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Potential Chemotherapeutics

II. 2-(5-Nitro-2-furyl)-5-amino-1,3,4-thiadiazoles *

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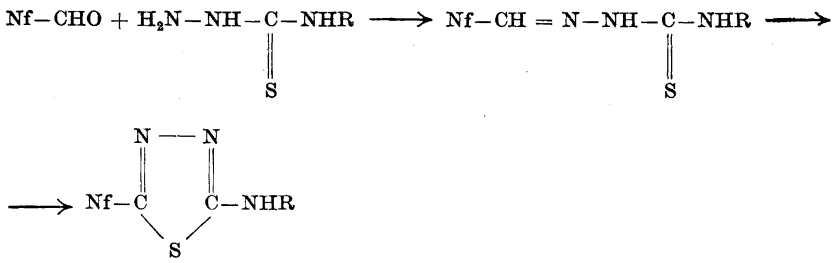
Seventeen 2-(5-nitro-2-furyl)-5-amino-1,3,4-thiadiazoles were prepared by oxidation of the corresponding thiosemicarbazones with ferric salts according to Young and Eyre or electrolytically in the presence of ferric salts.

In 1944 Dodd and Stillman discovered the antibacterial activity of the nitrofurans [1]. Since then a great number of nitrofuryl derivatives have been synthesized and tested. Several of these compounds are hydrazone-type derivatives of 5-nitrofurfural such as nitrofurazone, nitrofurantoin and furazolidone, all possessing a broad antibacterial spectrum [2]. It is also known, that 5-nitrofurfural thiosemicarbazone, in analogy to many other thiosemicarbazones, has a strong tuberculostatic activity [3].

Since the nitrofurans mentioned have a structure, which could be expected to be relatively easily destroyed, for example by acid hydrolysis, it was of interest to synthesize compounds with a more stable structure. This is readily accomplished for example by oxidative ring closure of nitrofurfural thiosemicarbazone to the corresponding aminothiadiazole. As it became clear that this compound had promising antibacterial properties, a series of derivatives were prepared for antibacterial screening.

All substances prepared are 5-(nitrofuryl)-thiadiazoles with an unsubstituted or a monosubstituted amino group in the 2-position of the thiadiazole ring system. They were prepared from nitrofurfural (or its diacetate) and the corresponding 4-substituted thiosemicarbazide as follows (Nf stands for 5-nitro-2-furyl):

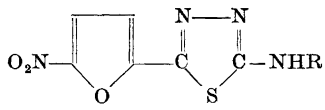
* During the preparation of this manuscript the authors were informed that Dr. W. Sherman at Abbott Labs. during the September Meeting of A.C.S. Division of Medicinal Chemistry lectured on nitrofuryl compounds, comprising some of the compounds mentioned in this paper.



The thiosemicarbazides were prepared from the corresponding *iso*thiocyanates and hydrazine hydrate, with the exception of 4-(2-pyridyl)-thiosemicarbazide. When 2-pyridyl-*iso*thiocyanate was allowed to react with hydrazine hydrate in ethanol under various conditions, only 1,6-dipyridyl-2,5-dithiobiurea was obtained. Therefore, 4-pyridylthiosemicarbazide was prepared from the triethylammonium salt of 2-pyridyldithiocarbamic acid.

The oxidation of thiosemicarbazones of aromatic aldehydes is readily performed with ferric chloride [4]. However, when applied to heterocyclic derivatives the method does not always seem to be convenient. Thus, Erlenmeyer *et al.*[5] obtained only a small yield on ferric chloride oxidation of 2-pyridylaldehyde thiosemicarbazone, and Maffii *et al.*[6] obtained 41.6 % yield on oxida-

Table 1. Substituted 2-(5-nitro-2-furyl)-5-amino-1,3,4-thiadiazoles.



Compd No.	Ph 778/ X	R	M. p.	M.p. of Thiosemicarb.	Yield %	Recrystallization solvent
1	1	H	272 — 3	250	87	DMSO; DMF; HOAc
2	6	CH_3	219 —21	216 —18	63	Ethanol-dioxane
3	8	C_2H_5	219 —21.5	181 — 3	71	Ethanol-dioxane
4	10	*n*-C_3H_7	190 — 1	170 — 1.5	87	Ethanol
5	7	*i*-C_3H_7	213 —15	188 — 9	75	Ethanol-dioxane
6	9	*n*-C_4H_9	186 — 7	152.5— 4	74	Ethanol-dioxane
7	14	*i*-C_4H_9	183.5— 4.5	173.5— 5	80	Ethanol
8	55	*n*-C_6H_{13}	162.5— 3.5	128.5—9.5	50	Ethanol
9	22	C_6H_{11}	214 —15	194 — 5	76	Ethanol-dioxane
10	21	$-CH_2-CH=CH_2$	172.5— 5.5	177 — 8	71	Ethanol
11	12	$-CH_2-C_6H_5$	190 — 1.5	211 —12	50	Ethanol-dioxane
12	43	$-CH_2-CH_2-C_6H_5$	160 — 9	188 — 9	50	Ethanol
13	41	$-CH_2-CH_2-O-CH_3$	169 —71	169 —70	60	Ethanol
14	16	$-CH_2-CO_2-Et$	172.5— 4	178.5— 9.5	77	Ethanol
15	18	$-CH_2-COOH$	243.5— 4	—	—	Ethanol-dioxane
16	19	$-C_6H_5$	253 — 4.5	196.5— 7	62	Ethanol-dioxane
17	24	$-2-C_5H_4N$	dec.	dec.	58	DMF

tion of 2-furfural thiosemicarbazone. In the present work, however, oxidation of 5-nitrofurfural thiosemicarbazone yields about 84 % of the aminothiadiazole. The difference in yield between the furyl and 5-nitrofuryl derivative might be attributed to the inhibitory influence of the nitro group on the acid cleavage of the furyl group.

Attempts were made to use other oxidation agents such as cerium sulphate, lead tetraacetate, chromium trioxide, selenium dioxide, potassium permanganate and hydrogen peroxide. However, either no reaction occurred or unidentified substances were formed (*cf.* Ref.[7]). On the other hand, it was possible to perform an electrolytic oxidation in the presence of small amounts of ferric salts. Probably, the ferrous ions formed by the ring closure were oxidized, thus allowing the reaction to proceed. However, attempts to replace the electric current with potassium permanganate failed, and the yield of aminothiadiazole formed in this case corresponded to the amount of ferric ions originally present in the reaction.

Under alkaline conditions the nitrofuryl group is fairly labile and extensive decomposition may occur. It was found, however, that the oxidation could be performed with potassium ferricyanide in the presence of sodium acetate [8], but the time of reaction was considerably increased.

Compound No. 12 in Table 1 (the 2-phenylethyl derivative) has a very unsharp melting-point in spite of several consecutive recrystallizations. Melting-point depression is obtained when the substance is mixed with the thiosemicarbazone used as starting material. It seems probable, that the broad melting-point interval is due to polymorfism. A similar behaviour is shown by the substance No. 10 (allyl derivative). On recrystallization of the latter compound two kinds of crystals are obtained — brown rods and yellow flakes. At about 150°C the shells turn brown, and a sharp melting point is obtained.

The thiadiazoles prepared are yellow substances with slightly alkaline properties. In contrast to the corresponding thiosemicarbazones, they are readily soluble in cold formic acid, which could be used to separate the substances, when mixtures were obtained. Compound No. 1 (unsubstituted amino group), which is the most promising as a potential chemotherapeutic, is slightly soluble in most common solvents but comparatively soluble in formic acid, dimethyl formamide and dimethyl sulfoxide.

In serial dilution tests *in vitro* Compound No. 1 inhibits growth of a series of strains of *Staphylococcus aureus* in a concentration of 3.2—25 μg/ml, of *Salmonella*, *Shigella* and *Coli* in concentrations of 0.4—6.3 μg/ml and of *Streptococcus* in concentrations of 12.5—200 μg/ml. Further details of the antibacterial screening will be published separately.

Table 2. Analytical results.

Compd No.	Summary formula	% C calc.	% C found	% H calc.	% H found	% N calc.	% N found	% S calc.	% S found
1	$C_6H_4N_4O_3S$	34.0	33.8	1.9	1.9	26.4	26.5	15.1	15.0
2	$C_7H_6N_4O_3S$	37.2	37.0	2.7	3.0	24.8	24.7	14.2	14.0
3	$C_8H_8N_4O_3S$	40.0	40.2	3.4	3.4	23.3	23.3	13.4	13.3
4	$C_9H_{10}N_4O_3S$	42.5	42.4	4.0	4.1	22.0	21.8	12.6	12.8
5	$C_9H_{10}N_4O_3S$	42.5	42.5	4.0	4.1	22.0	21.9	12.6	12.7
6	$C_{10}H_{12}N_4O_3S$	44.8	45.0	4.5	4.6	20.9	20.7	12.0	11.9
7	$C_{10}H_{12}N_4O_3S$	44.8	44.9	4.5	4.6	20.9	20.4	12.0	11.8
8	$C_{12}H_{16}N_4O_3S$	48.6	48.7	5.4	5.6	18.9	18.9	10.8	10.8
9	$C_{12}H_{14}N_4O_3S$	49.0	49.0	4.8	4.9	19.0	19.0	10.9	11.0
10	$C_9H_8N_4O_3S$	43.0	43.0	3.2	3.2	22.3	22.2	12.8	12.8
11	$C_{13}H_{10}N_4O_3S$	51.7	52.0	3.3	3.4	18.5	18.2	10.6	10.2
12	$C_{14}H_{12}N_4O_3S$	53.2	53.3	3.8	3.9	17.7	17.6	10.1	10.4
13	$C_9H_{10}N_4O_4S$	40.0	40.0	3.7	4.0	20.7	20.6	11.9	11.2
14	$C_{10}H_{10}N_4O_5S$	40.3	40.4	3.4	3.4	18.8	18.4	10.8	10.7
16	$C_{12}H_8N_4O_3S$	50.0	49.9	2.8	2.9	19.4	19.2	11.1	11.1
17	$C_{11}H_7N_5O_3S$	45.7	45.9	2.4	2.6	24.2	24.4	11.1	11.0

EXPERIMENTAL *

5-Nitro-2-furyl thiosemicarbazones. The thiosemicarbazones were prepared by refluxing 5-nitro-furfural diacetate and the corresponding thiosemicarbazide in a mixture of ethanol and dilute sulfuric acid, or by heating 5-nitrofurfural and the thiosemicarbazide in ethanol. The product obtained, usually orange-coloured, was recrystallized from dimethyl formamide, dimethyl sulfoxide, dioxane or ethanol. The products decompose on heating at temperatures given in Table 1.

2-(5-Nitro-2-furyl)-5-amino-1,3,4-thiadiazole. a) 125 g (0.58 mole) of 5-nitro-2-furfural thiosemicarbazone and 1 160 g of a 25 % solution of ferric chloride (490 g of $FeCl_3 \cdot 6\ H_2O$) in water are placed in a 1.5 l beaker, fitted with a stirrer and a thermometer. The mixture is stirred on a water bath at 80°C for 90 min and then chilled to room temperature. The precipitate is filtered off and washed with 800 ml of hot water, followed by 800 ml of cold water, and dried at 60°C. Weight 118 g. The substance is dissolved in 360 g of dimethyl sulfoxide, treated with Norite and filtered. 30 ml of distilled water is added to the hot solution and the mixture is chilled in a refrigerator. The yellow crystals are filtered off, washed with 140 ml of ethanol, 180 ml of water and dried. Yield 108 g (87 %). M. p. 270–271° (decomp.). If the substance is heated slowly it darkens gradually without melting.

b) A porcelain cell with a volume of about 200 ml is placed in a 500 ml vessel, used for electrolysis. A mixture of 21.4 g (0.1 mole) of 5-nitro-2-furfural thiosemicarbazone and 125 ml of 15 % potassium ferrisulfate in water is poured in the cell. The mixture is stirred thoroughly with a perforated platinum anode about 5 × 4 cm^2 in size. A cylinder-shaped copper cathode is placed in the cathode-room outside the cell, which is filled with dilute sulfuric acid.

The electrolysis is performed at about 70°C with about 2 A for 4 h. The solid in the anode-cell is filtered off, washed with hot water and recrystallized from DMF-water as described in method a). Yield 14 g. M. p. 270–272°C.

c) 8.5 g (0.04 mole) of nitrofurfural thiosemicarbazone and a solution of 29 g (0.09 mole) of potassium ferricyanide and 25 g of sodium acetate in 200 ml of water were stirred for 8 h on a water bath. The precipitate obtained on cooling was filtered off, washed and dried. 6.3 g (75 %) of yellow crystals were obtained, which decomposed at 272°C. A reaction time of 2 h and 5 h yielded, respectively, 20–30 % and 50 %.

* Microanalyses by Dr. Wolfgang Kirsten.

The 2-(5-nitro-2-furyl)-5-amino-1,3,4-thiadiazoles, listed in Table 1, were prepared in analogy to method a) above. A few examples further illustrate the procedure:

2-(5-Nitro-2-furyl)-5-allylamino-1,3,4-thiadiazole. 25.4 g (0.1 mole) of 1-(nitrofurfural)-4-allylthiosemicarbazone is stirred on a water bath for 2 h with a solution of 50 ml of 50 % $FeCl_3$ and 70 ml of water. The dark product obtained was dissolved in hot ethanol, carbon added, the hot solution filtered and allowed to crystallize. 12.5 g of yellow-brown needles and 5.8 g of yellow flakes were obtained. The needles melt at 172.5–173.5°C. The flakes rearrange to the brown needles at about 150°C and melt at 172.5–173.5°C.

2-(5-Nitro-2-furyl)-5-carboxymethylamino-1,3,4-thiadiazole. 2.5 g of 1-(nitrofurfural)-4-carbethoxymethylthiosemicarbazone is stirred with 15 ml of 15 % $FeCl_3$ and 20 ml of H_2O on a water-bath for one hour. The dark precipitate was dissolved in ethanol, carbon added, and the hot mixture was filtered. After cooling, 1.95 g of 2-(nitrofuryl)-5-carbethoxymethylaminothiadiazole was obtained as yellow flakes melting at 172.5–174°C.

0.9 g of the ester was heated on a water bath for 3 h with 20 ml of 5 N sulfuric acid. The yellow precipitate was filtered off, and washed with ethanol. 0.8 g of needles were obtained which decomposed at 246–248°C. 0.5 g of the substance was dissolved in 50 ml of ethanol-dioxane 1:1 and filtered. After cooling, 0.4 g of yellow crystals was obtained. M.p. 243.5–244°C (decomp.). Equiv. wt: Found 269. Calc. 270.

REFERENCES

1. Dodd, M. C. and Stillman, W. B. *J. Pharmacol. Exptl. Therap.* **82** (1944) 11.
2. *Introduction to the Nitrofurans.* Eaton Laboratories 1958.
3. Freedlander, B. L. and Furst, A. *Wasmann* J. Biol. **9** (1951) 355.
4. Young, G. and Eyre, W. *J. Chem. Soc.* **79** (1901) 54.
5. Hemmerich, P., Prijs, B. and Erlenmeyer, H. *Helv. Chim. Acta* **41** (1958) 2058.
6. Maffii, G., Testa, E. and Ettorre, E. *Il Farmaco* **13** (1958) 187.
7. Hoggarth, E. *J. Chem. Soc.* **1951** 2202.
8. Brown, E. V. *Iowa State Coll. J. Sci.* **11** (1937) 227. *Chem. Abstr.* **31** (1937) 8528.

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