the solution against *E. coli* was maintained without lowering even after 96 hours. It had already been reported[3] that the N-oxides in this condition are invariably transformed into the ring-oxides, *e.g.* N,N-methyl-β-chloroethyldimethylene-1,2-oximinium chloride (in case of the compound No. 10), within several hours. Therefore the transformed compound should be as active against *E. coli* as the original N-oxide and the lowering of activity, caused by hydrolysis of the chlorine atoms, could only be first observed after 192 hours (Table III).

TABLE III

Hours after being dissolved in water with $NaHCO_3$	HN_2 N-Oxide Hydrochloride Minimum Concentration for Inhibition *E. coli*: 24 hrs.' incubation		
	100 γ/cc.	50 γ/cc.	25 γ/cc.
0	—	—	+
72	—	—	++
96	—	—	++
192	—	++	++

The minimum concentration for inhibition of the compound (No. 10) fell to $5 \cdot 10^{-3}$ mole in the medium containing casein hydrolyzate and similarly its activity fell almost to zero when sodium thiosulfate was added to the pure synthetic medium[2] in concentration of 0.5%.

Summary

Twenty-three derivatives of nitrogen mustards and their N-oxides were examined as to their bacteriostatic activities *in vitro* using *Escherichia coli*. It was found that, in general, the bi- or polyfunctional N-oxides were more active than the corresponding tertiary nitrogen mustards, but the monofunctional N-oxides were far less active, regardless of whether they were N-oxide derivatives or tertiary ones. The results agreed well with experiments with Yoshida sarcoma animals.

(Received September 4, 1953)

3) M. Ishidate, Y. Sakurai: J. Pharm. Soc. Japan, 72, 1297(1952).

---

82. **Tadashi Sasaki:** Synthesis of Nitrofuran Sulfonic Acid Derivatives.*

(*Institute of Scientific Research for Practical Life, Medical Faculty, University of Kyoto***)

Nitrofuran derivatives with strong bactericidal activities, such as nitrofurfural semicarbazone or nitrofurylacrolein semicarbazone, are in general difficult to put into solution with ordinary organic solvents. This experiment was carried out in order to obtain nitrofuran derivatives having both characteristics of being easily soluble and also of being active in bacteriostasis. Several attempts[1] have been reported concerning this type of experiment. According to Hill and White 5-sulfofuran-2-carboxylic acid is nitrated by fuming nitric acid into nitrofuran-carboxylic acid and -sulfonic acid.[2]

Upon reëxamining this reaction, optimum conditions for preparation of nitrofuran

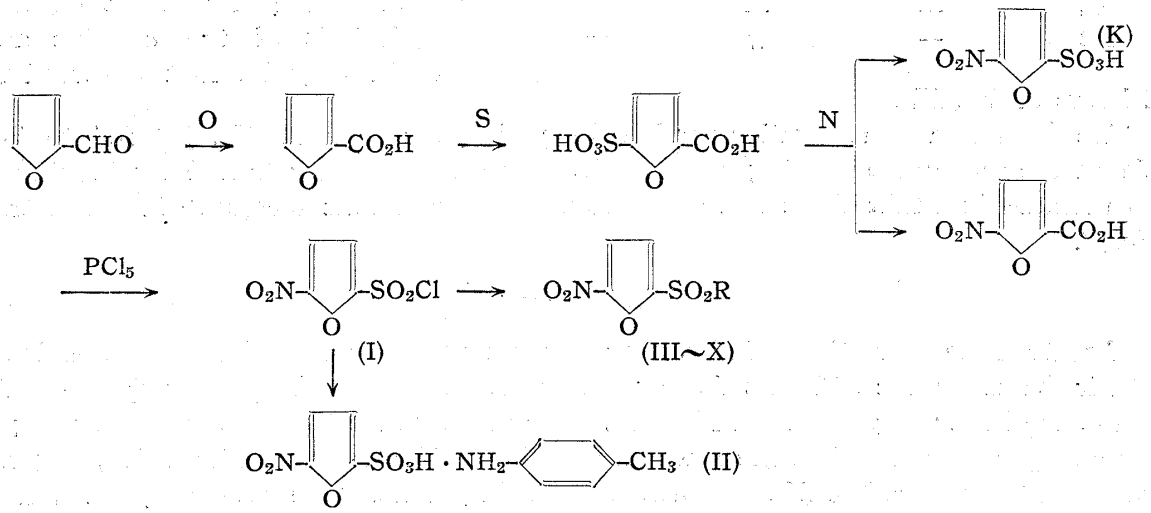
* Presented before the Annual Meeting of the Pharmaceutical Society of Japan, July, 1949.

** Yoshida-konoe-cho, Sakyo-ku, Kyoto (佐々木 正).

1) As a soluble furan, guanofuracin was prepared by Dr. Uota. Dr. Emoto later reported on some soluble furacin derivatives. Cf. M. Emoto: J. Soc. Chem. Ind. Japan, 55, 593(1952).

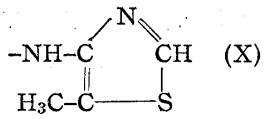
2) H. B. Hill, R. White: Am. Chem. J., 27, 196(1902).

sulfonic acid[3] were found. The potassium salt of nitrofuran sulfonic acid is converted into nitrofuran sulfochloride by treating it with phosphorus pentachloride. The free nitrofuran sulfonic acid is obtained by hydrolyzing the corresponding sulfochloride. This acid is very hygroscopic and is identified by its toluidine salt (II).[4] Sulfochloride (I) is condensed with several types of aliphatic and aromatic amines. The results concerning this are given in the experimental section. Another method of preparing sulfochloride (I), to give a better yield, was attempted, for example, by sulfonation or sulfochlorination of nitrofuran[5], but this has been unsuccessful. Furyl-sulfonamide without a nitro radical was recently obtained by another method.[6]



R: $-NH_2$ (III) $-NH-C_6H_5$ (IV) $-NH-C_6H_4-CH_3(p)$ (V) $-NH-C_6H_4-OCH_3(p)$ (VI)
$-NH-C_6H_4-OC_2H_5(p)$ (VII) $-NH-C_6H_4-COOC_2H_5(p)$ (VIII)

$-NH-C_{12}H_7$ (IX)



(X)

Among these nitrofurylsulfonic acid derivatives, nitrofurylsulfonylamide (III) is very active against *Staphylococcus aureus* and *Eberthella typhosa*, although nitrofuryl-carboxyl-amide is very inactive[7] as shown in Table I.

TABLE I. Antibacterial Action of 5-Nitrofurylsulfonamide Derivatives*

Compd.	Bacteria	*Staph. aur.*		*Strept. haemol.*		*Diploc. pneum.* 1.		*Esch. coli*	
	Hours	24	96	24	96	24	96	24	96
(III)		20	10	<1	<1	<1	<1	10	10
(IX)		2.5	1			<1	<1	1	<1
(VIII)		5	2.5			<1	<1	1	<1
(X)		1	<1			1	<1	2.5	1
K-salt		<1	<1			<1	<1	<1	<1

3) T. Sasaki: Bull. Chem. Soc. Japan (in press). Presented before the Annual Meeting of the Chemical Society of Japan, April, 1949.
4) L. F. Fieser: "Experiments in Organic Chemistry", 140(1941); J. Am. Chem. Soc., 51, 2460(1927); Org. Syntheses, Coll. Vol. II, 604(1943).
5) Terentiev, Kazitsyana, and Vestinik reported on the sulfonation of furan, methylfuran, and acetylfuran by using pyridine sulfonium salt.
6) R. Ocinneide: Nature, 160, 260(1947).
7) M. C. Dodd, W. B. Stillman: J. Pharmacol, Exptl. Therap, 82, 11(1944).

Compd.	Bacteria	*Eber. typhosa*		*S. dysenteriae*		*Pseudomonas aeruginosae*	
	Hours	24	96	24	96	24	96
(III)		20	10	10	5	<1	<1
(IX)		1	<1	1	<1	<1	<1
(VIII)		1	<1	<1	<1	<1	<1
(X)		2.5	1	<1	<1		
K-salt		<1	<1	2.5	1	<1 (Unit 10,000)	<1

* These tests were carried out by Dr. K. Ikegaki at the R. Kimura Laboratory, Medical Faculty, University of Kyoto.

Diffusibility of Nitrofuran Derivatives (mm.)

Concentration	1:20,000	1:80,000
Furacin	14.5	9.0
Nitrofurylsulfonamide	14.5	9.8

LD_{50} per 10 g. mouse (mg.)

	Subcutan.	Oral
Furacin	6.25	2.08
Nitrofurylsulfonamide	2.18	2.50

The author wishes to express his gratitude to Prof. H. Saikachi of the University of Kyushu for his kind advices. Elemental analysis was carried out by Miss Iwata of the University of Kyoto.

Experimental[8]

2-(5-Nitro)-furyl sulfochloride (I)—Ten g. of potassium nitrofuryl sulfonate is added to 15 g. of PCl_5 and gently heated to 140° under reflux in an oil bath for 4 hours. After adding further 10 g. of PCl_5, heating is continued for 6 more hours. When needle crystals of the potassium salt disappear, the reaction mixture is poured onto cracked ice, and then extracted with ether. The ether layer is dried with anhyd. Na_2SO_4 and after removing the ether, the residual yellow oil is purified by distillation under a reduced pressure. The oil, b.p_7 117~123°, gradually formed yellow crystals, m.p. 43°, on standing. These yellow crystals are easily soluble in ordinary organic solvents. Yield, 5.5 g. *Anal.* Calcd. for $C_4H_2O_5NClS$: C, 22.72; H, 0.95; N, 6.62. Found; C, 22.80; H, 1.10; N, 6.24.*

2-(5-Nitro)-furylsulfonic acid *p*-toluidine salt (II)—Sulfochloride (I) is suspended in the proper amount of water and hydrolyzed by gently warming in a water bath until it dissolves completely. This mixture is concentrated as much as possible to a syrup and dried completely for several days in a desiccator containing conc. H_2SO_4. The free acid thus obtained forms colorless needle crystals. Being very hygroscopic and unstable, it gradually darkens upon exposure to air. The toluidine salt is prepared as follows[4]: To a water solution of free acid (0.4 g.) is added 0.3 g. of *p*-toluidine and the mixture is warmed in a water bath under reflux for several minutes. After cooling, the precipitated crystals are recrystallized from hot water to light yellow crystals, m.p. 185°(decomp.). Yield, 0.3 g. *Anal.* Calcd. for $C_{11}H_{12}O_6N_2S$: C, 44.00; H, 4.00; N, 9.39. Found: C, 43.39; H, 4.33; N, 9.47.

2-(5-Nitro)-furylsulfonamide (III)—One g. of sulfochloride (I) is mixed with 2 g. of ammonium carbonate and melted slowly in a water bath at about 50° and stirred occasionally with a glass rod. During this process CO_2 gas evolves violently for about 1 hour. Heating is continued for further 10 minutes to complete the reaction at 60°, then cooled, and extracted with cold and hot absolute alcohol to remove the resulting ammonium chloride. After removing the alcohol the residual mass is extracted with acetone and the acetone layer evaporated to dryness to give 0.2 g. of light yellow crystals, m.p. 184.5~185°(decomp.).** *Anal.* Calcd. for $C_4H_4O_5N_2S$: C, 25.00; H, 2.08. Found: C, 24.99; H, 2.43.

8) All melting points are not corrected.
* A very small amount of ash remained.
** This compound is very difficult to purify. Hill and Sylvester reported on this fact on another furansulfonamide. Cf. Am. Chem. J., 32, 281(1910). Microanalysis showed the presence of ash which was excluded from the calculation.

2-(5-Nitro)-furylsulfonylanilide (IV)—To 0.5 g. of the sulfochloride (I) dissolved in dry ether is added one drop of dry pyridine which causes the solution to become cloudy and results in the formation of pyridinium salt. Shaking this solution and simultaneously adding dropwise freshly distilled aniline (0.25 g.) causes a red, bulky mass to precipitate immediately. After letting this mixture stand overnight at a room temperature, the ether is distilled and the residual crystals dried on a porcelain plate. Repeated recrystallization from ethanol-water (1 : 1) mixture gives thin yellow crystals (0.08 g.), m.p. 108.5~109°. *Anal.* Calcd. for $C_{10}H_8O_5N_2S$: C, 44.77; H, 2.99; N, 10.44. Found: C, 44.08; H, 3.32; N, 9.62.

2-(5-Nitro)-furylsulfonic *p*-toluidide (V)—This compound is obtained from 0.5 g. of (I) and 0.5 g. of *p*-toluidine by the procedure used for the preparation of (IV) and forms thin, brown plate crystals, m.p. 133~134°. Yield, 0.2 g. after being recrystallized from methanol-water mixture (1 : 1). *Anal.* Calcd. for $C_{11}H_{10}O_5N_2S$: C, 46.80; H, 3.50; N, 9.93. Found: C, 46.25; H, 3.48; N, 10.26.

2-(5-Nitro)-furylsulfonic *p*-anisidide (VI)—A mixture of 0.5 g. of (I) and 0.5 g. of *p*-anisidine, dissolved in 10 cc. of dry acetone is heated gently in a water bath for 1 hour. After letting it stand overnight, the acetone evaporates to a brownish oil, which gradually solidifies. It is dried on a porcelain plate and purified by recrystallization from ethanol-water mixture (1 : 1) to 0.2 g. of thin brown crystals, m.p. 178~179°(decomp.). *Anal.* Calcd. for $C_{11}H_{10}O_6N_2S$: C, 44.00; H, 4.00; N, 9.33. Found: C, 43.39; H, 4.33; N, 9.47.

2-(5-Nitro)-furylsulfonic *p*-phenetidide (VII)—The same procedure as used above is utilized for the preparation of this compound. From 0.5 g. of (I) and 0.5 g. of phenetidine, 0.3 g. of thin yellow scaly crystals, m.p. 175°(decomp.), are obtained after being recrystallized from ethanol. *Anal.* Calcd. for $C_{12}H_{12}O_2N_6S$: N, 8.97. Found: N, 8.40.

2-(5-Nitro)-furylsulfonic *p*-ethylaminobenzoate (VIII)—0.5 g. of (I) and 0.3 g. of ethyl *p*-aminobenzoate are treated by the same procedure as described above and recrystallized from methanol to give 0.3 g. of thin brown plate crystals, m.p. 163°. *Anal.* Calcd. for $C_{13}H_{12}O_7N_2S$: N, 8.23. Found: N, 7.96.

2-(5-Nitro)-furylsulfonyl α-naphthylamide (IX)—Utilizing the same procedure as above, from 0.5 g. of (I) and 0.3 g. of α-naphthylamine is obtained 0.5 g. of colorless plate crystals, m.p. 223°(decomp.), after recrystallization from ethanol. *Anal.* Calcd. for $C_{14}H_{10}O_5N_2S$: N, 8.80. Found: N, 8.63.

2-(5-Nitro)-furylsulfonyl-5′-methylsulfathiazole-2′—0.5 g. of (I) and 0.3 g. of 5′-methylsulfathiazole-2′ produces 0.4 g. of light yellow granular crystals, m.p. 180°(decomp.), after being recrystallized from ethanol. *Anal.* Calcd. for $C_8H_7O_5N_3S$: N, 14.52. Found: N, 14.23.

Summary

Taking nitrofuran sulfochloride as the starting material, several nitrofurylsulfonic acid derivatives (easily soluble in water) were synthesized.

(Received September 7, 1953)