activity relationships have been discussed (1). Aminoalkylthiols constitute the most effective class of radioprotective agents. The initial discovery that 2-mercaptoethylamine (MEA) offered protection to mice against ionizing radiation (2) led to the synthesis of several hundred derivatives of this compound. Structural requirements necessary for radioprotective activity have evolved from the test results on these compounds and have been summarized (1). This effect was not observable when one or two alkyl substituents were placed on the carbon containing the thiol function of MEA (3, 4).[1] Subsequently, it was found that some *N*-substituted aminoethanethiols and *N*-substituted aminoethanethiol *S*-sulfonic acids, prepared in this laboratory, showed significant protection against ionizing radiation. In this report the radioprotection test results on these compounds are presented, and their structure–activity relationships are discussed.

[1] Subsequent antiradiation test results have shown that 2-mercapto-2-methylaminopropane hydrochloride, when administered at 90 mg./kg. i.p. using CMCTW as vehicle, gave 67% survival to mice irradiated with 825 r. (See footnotes to Table II for explanation of test data.)