acid chloride which was again diluted (CH_2Cl_2) and added dropwise to a stirred mixture of 15.7 g (0.1 mole) of 5-nitro-2-furaldehyde hydrazone[15] in 160 ml of dry pyridine cooled below 15°. After completing the addition, stirring was continued 1.5 hr and then the brown solid was collected on a filter and washed (dilute AcOH). The crude product was crystallized from AcOH (see Table I).

The following compounds were prepared similarly: IV, V, XIV, XV, and XXI except that bis(2-methoxyethyl) ether replaced CH_2Cl_2 in preparing IV. The hydrazones used for the last three nitrofurans were 5-nitro-2-furaldehyde methyl hydrazone, 5-nitro-2-furaldehyde hydroxyethyl hydrazone,[16] and 5-nitro-2-thiophenecarboxaldehyde hydrazone, respectively. The preparation of the latter derivative and others not previously described in the literature is presented as follows.

2,6-Dihydroxy-3,5-dinitrobenzoic Acid.—To a mixture of 15.4 g (0.1 mole) of finely divided 2,6-dihydroxybenzoic acid and 40 ml of concentrated H_2SO_4 cooled below 10° was added dropwise with stirring at −5 to 5° about three-fifths of a cold mixture containing 19 g (0.21 mole) of HNO_3 (70%) and 10 ml of concentrated H_2SO_4. The remainder of the mixed acid was added at 5–10°. The mixture was poured on ice after stirring at room temperature 1 hr. The solid was filtered and sucked as free of liquid as possible before drying *in vacuo* at 60° to constant weight. The yield of crude pink acid was 21 g (86%), mp 145–174° dec. The acid was exceedingly water soluble and an analytical sample was prepared by crystallizing twice from AcOH and drying *in vacuo* at 100°: pale yellow solid, mp 186.5–188°. *Anal.* ($C_7H_4N_2O_8$) C, H, N.

Methyl 2,6-Dihydroxy-3,5-dinitrobenzoate.—A solution of 21 g (*ca.* 0.086 mole) of the crude acid (mp 145–174° dec) was refluxed with 100 ml of $SOCl_2$ for 4.5 hr and then most of the excess $SOCl_2$ was distilled *in vacuo*. MeOH (250 ml) was added to the acid chloride, and the solution was refluxed for 1 hr and then concentrated until the ester started to precipitate (final volume ~125 ml). The solution was cooled near 0° overnight, and the crystals were collected by filtration and dried at 85°. Glistening white plates were obtained that weighed 14 g (*ca.* 63%), mp 135–139°. A purified sample was prepared by crystallizing twice (MeOH): pale cream needles, mp 139–140.5°. *Anal.* ($C_8H_6N_2O_8$) C, H, N.

2,4-Dihydroxy-3,5-dinitrobenzoic Acid.—The nitration of β-resorcylic acid to prepare the dinitro acid was unsuccessful in our hands and the following procedure was found to be more expedient than reported methods.[17] A mixture of 50 g (0.178 mole) of 2,4-dichloro-3,5-dinitrobenzoic acid[18] and 1000 ml of 10% NaOH was heated on a steam bath for 7 hr and then cooled near 0°. The Na salt was collected on a filter, dissolved in water, and acidified with HCl. On recrystallizing (charcoal) the solid from H_2O and drying at 115° there was obtained 28 g (64.4%) of white acid, mp 200–203°.

Methyl 2,4-Dihydroxy-3,5-dinitrobenzoate.—The methyl ester was prepared from 22 g (0.09 mole) of the acid *via* the acid chloride as described above for methyl 2,6-dihydroxy-3,5-dinitrobenzoate. The yield of white ester was 15 g (60%), mp 196–200°. A purified sample was obtained by crystallizing (MeOH): pale yellow crystals, mp 197–200°. *Anal.* ($C_8H_6N_2O_8$) C, H, N.

Acetyl-3,5-dinitrosalicylic Acid (5-Nitrofurfurylidene)-N-acetylhydrazide (XVI).—A solution of 5.0 g (0.014 mole) of I in 25 ml of Ac_2O–AcOH (4:1) was obtained by warming and after 15 min it was cooled to 25° and an equal volume of H_2O was slowly added with swirling. Then the diacetyl derivative was crystallized by chilling near 0°. The pale yellow solid was filtered and dried at 85° to give 3.0 g (49%), mp 159–163°. A purified sample of pale yellow square tablets (see Table I) was prepared by crystallization (charcoal) twice (Me_2CO–H_2O).

5-Nitro-2-thiophenecarboxaldehyde Hydrazone.—5-Nitro-2-thiophenemethanediol diacetate[19] (10 g, 0.0386 mole) was dissolved in 225 ml of dry MeOH and cooled to 5°. A solution of 5.8 g (0.116 mole) of hydrazine hydrate and 15 ml of MeOH was slowly added to this solution. A red solid precipitated in a few minutes. The MeOH was allowed to evaporate at room temperature after stirring for 2 hr below 5°. The solids were crystallized (charcoal) from EtOH–H_2O to yield 4.0 g (61%) of red solid, mp 140–142°. An analytical sample was prepared by crystallization from this solvent pair: red solid, mp 142–144°. *Anal.* ($C_5H_5N_3O_2S$) C, H, N.

Acknowledgments.—The authors wish to thank Mr. E. L. Brunsting and Mr. J. W. Gruber for assistance with the syntheses and Mr. M. R. Carr and Miss C. A. Page for conducting the elemental analyses. The pharmacology data were furnished by Dr. H. W. Reuber, and Mr. D. C. Leggett determined the pK_a of I.

(15) Imperial Chemical Industries Ltd. (by Roy Hull), British Patent 816,886 (July 22, 1959).

(16) J. C. Howard, G. Gever, and P. H. I. Wei, *J. Org. Chem.*, **28**, 868 (1963).

(17) H. Einbeck and L. Jablonski, *Ber.*, **56**, 1907 (1923).

(18) W. Borsche and H. Fabr, *Ann. Chem.*, **402**, 91 (1913).

(19) C. Yuan, C. Yao, K. Su, and F. Yang, *Yao Hsueh Hsueh Pao*, **7**, 245 (1959); *Chem. Abstr.*, **54**, 12097*h* (1960).

Nitroheterocyclic Antimicrobial Agents. I. Nitrothiazolecarboxaldehyde Derivatives

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A series of derivatives of 2-thiazolecarboxaldehyde, 2-nitro-5-thiazolecarboxaldehyde, 5-nitro-2-thiazolecarboxaldehyde, and 4-nitro-2-thiazolecarboxaldehyde was synthesized and assayed for antimicrobial activity. Only 2,5-disubstituted thiazoles in which one substituent is a nitro group were active against microorganisms *in vitro*. 1-{[(5-Nitro-2-thiazolyl)methylene]amino}-2-imidazolidinone exhibited activity against *Staphylococcus aureus* and *Escherichia coli* infections in mice.

The disclosure of the antibacterial activity of 5-nitrofurfural derivatives by Dodd and Stillman[2] has spurred the syntheses of a large number of new nitrofuryl compounds.[3,4] In the majority of these compounds a nitro group in position 5 is necessary and a conjugated –C=N moiety in position 2 is desirable for antimicrobial activity. The search for new antimicrobial agents with the nitrofuran moiety replaced by nitropyrrole[5,6] and nitrothiophene[5,7–9] has been pursued, and, although interesting antimicrobial activities were reported in some cases, no clinically useful drug has yet emerged.

(1) Author to whom inquiries should be addressed.

(2) M. C. Dodd and W. B. Stillman, *J. Pharmacol. Exp. Ther.*, **82**, 11 (1944).

(3) H. E. Paul and M. F. Paul in "Experimental Chemotherapy," Vol. II, Part I, R. J. Schnitzer and F. Hawking, Ed., Academic Press Inc., New York, N. Y., 1964, pp 307–370.

(4) K. Miura and H. K. Reekendorf, *Progr. Med. Chem.*, **5**, 320 (1967).

(5) Reference 3, p 316.

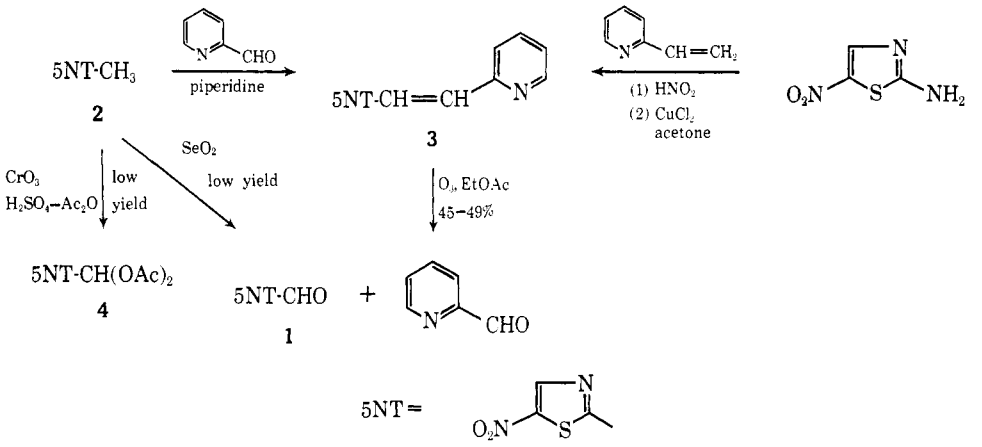
(6) W. T. Colwell, J. H. Lange, and D. W. Henry, *J. Med. Chem.*, **11**, 282 (1968), and references cited therein.

(7) G. L. Dunn, P. Actor, and V. J. DiPasquo, *ibid.*, **9**, 751 (1966).

(8) T. Kametani, O. Umezawa, M. Sato, and F. Kobayashi, *J. Pharm. Soc. Jap.*, **83**, 174 (1963).

(9) Smith Kline and French, South African Patent 65/5968 (1966).

SCHEME I



In this laboratory, it was decided to substitute some hitherto unknown isosteric nitroheteroaromatic groups for the nitrofuran ring in compounds known to be active antiinfective agents. After this work was begun, derivatives of nitroisoxazolecarboxaldehyde,[10] nitropyrazolecarboxaldehyde,[10] nitrobenzothiazolecarboxaldehyde,[11] and 1-methyl-5-nitro-2-imidazolecarboxaldehyde[12] and their antibacterial activities were reported. In general, the activities of these compounds were inferior to those reported for nitrofurans.

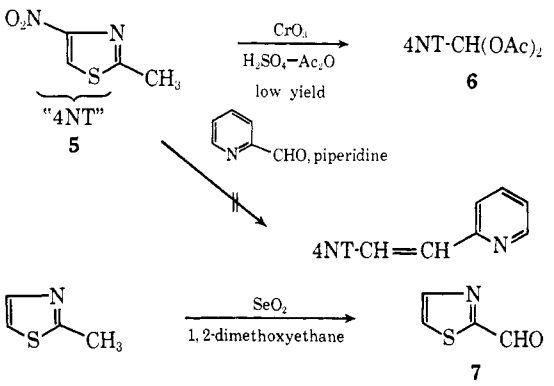
We now report the syntheses and antimicrobial activities of a series of derivatives of 2-nitro-5-thiazolecarboxaldehyde and 5-nitro-2-thiazolecarboxaldehyde.[13] For purposes of structure–activity correlation, derivatives of 4-nitro-2-thiazolecarboxaldehyde and 2-thiazolecarboxaldehyde were also synthesized and tested.

Chemistry.—The primary precursor chosen for the synthesis of 5-nitro-2-thiazolecarboxaldehyde (**1**) was 2-methyl-5-nitrothiazole[14] (**2**) (Scheme I). It was condensed with 2-pyridinecarboxaldehyde in *n*-PrOH in the presence of piperidine to give **3**,[14] which was also obtained by the Meerwein reaction of 2-amino-5-nitrothiazole with 2-vinylpyridine.[14] The olefin **3** was then ozonized in ethyl acetate at *ca.* −40° to give **1**, in 42–51% yield, and 2-pyridinecarboxaldehyde, which was removed from the reaction mixture by extraction with acid. The pure aldehyde **1** melted at 50.5°. SeO_2 oxidation of **2** also afforded **1**, while chromic acid led to the diacetate **4** of **1**, both in trace amounts.

Chromic acid oxidation of 2-methyl-4-nitrothiazole[14] (**5**) also gave the aldehyde diacetate **6** in trace amounts (Scheme II). On the other hand, SeO_2 oxidation of **5** failed. Attempts to condense **5** with 2-pyridinecarboxaldehyde in the presence of piperidine in *n*-PrOH to give the 2-pyridylvinyl compound for subsequent ozonation also failed. SeO_2 oxidation of 2-methylthiazole led to a low yield of 2-thiazolecarboxaldehyde (**7**).[15]

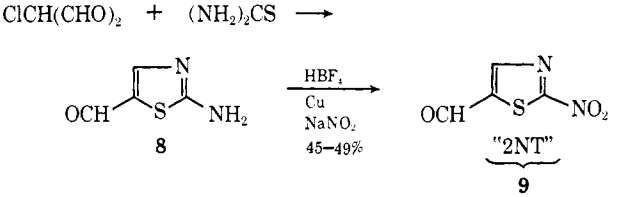
Since the synthesis of 2-amino-5-thiazolecarboxaldehyde (**8**) had been reported[16] previously *via* the reaction of chloromalondialdehyde with thiourea, the synthesis

SCHME II



of 2-nitro-5-thiazolecarboxaldehyde (**9**) was straightforward (Scheme III). Thus, **8** was diazotized in HBF_4 and the diazonium salt was allowed to react with excess NO_2^- in the presence of Cu to afford 45–49% crude yields of **9**. The nmr spectrum of **9** revealed two downfield protons at τ −0.22 (CHO) and 1.30 (ring H), which supported its structure.

SCHEME III

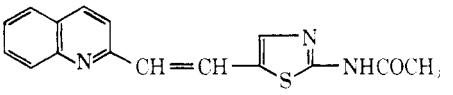


A survey of the known active nitrofurans led us to choose azomethine, β-arylvinyl, and heterocyclic groups for attachment to the 2 and 5 positions of the thiazole ring. The azomethine compounds were prepared readily by standard techniques from the aldehydes. In addition, the oxime **10** was prepared by the reaction of 2-methyl-5-nitrothiazole with BuONO and EtOH–HCl.

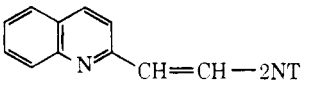
The β-arylvinyl compounds in the 5-nitrothiazole series were prepared by the condensation of 2-methyl-5-nitrothiazole with aromatic aldehydes. In contrast, attempts to condense 2-nitro-5-thiazolecarboxaldehyde with 2-picoline, 2-picoline N-oxide, and quinaldine were unsuccessful, the aldehyde being unstable in hot AcOH or Ac_2O reaction mixtures. In these solvents evolution of a brown gas (NO_2?) was observed during heating and no nitro compounds were isolated after work-up,

(10) C. Caradonna and M. L. Stein, *Ann. Chim.* (Rome), **54**, 539 (1964).
(11) R. G. Johnston and D. Kidd, *J. Chem. Soc.*, 4730, 4734 (1964).
(12) Merck and Co., Inc., Netherlands Patent 6,503,442 (1965). A communication describing our work in this series has been submitted elsewhere.
(13) D. W. Henry [*J. Med. Chem.*, **12**, 303 (1969)] also describes the synthesis of 5-nitro-2-thiazolecarboxaldehyde and some of its derivatives.
(14) G. Asato, *J. Org Chem.*, **33**, 2544 (1968).
(15) After this work was completed, P. E. Iversen and H. Lund [*Acta Chem. Scand.*, **20**, 2649 (1966)] reported a better synthesis of this aldehyde.
(16) Farbenfabriken Bayer A. G., German Patent 1,182,234 (1964).

except in the case of 2-quinaldine where a trace of likely product was isolated from an AcOH–Ac_2O mixture. Alternatively, 2-acetamido-5-thiazolecarboxaldehyde was condensed with quinaldine in refluxing Ac_2O to afford a 61% yield of **15**. This compound, however, after acid hydrolysis, followed by diazotization and displacement with NO_2^-, led at most to traces of **16**.



15



16

The nitrothiazolyl heterocyclic derivatives were represented by the aminothiadiazoles, which were derived from the thiosemicarbazones by oxidative cyclizations with Fe^{3+}.

Biological Activity.—The *in vitro* antibacterial activity of most of the nitrothiazolyl compounds is summarized in Table I. In general, the nitrothiazole derivatives have fair-to-good broad-spectrum *in vitro* activity. In these tests the response of the selected organisms to furazolidone (F) was readily demonstrated. The nitrothiazoles were tested *in vivo* orally against *Salmonella gallinarum* in chicks and *Staphylococcus aureus* (Smith), *Escherichia coli*, and *Mycobacterium tuberculosis* in mice. In addition, compounds **18**, **20**, **22**, **26**, and **27** were assayed orally against *Pasteurella multocida* in mice. The aminoimidazolidinone derivative **21** was the only *in vivo* antibacterially active compound. Against an *S. aureus* infection in mice a single dose of **21** saved 4/5 when given by oral tubing at 128 mg/kg. Sulfadiazine at 64 mg/kg saved 5/5 and there was 0/10 survival in the infected, untreated controls. Against a fatal *E. coli* 311 infection, **21** resulted in 5/5 and 3/5 survival at 512 and 128 mg/kg, respectively (oral tubing). The styryl compound **13** was active against *Trichomonas vaginalis* in mice at 0.05% in the diet, but inactive at 0.025%. Against selected fungi, some of the derivatives displayed broad *in vitro* activity (Table II), but, when one of the more active compounds, **3**, was tested dermally as a 1% ointment against *Trichophyton mentagrophytes* on guinea pigs, it was inactive.

The presence of the nitro group, as well as its position, is critical in this series as it is in the nitrofurans. Both the 2-nitro-5-thiazolyl and 5-nitro-2-thiazolyl analogs of furazolidone, **17** and **24**, respectively, were active *in vitro* against bacterial microorganisms, whereas the 4-nitro-2-thiazolyl analog **30** and desnitro derivative **29** were essentially inactive in the same test.

Experimental Section

Melting points were determined on a Thomas–Hoover apparatus and are uncorrected. Ir spectra were taken on a Perkin-Elmer Model 137 spectrophotometer, nmr spectra on a Varian A-60 instrument (Me_4Si). Microanalyses were performed by Galbraith Laboratories, Inc., Knoxville, Tenn. Where analyses are indicated only by symbols of the elements, analytical results obtained for the elements were within ±0.4% of the theoretical values.

5-Nitro-2-thiazolecarboxaldehyde (1).—A suspension of 12 g (51.5 mmoles) of 2-[2-(5-nitro-2-thiazolyl)vinyl]pyridine[14] in 450 ml of EtOAc was stirred at −30 to −20°, and O_3 (generated from a Welsbach Corp. ozonator) was bubbled into the mixture at *ca.* 0.081 mole hr until the yellow color disappeared (1.5 hr). Stirring was continued for an additional 10 min, the system then was purged of excess O_3 with N_2, and the mixture was treated with 9 g of NaI in 10 ml of glacial HOAc and 100 ml of H_2O at −10 to 10°. The I_2 was reduced with 19 g of $Na_2S_2O_3$ in 200 ml of H_2O, the organic phase was separated, and the aqueous layer was extracted with 100 ml of Et_2O. The combined organic phases were washed three times with 50 ml of 10% HCl, followed by a washing with 50 ml of saturated aqueous Na_2CO_3. The organic phase was then dried ($MgSO_4$) and filtered, and the volatile solvents were removed *in vacuo* to afford a red-brown oil. The oil was further swirled with ether, filtered to remove insoluble contaminants, and evaporated *in vacuo* to give 4.16 g (51%) of oily product. Analysis by glpc (20% SE 30 6-ft column at 175°) showed that this aldehyde was *ca.* 96% pure. A sample collected from the column melted at 50.5° (softened at 48°). *Anal.* ($C_4H_2N_2O_3S$) C, H, N, S.

5-Nitro-2-thiazolecarboxaldehyde Derivatives.—Unless the methods are described below, standard techniques or methods cited were used. The corresponding 2-nitro derivatives were also similarly prepared.

3-{[(5-Nitro-2-thiazolyl)methylene]amino}-2-oxazolidinone (17).—The method of Gever[17] was used to prepare 3-amino-2-oxazolidinone; 86.4% crude yield for **17**, recrystallized from MeOH, yellow crystals, mp 221–222°. *Anal.* ($C_7H_6N_4SO_4$) C, H, N, S.

5-Nitro-2-thiazolecarboxaldehyde semicarbazone (18): 85% crude yield, recrystallized from EtOH, yellow-brown crystals, mp 274° dec. *Anal.* ($C_5H_5N_5SO_3$) C, H, N, S.

5-Nitro-2-thiazolecarboxaldehyde thiosemicarbazone (19): 94% crude yield, recrystallized from EtOH–DMF, orange crystals, mp 250–251°. *Anal.* ($C_5H_5N_5O_2S_2$) C, H, N, S.

1-{[(5-Nitro-2-thiazolyl)methylene]amino}hydantoin (20): 35.5% crude yield, recrystallized from aqueous EtOH, gray crystals, mp 244–245°. *Anal.* ($C_7H_5N_5O_4S$) C, H, N, S.

1-{[(5-Nitro-2-thiazolyl)methylene]amino}-2-imidazolidinone (21).—The method of Michels and Gever[18] was used; 65% yield (after purification), recrystallized from DMF, yellow crystals, mp 256–257°. *Anal.* ($C_7H_7N_5O_3S$) C, H, N, S.

5-Nitro-2-thiazolecarboxaldehyde Oxime (10). Method A.[19]—A mixture of 1.16 g (8 mmoles) of 2-methyl-5-nitrothiazole, 0.9 g of 36% ethanolic HCl, and 1 g of BuONO was heated under reflux in 10 ml of EtOH. After 1.75 hr an additional 0.5 g of BuONO and 0.5 g of ethanolic HCl was added and refluxing continued for a total of 3.75 hr. The solution was cooled and evaporated *in vacuo* to give a red-brown oil. Ether was added to the oil, the mixture was filtered, and the ether filtrate was decolorized with activated carbon. The ether solution was evaporated to give an oil, which was then washed with petroleum ether. The insoluble material was dissolved in hot H_2O, filtered, and cooled to afford 0.2 g (15%) of the oxime which gradually turned from white to pale brown in about 1 day; mp 149.5–151°. Recrystallizations (H_2O) gave an analytical sample, mp 167–169° dec. *Anal.* ($C_4H_3N_3O_3S$) C, H, S.

Method B.—The oxime was prepared from the aldehyde in EtOH with $NH_2OH \cdot HCl$ and pyridine and the crude material was isolated by evaporating the reaction mixture to dryness *in vacuo*. It was purified by recrystallization from 50% aqueous EtOH, to which a trace of Me_2CO was added to solubilize the solid, to give a 50% yield of product, mp 169–170°.

4-[2-(5-Nitro-2-thiazolyl)vinyl]pyridine (11).—This product was prepared from 4-pyridinecarboxaldehyde in the same manner as 2-[2-(5-nitro-2-thiazolyl)vinyl]pyridine was prepared[14] from 2-methyl-5-nitrothiazole and 2-pyridinecarboxaldehyde; 8% crude yield, recrystallized from MeOH, yellow needles, mp 167–168°. *Anal.* ($C_{10}H_7N_3SO_2$) C, H, N, S.

2-[2-(5-Nitro-2-thiazolyl)vinyl]quinoline (12).—A mixture of 1.44 g (0.01 mole) of 2-methyl-5-nitrothiazole, 3.14 g (0.02 mole) of recrystallized 2-quinolinecarboxaldehyde and 5 drops of piperidine in 10 ml of *n*-PrOH was refluxed for 1 hr and then cooled in ice to give a dark green solid (750 mg), which was collected; mp 190–205°. Recrystallization from EtOH–Me_2CO gave 325 mg of yellow crystals, mp 205–207°. Further recrystallization

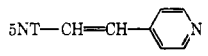
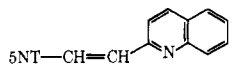
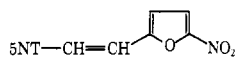
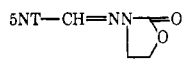
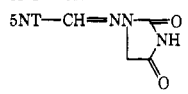
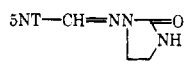
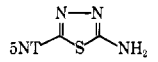
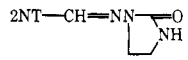
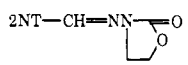
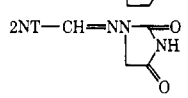
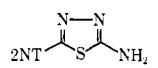
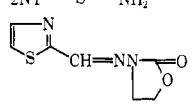
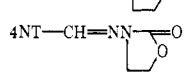
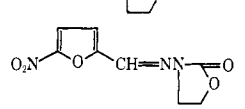
(17) G. Gever, U. S. Patent 2,652,402 (1953).

(18) J. G. Michels and G. Gever, *J. Amer. Chem. Soc.*, **78**, 5349 (1956).

(19) H. Bredereck and G. Simchen, *Angew. Chem. Intern. Ed. Engl.*, **2**, 738 (1963).

TABLE I

In Vitro ACTIVITY OF NITROTHIAZOLES AGAINST SELECTED ORGANISMS[a]

No.	Structure	Min inhib concn, μg/ml[b]																
		B.c.	B.s.	B.t.	M.	S.a.	St.a.	S.f.	A.a.	A.f.	B.b.	E.c.	P.m.	S.c.	S.d.	S.g.	S.t.	S.ty.
1		125	31	—	31	—	31	—	—	—	—	—	31	250	—	—	—	—
2		250	125	—	62.5	—	125	—	125	—	—	125	62.5	31	250	62.5	125	—
3		4	<1	—	8	8	16	—	>250	—	—	>250	>2	250	—	16	—	250
5		—	—	—	—	—	—	—	—	—	—	—	>250	>250	—	—	—	—
9		250	125	—	250	—	250	—	—	—	250	—	62	>250	—	>250	—	>250
10	5NT—CH=NOH	31	8	—	8	62.5	16	—	31	62.5	62.5	31	2	31	62.5	16	31	62.5
11	5NT—CH=CH— 	4	2	—	1	—	8	—	—	125	125	125	2	31	—	62	125	62
12	5NT—CH=CH— 	<16	<16	—	—	<16	—	—	—	—	—	—	>250	—	—	—	—	—
13	5NT—CH=CHC$_6$H$_5$	—	4	—	4	—	—	—	—	—	—	—	250	—	—	—	—	250
14	5NT—CH=CH— 	4	<2	250		4	4	250	—	—	—	>250	4	>250	—	250	—	250
17	5NT—CH=NN— 	16	8	250		31	16	—	31	250	250	31	2	16	62.5	31	62.5	8
18	5NT—CH=NNHCONH$_2$	31	16	—		>250	16	—	—	—	—	250	2	16	—	>250	—	16
19	5NT—CH=NNHCSNH$_2$	16	8	—	2	31	8	—	—	—	—	62.5	2	31	—	62.5	—	16
20	5NT—CH=NN— 	62.5	31	250	16	31	16	—	250	—	—	250	4	62.5	250	250	250	31
21	5NT—CH=NN— 	8	16	—	8	31	16	—	—	250	—	31	2	4	62.5	16	16	2
22		8	4	250	1	31	8	—	62.5	62.5	125	31	1	31	62.5	31	62.5	62.5
23	2NT—CH=NN— 	16	31	—	8	—	31	—	62	125	125	31	2	31	125	62	125	31
24	2NT—CH=NN— 	8	8	—	8	4	62	—	125	—	125	62	1	31	125	62	125	31
25	2NT—CH=NN— 	125	250	250	62	250	125	250	250	250	>250	250	8	250	250	250	250	125
26	2NT—CH=NNHCSNH$_2$	31	16	16	31	>250	125	—	>250	62	62	125	4	125	125	125	250	125
27	2NT—CH=NOH	31	16	250	31	250	125	—	125	31	31	125	4	62	125	62	125	62
28		8	8	250	8	250	3	250	125	62	16	62	4	31	62	62	62	62
29		—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
30	4NT—CH=NN— 	—	250	—	—	—	—	—	—	—	—	—	250	—	—	—	—	250
F[c]		4	1	31	2	16	16	>250	8	62	>250	4	4	2	4	2	4	2

[a] Agar dilution tests. [b] Dash indicates compound was inactive at the highest test level, 250 μg/ml; >250 indicates partial inhibition of highest test level. B.c. = *Bacillus cereus* ATCC 10702, B.s. = *Bacillus subtilis* ATCC 6633, B.t. = *Bacillus thuringiensis*, M. = *Micrococcus*, S.a. = *Staphylococcus aureus* ATCC 6538, St. a. = *Streptococcus agalactiae*, S.f. = *Streptococcus faecalis* ATCC 8043, A.a. = *Aerobacter aerogenes*, A.f. = *Alcaligenes faecalis* ATCC 8750, B.b. = *Bordetella bronchiseptica*, E.c. = *Escherichia coli* 2, P.m. = *Pasteurella multocida* RC 315, S.c. = *Salmonella choleraesuis* var. *kunzendorf*, S.d. = *Salmonella dublin*, S.g. = *Salmonella gallinarum* 605, S.t. = *Salmonella typhimurium*, S. ty. = *Salmonella typhosa* ATCC 6539. [c] F = furazolidone.

(Me_2CO) afforded an analytical sample, mp 208–209°. *Anal.* ($C_{14}H_9N_3O_2S$) C, H, N, S.

β-(5-Nitro-2-thiazolyl)styrene (13).—Benzaldehyde (3 ml) and 0.5 g (3.47 mmoles) of 2-methyl-5-nitrothiazole were heated at reflux temperature in the presence of 5 drops of piperidine for 15 min. The mixture was cooled, and 15 ml of Et_2O was added to precipitate a solid, which was collected and washed thoroughly with Et_2O; this gave 0.48 g, mp 159–163°. The solid was recrystallized from 95% EtOH and decolorized with activated C to give 0.34 g (42%) of yellow-orange crystals, mp 164.5–166°. The analytical sample was obtained by recrystallization (MeCO); mp 164.5–166°. *Anal.* ($C_{11}H_8N_2O_2S$) C, H, N, S.

5-Nitro-2-[2-(5-nitro-2-thiazolyl)vinyl]furan (14).—In 30 ml of glacial HOAc, 1.44 g (0.01 mole) of 2-methyl-5-nitrothiazole, and 1.69 g (0.012 mole) of 5-nitro-2-furfuraldehyde were dissolved, and a catalytic amount of freshly fused $ZnCl_2$ was added. The mixture was stirred, heated at reflux for 3 hr, cooled, and evaporated to dryness *in vacuo* to give a solid which was washed with EtOH and collected. The crude yield was 0.6 g and this material was recrystallized from 2:1 EtOH–Me_2CO to give orange crystals,

TABLE II

In Vitro ANTIFUNGAL ACTIVITY OF NITROTHIAZOLE DERIVATIVES[a]

	Min inhib concn, μg/ml[b]											
Compd	C.a.	C.m.	S.c.	M.r.	F.e.	H.c.	T.m.	M.g.	P.d.	M.e.	C.g.	A.f.
10	125	62	31	62	31	15		15	31	4	15	62
3	31	125	15	15			8	8	8	31	15	125
14	4	8	4	15			2	4	15		4	15
17			125	125		250	62	62	62		62	250
22	125	250	31		62	62		15	31	31	15	62
24							62	125	125		62	
28	125	125	62	250	125	125	31	31	62	125	15	125

[a] Agar dilution tests. Compounds **23**, **25**, and **26** were inactive against all organisms at 250 μg/ml. [b] Dash indicates compound was inactive at the highest test level, 250 μg/ml. C.a. = *Candida albicans* Bergen strain E-3, C.m. = *Candida mycoderma* ATCC 9888, S.c. = *Saccharomyces cerevisiae* ATCC 4100, M.r. = *Mucor ramannianus* M-143, F.e. = *Fusarium episphaeria* F-105, H.c. = *Hormodendrum cladosporoides* Z-516, T.m. = *Trichophyton mentagrophytes* E-11, M.g. = *Microsporum gypseum* E-28, P.d. = *Penicillium digitatum* P-308B, M.e. = *Memnoniella echinata* Z-583, C.g. = *Chaetomium globosum* H-71 QM 6694, A.f. = *Aspergillus fumigatus* S-246.,

mp 196–197°. In another run, multiple extraction of the crude material (Me_2CO) gave the product in 40% yield. *Anal.* ($C_9H_5N_3O_3S$) C, H, N, S.

2-Amino-5-(5-nitro-2-thiazolyl)-1,3,4-thiadiazole (22).—A heterogeneous mixture of 0.655 g (2.8 mmoles) of 5-nitro-2-thiazolecarboxaldehyde thiosemicarbazone in a solution of 5.5 g of $FeNH_4(SO_4)_2 \cdot 12H_2O$ in 10 ml of H_2O was stirred and heated at 80–90° for 2 hr and cooled and the product was collected. Recrystallization of the crude material from EtOH–DMF afforded 0.35 g of yellow solid, mp 249° dec. *Anal.* ($C_5H_3N_5O_2S_2$) C, H, N, S.

2-Amino-5-thiazolecarboxaldehyde (8).—Chloromalondialdehyde was treated with thiourea in 50% aqueous HOAc to give 32–42% crude yields of the aldehyde[16] **8**, mp 160–167° (lit.[17] mp 172–175°), which was used without purification.

2-Nitro-5-thiazolecarboxaldehyde (9). Method A.—A solution of 0.5 g (4 mmoles) of 2-amino-5-thiazolecarboxaldehyde in 3 ml of 48–50% aqueous HBF_4 was cooled to 0° and stirred as 0.27 g (4 mmoles) of $NaNO_2$ was added gradually. The mixture was stirred for 50 min at 0°, and then added, in portions, to a vigorously stirred suspension of 0.8 g of Cu powder in 10 ml of 30% aqueous $NaNO_2$ solution at 25°. This caused foaming and NO_2 was liberated. After 1 hr of stirring, the mixture was filtered and the filtrate was diluted with 15 ml of H_2O and extracted twice with 40 ml of benzene. The combined extracts were dried ($MgSO_4$) and evaporated to dryness *in vacuo* to afford a yellow syrup (0.275 g or 45%), which gradually solidified. Recrystallization of the solid (Et_2O) afforded yellow crystals, mp 84.5–85°. The ir and nmr spectra affirmed the structure of the aldehyde. *Anal.* ($C_4H_2N_2O_3S$) C, H, N, S.

Method B.—A solution of 10 g (0.078 mole) of 2-amino-5-thiazolecarboxaldehyde in 80 ml of 18% aqueous HBF_4 at 40–60° was added slowly to a vigorously stirred suspension of 5 g of Cu powder in 100 ml of 20% aqueous $NaNO_2$ solution at 10–15°. The mixture was stirred 2 hr and filtered, and the product was isolated by benzene extractions. The crude yield was 49%.

1-{[(2-Nitro-5-thiazolyl)methylene]amino}-2-imidazolidinone (23): 96% yield, red crystals, mp 252–254°. *Anal.* ($C_7H_7N_5O_3S$) C, H, N, S.

3-{[(2-Nitro-5-thiazolyl)methylene]amino}-2-oxazolidinone (24): 84% crude yield, recrystallized from DMF–EtOH (1:2), orange crystals, mp 254–255° dec. *Anal.* ($C_7H_6O_4N_4S$) C, H, N, S.

1-{[(2-Nitro-5-thiazolyl)methylene]amino}hydantoin (25): 70% crude yield, recrystallized from aqueous EtOH, yellow crystals, mp 239–240°. *Anal.* ($C_7H_5N_5O_4S$) C, H, N, S.

2-Nitro-5-thiazolecarboxaldehyde thiosemicarbazone (26): 87% crude yield, recrystallized from DMF–Me_2CO–EtOH (5:5:1), dark red powder, mp >285°. *Anal.* ($C_5H_5N_5O_2S_2$) C, H, N, S.

2-Nitro-5-thiazolecarboxaldehyde oxime (27) had an 82% crude yield, mp 160–163°. Recrystallization from aqueous EtOH lowered the melting point and the recrystallized sample after sublimation melted at 149°. The nmr spectrum (Me_2CO-d_6) showed two singlets at τ 2.8 and 2.66. *Anal.* ($C_4H_3N_3O_3S$) H, N, S, C: calcd, 27.74; found, 28.82.

This oxime is apparently dehydrated easily since a crude sample after sublimation exhibited a –CN absorption at 2235 cm^{-1}.

2-Amino-5-(2-nitro-5-thiazolyl)thiadiazole (28).—To a mixture containing 40 ml of DMF, 20 ml of H_2O, and 20 ml of 50% aqueous $FeCl_3$ solution, 4.6 g (0.02 mole) of 2-nitro-5-thiazolecarboxaldehyde thiosemicarbazone was added and the mixture was heated at 100° for 2 hr. After stirring overnight at room temperature, the mixture was diluted with 100 ml of H_2O and filtered to give a dark brown solid. This solid was thoroughly extracted with Me_2CO and the extracts were decolorized with activated C and evaporated to dryness to give 1 g (22%) of orange product, mp >300°. *Anal.* ($C_5H_3N_5O_2S_2$) C, H, N, S.

3-(2-Thiazolylmethyleneamino)-2-oxazolidinone (29).—A solution of 3.8 g (0.38 mole) of 2-methylthiazole in 15 ml of 1,2-dimethoxyethane was heated under reflux while 4.26 g (0.38 mole) of freshly prepared SeO_2 in 15 ml of dimethoxyethane and 2.5 ml of H_2O was added in 1 hr. The mixture was cooled after 32.5 hr and filtered through $MgSO_4$, the filter cake was washed (Et_2O), and the filtrate and washings were steam distilled. The distillate was then saturated with NaCl and extracted with $CHCl_3$, and the extracts were dried ($MgSO_4$) and evaporated *in vacuo* to give 0.56 g of liquid. Glpc analysis on a 20% SE 30 6-ft column at 140° revealed the presence of only *ca.* 56% of the aldehyde (equal to *ca.* 7% yield) along with *ca.* 43% of 2-methylthiazole in the distillate. To this mixture 0.3 g of aminooxazolidinone in 2 ml of EtOH was added along with a drop of concentrated HCl to give a white solid. An additional 2 ml of EtOH was added and the mixture refluxed for 3–4 min. The mixture was cooled and the white product was collected. It melted at 190–191.5°, 0.48 g (6.4%). A sample recrystallized from MeOH melted at 190–192°. *Anal.* ($C_7H_7N_3O_2S$) C, H, N, S.

3-{[(4-Nitro-2-thiazolyl)methylene]amino}-2-oxazolidinone (30).—A solution of 0.65 g (4.5 mmoles) of 2-methyl-4-nitrothiazole[14] in 15 ml of glacial HOAc and 15 ml of Ac_2O was cooled to 5° and 2 ml of concentrated H_2SO_4 was added. To this was added 2 g (0.02 mole) of CrO_3 in 45 min at 5–10°. After 1 hr at this temperature, the mixture was allowed to rise to room temperature. Since no C=O band was observed in the ir spectrum of a sample (CH_2Cl_2 extract of an aliquot which had been neutralized with K_2CO_3), an additional 2 g of CrO_3 was added in 20 min and the temperature was allowed to rise to 32.5°. After 6.75 hr, the mixture was poured on ice-cold, saturated K_2CO_3 and the resultant solution was extracted (CH_2Cl_2). The extracts were dried ($MgSO_4$) and evaporated *in vacuo* to give 0.1 g of 4-nitro-2-thiazolecarboxaldehyde diacetate[20] [ν_{max} (neat) 1775 cm^{-1}] and starting material. The crude diacetate was dissolved in cold CCl_4 and filtered to remove starting material, and the filtrate was evaporated to dryness to give *ca.* 20 mg of partially purified material. This was derivatized with aminooxazolidinone in the manner of Gever[17] to give white needles of product, mp 273–275° (after recrystallization from Me_2CO). *Anal.* ($C_7H_6N_4SO_4$) C, H, N, S.

Attempts at SeO_2 oxidation of 2-methyl-4-nitrothiazole in dioxane or dimethoxyethane were unsuccessful.

2-[2-(2-Acetamido-5-thiazolyl)vinyl]quinoline (15).—A suspension of 5.0 g (0.039 mole) of 2-amino-5-thiazolecarboxaldehyde in 15 ml of Ac_2O was heated at reflux for 2 hr, the mixture was cooled, and the solid was collected and dried to give 4.7 g (70%)

(20) Although no elemental analyses were obtained for this material, its ir spectrum, the elemental analyses of **30**, and the unequivocal structure proof of 5[13] support its structure.

of tan solid, mp 234–235°.[21] This material was suspended in 20 ml of glacial HOAc and heated to 60° and 4.0 g (0.028 mole) of freshly distilled quinaldine and 5 ml of Ac_2O were added. The mixture was heated at reflux for 4 hr and cooled over a week-end, and 100 ml of Et_2O was added. After filtering and drying, 5 g (61%) of product, mp 269–272°, was obtained. A sample was recrystallized from EtOH–Me_2CO (1:1) with added DMF to increase the solubility; mp 273–274°. *Anal.* ($C_{16}H_{13}N_3OS$) C, H, N, S.

2-[2-(2-Amino-5-thiazolyl)vinyl]quinoline (31).—A suspension of 0.4 g (1.35 mmoles) of **15** in a mixture of 2 ml of glacial HOAc and 2 ml of concentrated HCl was heated at reflux for 2 hr to give a clear, dark solution. The solution was evaporated to dryness, and the residue was dissolved in 100 ml of H_2O and treated with saturated $NaHCO_3$ until gas evolution stopped. The precipitate was collected, washed (H_2O), and dried to give 0.275 g (80%) of yellow crystals, mp 244–246°. *Anal.* ($C_{14}H_{11}N_3S$) C, H, N, S.

Attempts to convert the NH_2 group to NO_2 by diazotization in the presence of Cu and excess $NaNO_2$ gave traces of a semisolid which had an ir spectrum showing NO_2 bands (1510 and 1340 cm^{-1}), which was similar to the spectrum of **12**. A virtually identical spectrum was obtained from a crude solid which had been isolated from an attempt to condense 2-nitro-5-thiazolecarboxaldehyde with quinaldine in refluxing HOAc–Ac_2O.

(21) H. Taniyama, B. Yasui, and F. Inoue [*J. Pharm. Soc. Jap.*, **73**, 276 (1953)] report mp 207° dec.

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Synthesis of 3-[(5-Nitrofurfurylidene)amino]hydantoins and N-Ethoxycarbonylamino Acid Nitrofurfurylidenehydrazides

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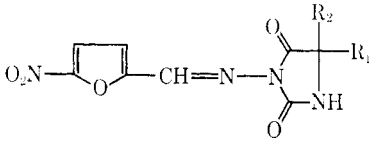
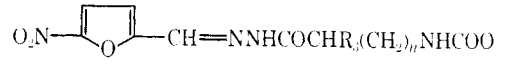
Some 3-[(5-nitrofurfurylidene)amino]hydantoins and some N-ethoxycarbonylamino acid nitrofurfurylidenehydrazides have been synthesized for antibacterial screening. Improved procedures for the preparation of 3-aminohydantoins have been developed.

In view of the chemotherapeutic properties of 1-[(5-nitrofurfurylidene)amino]hydantoins,[1] we synthesized and screened several 3-[(5-nitrofurfurylidene)amino]hydantoins (I). Three examples of N-ethoxycarbonylamino acid 5-nitrofurfurylidenehydrazides (II), open-chain forms of the hydantoins, were also prepared for antibacterial screening.

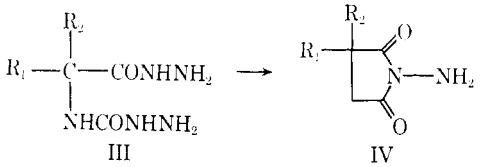


I



II, $n = 0, 1$

The synthesis of several 5,5-disubstituted 3-aminohydantoins (IV) by heating aqueous solutions of N-carboxy-α-amino acid dihydrazides (III) at atmospheric pressure has been reported by Taub[2] (method B).



III IV

Earlier, Schlögl, *et al.*,[3,4] had found this method unsatisfactory for the synthesis of monosubstituted hydantoins; yields decreased as the size of the substituent decreased, and they were unable to prepare the unsubstituted 3-aminohydantoin or its 5-hydroxymethyl analog. We also were unable to prepare either the unsubstituted or the 5-methyl compound by heating aqueous solutions of dihydrazides.

More recently another synthesis of 5,5-disubstituted 3-aminohydantoin from 5,5-disubstituted hydantoins and hydrazine hydrate was devised by Davidson.[5] The applicability of this method to the preparation of 5-monosubstituted 3-aminohydantoins or to unsubstituted 3-aminohydantoin was not mentioned. These methods, then, are of limited value for the preparation of 3-aminohydantoins.

We have developed a reliable procedure for the preparation of 5-monosubstituted 3-aminohydantoins (IV, R_1 = H), which consists of heating under reflux a dilute solution of the dihydrazide (III) in DMF. The compounds prepared in this way are listed in Table I (method A). This method is applicable for either large or small substituents, as well as the unsubstituted compound.

Although the procedure of Taub[2] was used for preparation of the dimethyl and methylethyl compounds (Table I, method B), we found that the low yield of the latter compound could be doubled by a third procedure (method C).[3] This consisted of heating under reflux an ethanol solution of ethyl N-ethoxycarbonyl-DL-iso-

(1) (a) M. Abrams and B. Prophete, *Missouri Med.*, **51**, 280 (1954); (b) K. J. Hayes, U. S. Patent 2,610,181 (1952); *Chem. Abstr.*, **47**, 6980*i* (1953); (c) J. G. Michels, U. S. Patent 3,075,973 (1963).

(2) W. Taub, U. S. Patent 2,767,193 (1956); *Chem. Abstr.*, **51**, 5841*h* (1957).

(3) K. Schlögl and G. Korger, *Monatsh. Chem.*, **82**, 799 (1951); *Chem. Abstr.*, **47**, 7511*a* (1953).

(4) K. Schölgl, J. Derkosch, and E. Wawersich, *Monatsh. Chem.*, **85**, 607 (1954); *Chem. Abstr.*, **49**, 9511*d* (1955).

(5) J. S. Davidson, *J. Chem. Soc.*, 4646 (1964).