New Compounds: Derivatives of Fluorene XXXVII: 9-Substituted 3-Nitrofluorenes

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Abstract □ New 9-substituted 3-nitrofluorenes were prepared as potential intermediates in a study of modified electrophilicity and mutagenicity of the carcinogen 3-*N,O*-diacetylhydroxylaminofluorene. The reported derivatives, together with some already known 9-substituted 3-nitrofluorenes, were too unstable to survive the reducing conditions required to transform the nitro group to the corresponding hydroxylamine.

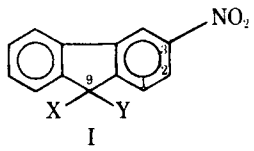
Keyphrases □ Fluorenes—synthesis of 3-nitro-9-substituted derivatives, potential enhancement of 3-*N,O*-diacetylhydroxylaminofluorene carcinogenicity □ 3-*N,O*-Diacetylhydroxylaminofluorene—synthesis of 9-substituted 3-nitrofluorene intermediates, effect on carcinogenicity □ Carcinogens—synthesis of 9-substituted 3-nitrofluorene intermediates for study of 3-*N,O*-diacetylhydroxylaminofluorene mutagenicity

Oncogenic arylamines and amides generally are believed to exert their baneful effects by undergoing enzymatic oxidation to an *N*-hydroxy derivative (a proximate carcinogen) and then by further reaction to form an ester such as the acetate or sulfate (1). Many of these esters are ultimate carcinogens, and they cleave heterolytically, with differing facility, at the nitrogen–oxygen bond. The electron-deficient nitrogen atom is left as an electrophilic center (acetamidonium ion). Such moieties are capable of an *in vivo* attack on various nucleophiles in tissue constituents such as DNA, RNA, and proteins. Some of these reactions appear to be crucial in carcinogenesis.

DISCUSSION

Much of the knowledge supporting the foregoing concepts was developed from studies on 2-fluorenamine and *N*-acetyl-2-fluorenamine, a strong hepatocarcinogen (1). A closely related isomer, *N*-acetyl-3-fluorenamine, is of interest because its *N*-acetoxy derivative, 3-*N,O*-diacetylhydroxylaminofluorene, is a much weaker carcinogen (mammary) and is not hepatocarcinogenic (2). It is possible that one major reason for this difference in biological activity between the 2-substituted and 3-substituted compounds is simply the lower tendency of the acetoxy group of the latter to act as a leaving group and the lower electrophilicity of the resulting acetamidonium ion.

Accordingly, 3-nitrofluorene (I) derivatives were designed with various 9-substituents to increase the leaving group facility of the acetoxy group and, after reduction and acetylation to the *N*-acetoxyamide, to produce a compound with enhanced mutagenicity and carcinogenicity. The presence of an electron donor group in the single-ring compound *O*-acetyl-*N*-benzoylphenylhydroxylamine was shown to enhance heterolytic acyloxy migration to a nucleophilic carbon (3). 2-Methoxy-3-*N,O*-diacetylhydroxylaminofluorene showed slightly enhanced mutagenicity over that of 3-*N,O*-diacetylhydroxylaminofluorene[1]. Since it was assumed that the effect sought was lessened by steric hindrance and since other methoxylated 3-nitrofluorenes were not readily available, the 9-position was investigated.



I

Ia: X = H, Y = Br
Ib: X = H, Y = OCH_3
Ic: X = H, Y = OC_2H_5
Id: X = H, Y = $P^+(C_6H_5)_3Br^-$
Ie: X = H, Y = $SC(NH_2)=N^+H_2Br^-$
If: XY = $=NNHSO_2C_6H_4CH_3$-*p*

Unfortunately, *Ia–If* were either too unstable for the reducing conditions required to transform the nitro group to the corresponding hydroxylamine (ammonium sulfide in ethanol–dimethylformamide) (4) or gave relatively intractable mixtures in the workup. Since these compounds may be of interest in other areas and since known 3,9-disubstituted fluorenes are comparatively few, this report is pertinent. Ways of stabilizing the 9-position are being studied and will be the subject of a future article.

Attempts to make 9-methoxy-3-nitrofluorene (*Ib*) from 3-nitrofluorene-9-ol (5) with methyl iodide, dimethylformamide, and silver oxide (6) or with trimethylsulfoxonium iodide and silver oxide (7) led to recovery of 3-nitrofluorenone as the only recognizable product.

Attempts to make an ylid from *Id* (8) failed, as did efforts to produce 9-mercapto-3-nitrofluorene from *Ie* (9). Attempts to prepare 9-fluoro- or 9-cyano-3-nitrofluorene did not yield the desired products. Previously known 9-chloro-3-nitrofluorene and 9-acetoxy-3-nitrofluorene were included in the nitro reduction attempts.

EXPERIMENTAL[2]

9-Bromo-3-nitrofluorene (Ia)—3-Nitrofluoren-9-ol (7.5 g, 0.033 mole), prepared by sodium borohydride reduction of 3-nitrofluoren-9-one (5), was mixed with 100% acetic acid (120 ml). Aqueous 48% HBr (16 ml) was added to the stirred mixture all at once. The mixture was heated with stirring to boiling, and the solvent was distilled off. The residue was allowed to cool to room temperature and then was dissolved in hot benzene. The benzene solution was treated with activated charcoal[3], filtered, and concentrated to a small volume. Addition of petroleum ether (bp 30–60°) caused the product to crystallize as white needles, 9.5 g (99%), mp 154.5–156°. Recrystallization from benzene–petroleum ether (bp 30–60°) gave 9.1 g, mp 157–158°.

Anal.—Calc. for $C_{13}H_8BrNO_2$: C, 53.82; H, 2.78; Br, 27.54; N, 4.83. Found: C, 53.81; H, 2.81; Br, 27.75; N, 4.86.

9-Methoxy-3-nitrofluorene (Ib)—9-Chloro-3-nitrofluorene (10) (1 g, 0.004 mole) was mixed with powdered silver nitrate (0.7 g, 0.004 mole) and absolute methanol (40 ml). The mixture was refluxed under dry nitrogen for 2 hr and cooled to room temperature. The suspension was filtered into a slurry of crushed ice and water. The crystallized product in the ice slurry then was collected, washed with water, and dried over concentrated sulfuric acid *in vacuo* to give 0.9 g. Chromatography on silica gel and recrystallization from methanol–water gave a pure product, 0.45 g (47%), mp 103.5–104.5°; IR (mineral oil): 1060 cm^{-1}.

Anal.—Calc. for $C_{14}H_{11}NO_3$: C, 69.70; H, 4.60; N, 5.81. Found: C, 69.87; H, 4.37; N, 5:62.

9-Ethoxy-3-nitrofluorene (Ic)—In a manner similar to that for *Ib*, equimolar amounts of 9-chloro-3-nitrofluorene and silver nitrate in refluxing absolute ethanol gave crude *Ic*. Chromatography and recrystallization from ethanol–water gave a 40% yield of pure product, mp 88–89°; IR (mineral oil): 1060 cm^{-1}.

Anal.—Calc. for $C_{15}H_{13}NO_3$: C, 70.58; H, 5.13; N, 5.49. Found: C, 70.52; H, 5.01; N, 5.42.

9-(3-Nitrofluorenyl)triphenylphosphonium Bromide (Id)—To a stirred solution of *Ia* (1.5 g, 0.005 mole) in benzene (50 ml) was added triphenylphosphine (1.3 g, 0.005 mole) in one portion. The mixture was

[1] H.-L. Pan, C.-A. Cole, and T. L. Fletcher, unpublished results.

[2] Melting points were determined on a Fisher-Johns apparatus and are corrected. IR spectra were obtained using a Beckman IR-33 spectrophotometer. Elemental analyses were performed by commercial analytical laboratories.

[3] Darco.

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stirred further in nitrogen for 1 hr and then refluxed for 0.5 hr and stirred to cool. After filtration from the reaction mixture, the gray solid (0.7 g, 25%) was washed twice with benzene and then with ether and dried *in vacuo*, mp 210–214° dec.

Anal.—Calc. for $C_{31}H_{23}BrNO_2P$: C, 67.40; H, 4.20; N, 2.54. Found: C, 67.27; H, 4.18; N, 2.64.

9-(3-Nitrofluorenyl)isothiuronium Bromide (Ie)—Compound Ia (2.9 g, 0.01 mole) and thiourea (0.76 g, 0.01 mole) were ground together thoroughly. To the mixture were added 1-butanol (8 ml) and aqueous 48% HBr (1 ml). The mixture was refluxed with occasional shaking for 0.5 hr and cooled. The crystalline product was collected, washed successively with small amounts of 1-butanol, carbon tetrachloride, chloroform, and ether, and air dried, giving 3.45 g (93%), mp 210–215° dec.

Anal.—Calc. for $C_{14}H_{12}BrN_3O_2S$: C, 45.91; H, 3.30; N, 11.47; S, 8.75. Found: C, 45.77; H, 3.22; N, 11.28; S, 8.76.

3-Nitrofluoren-9-*p*-toluenesulfonylhydrazone (If)—*p*-Toluenesulfonylhydrazine (11) (15.4 g, 0.083 mole) and 3-nitrofluoren-9-one (15 g, 0.067 mole) were suspended in absolute ethanol (150 ml). The suspension was heated with stirring under reflux for 1.5 hr and cooled. The brown crystalline product was collected, washed with 100 ml of boiling ethanol, and dried, giving 20.5 g (78%), mp 194–197° dec.

The product (0.5 g) was purified by repeated washing with boiling ethanol and recrystallization from chloroform–benzene to give lustrous orange crystals, 0.3 g, mp 205–206° dec.; IR (mineral oil): 3230, 1583, 1515, 1365, 1330, and 1150 cm^{-1}.

Anal.—Calc. for $C_{20}H_{15}N_3O_4S$: C, 61.06; H, 3.84; N, 10.68. Found: C, 60.94; H, 3.93; N, 10.24.

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COMMUNICATIONS

Decarboxylation Kinetics of 5-(Tetradecyloxy)-2-furoic Acid

Keyphrases □ Decomposition—solid-state reactions, decarboxylation kinetics, hypolipidemic agent, 5-(tetradecyloxy)-2-furoic acid □ Kinetics—solid-state activation energy, decarboxylation, decomposition, 5-(tetradecyloxy)-2-furoic acid □ 2-Furoic acid, 5-tetradecyloxy—hypolipidemic agent, solid-state decomposition, kinetics

To the Editor:

Single-component solid reactions are of several types: *e.g.*, a solid can form a solid and a gas (type I) or a liquid and a gas (type II). A type I reaction is exemplified by oxygen formation from permanganates, whereas the formation of liquid aniline and gaseous carbon dioxide from decarboxylation of *p*-aminobenzoic acid denotes a type II reaction. This paper reports a type II reaction of a pharmaceutical solid, 5-(tetradecyloxy)-2-furoic acid[1], which is a novel hypolipidemic agent (1).

Decarboxylation kinetics in the solid state have been reported (2–7), and several reported reactions (3, 5, 6) were type II. This paper deals with carboxylic acid, RCOOH, where R = $C_{14}H_{29}O$–(C_4H_2O)– and where (C_4H_2O)– is a furan moiety. The compound decomposes *via* the reaction shown in Scheme I, where RH is liquid in the temperature range studied; hence, the reaction is type II.

$$RCOOH \rightarrow RH + CO_2$$

Scheme I

An apparatus such as the one described by Carstensen and Musa (Fig. 6 of Ref. 3) was constructed for the studies at each temperature. The assembly was completely tight since it was fused by glass blowing and sealed under vacuum (<0.1 μm Hg). The entire assembly was placed in a thermostated mineral oil bath or, for lower temperatures, in a water bath. The pressure evolution was followed as a function of time with a cathetometer[2]. All pertinent volumes were determined by water weight calibration. With knowledge of the volumes, the pressures were converted to moles of gas, thus yielding a decomposition *versus* time curve (Fig. 1). The gas evolution eventually accounted for 100% decomposition.

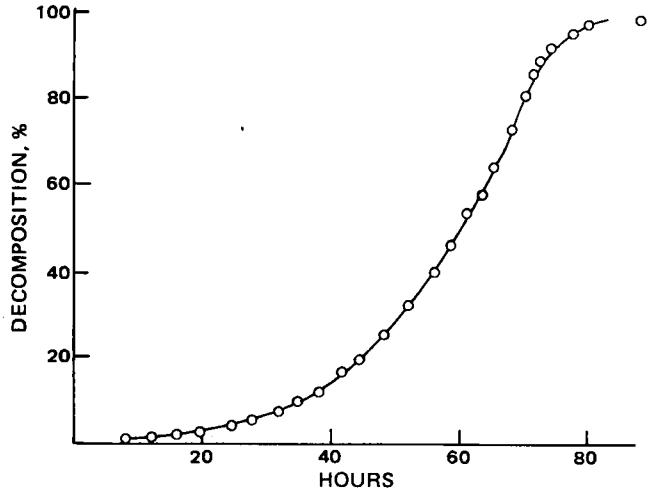

Figure 1—*Percent of parent compound decomposed as a function of time at 90°.*

[1] This agent is known as RMI 14514 and was supplied by R. A. Parker and M. A. Zoglio, Merrell National Laboratories, Cincinnati, OH 45215.

[2] Model M-911, Gaertner Scientific Corp., Chicago, IL 60614.

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