[CONTRIBUTION FROM THE CANCER RESEARCH LABORATORY OF THE UNIVERSITY OF FLORIDA]

2-ACETYL-7-AMINOFLUORENE AND ITS DERIVATIVES[1]

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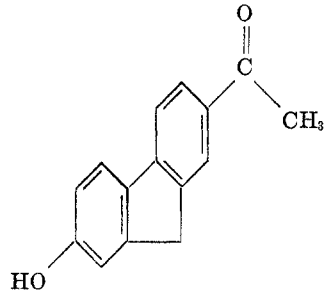
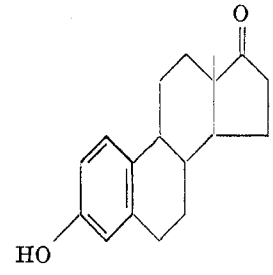
In a continuation of a study of the chemical, physical, and biological properties of carcinogenic amines and allied compounds, derivatives of 2-acetyl-7-aminofluorene and 2-acetyl-7-hydroxyfluorene were prepared.

2-Aminofluorene (1), 2-methylaminofluorene (2), 2-dimethylaminofluorene (2), 2-nitrofluorene (1), 2-acetylaminofluorene (1), 2-diacetylaminofluorene (1), N-(2-fluorenyl)hemisuccinamide (3), N-(2-fluorenyl)glycine (3), 7-fluoro-2-acetylaminofluorene (4), 9-hydroxy-2-acetylaminofluorene (4), 7-hydroxy-2-acetylaminofluorene (5), 2,7-bis-(acetylamino)fluorene (6), and 2-acetylaminofluorenone (4) have been shown to have cancer-producing activity in animals. For this reason these chemicals should be considered dangerous to any human in extended contact with them. To investigate this phenomenon further 2-acetyl-7-aminofluorene and N-substituted derivatives of the compound have been prepared.

Schueler (7, 8) has suggested that a given substance may be estrogenic if it is rather large, rigid, relatively inert, lipoid-soluble, and has two active hydrogen-bond forming groups located at an optimum distance of 14.5 Å units from one another. In this respect 2-acetyl-7-hydroxyfluorene (I) (which resembles estrone, II) or some derivative of I, could conceivably have estrogenic activity.



I



II

The spectra of 2-acetyl-7-hydroxyfluorene ($\lambda_{max.}$ 233; 331), 2-acetyl-7-aminofluorene ($\lambda_{max.}$ 243; 352), 2-acetyl-7-hydroxyfluorene anion ($\lambda_{max.}$ 252; 384) and 2-nitro-7-aminofluorene ($\lambda_{max.}$ 261; 400) (9) in Fig. 1 show the typical *p*-nitroaniline type of envelope. This consists of a narrow band of fairly strong intensity in the ultraviolet and a much more intense broad band at or near the visible end of the spectrum. Many phenols and anilines substituted in the *para* position with an electron-attracting group have this type of spectral envelope (10, 11).

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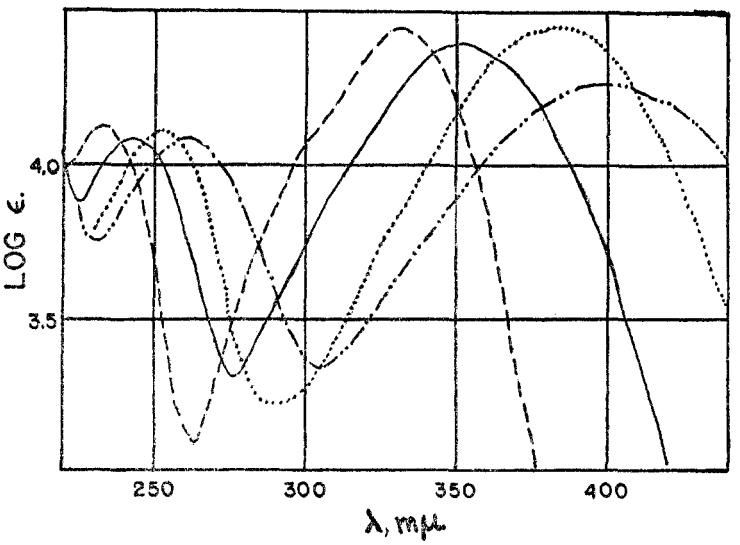

FIG. 1. ULTRAVIOLET ABSORPTION SPECTRA. 2-Acetyl-7-hydroxyfluorene in 95% ethanol (- - -); 2-Acetyl-7-aminofluorene in 95% ethanol (————); 2-Acetyl-7-hydroxyfluorene in 95% ethanol containing 10 ml. of 1 *N* aqueous potassium hydroxide per 100 ml. of solution (. . . .); and 2-Nitro-7-aminofluorene in 95% ethanol (—. .—. .).

The derivation of these two bands has been discussed at some length by Kumler (10) and Doub and Vandenbelt (11).

EXPERIMENTAL[2]

2-Acetyl-7-aminofluorene. To a thin paste of 25.3 g. of 2-acetyl-7-nitrofluorene (12) in 800 ml. of hot 95% ethanol was added a solution of 7 g. of calcium chloride in 180 ml. of water and 210 g. of zinc dust. The whole was thoroughly shaken, refluxed for four hours, and then filtered hot. The residue was extracted several times with boiling Methyl Cellosolve.[3] The combined filtrates were poured into 2 liters of water and 19 g. (85% yield) of yellow crystals were collected, m.p. 212–214°. Crystallization from alcohol gave light yellow lustrous crystals, m.p. 214–215°. A solution of the compound in alcohol has a green fluorescence in ultraviolet light.

Anal. Calc'd for $C_{15}H_{13}NO$: C, 80.7; H, 5.83; N, 6.28.
Found: C, 80.5; H, 5.75; N, 6.20.

2-Acetyl-7-acetylaminofluorene. This compound was obtained by the action of 1.0 ml. of acetic anhydride on a solution of 2.23 g. of 2-acetyl-7-aminofluorene in boiling xylene. Crystallization from dilute acetic acid gave 2.20 g. (83% yield) of light yellow needles, m.p. 257–258°.

Anal. Calc'd for $C_{17}H_{15}NO_2$: C, 77.0; H, 5.66; N, 5.28.
Found: C, 77.2; H, 5.51; N, 5.03.

2-Acetyl-7-trichloroacetylaminofluorene. To a warm solution of 0.22 g. of 2-acetyl-7-aminofluorene in 25 ml. of xylene and 0.08 ml. of pyridine was added 0.19 g. of trichloroacetyl chloride. Charcoal was added to the hot solution which was then filtered. Crystallization from aqueous ethanol gave 0.25 g. (68% yield) of colorless needles, m.p. 196–197°.

[2] Melting points are not corrected.

[3] Trade name for 2-methoxyethanol.

Anal. Calc'd for $C_{17}H_{12}Cl_3NO_2$: Cl, 29.0. Found: Cl, 29.9.

2-Acetyl-7-fluoroacetylaminofluorene. This compound was prepared by the same procedure used for the previous compound except that fluoroacetyl chloride was the acylating agent. Crystallization from xylene gave an 82% yield of faintly yellow crystals, m.p. 229–230°.

Anal. Calc'd for $C_{17}H_{14}FNO_2$: C, 72.1; H, 4.95.
Found: C, 71.9; H, 4.78.

2-Acetyl-7-diethylcarbamylaminofluorene or *N,N-diethyl-N'-(7-acetyl-2-fluorenyl)urea.* The same procedure was used as for the preceding compound except that diethyl carbamyl chloride[4] was the acylating agent. A 60% yield of yellow crystals was obtained, m.p. 225–227° (placed in bath at 220°).

Anal. Calc'd for $C_{20}H_{22}N_2O_2$: N, 8.70. Found: N, 8.50.

2-Acetyl-7-benzalaminofluorene. The reaction between benzaldehyde and a hot solution of 2-acetyl-7-aminofluorene in Methyl Cellosolve gave the Schiff base. Crystallization from Methyl Cellosolve gave a 90% yield of light yellow plates, m.p. 210–211°.

Anal. Calc'd for $C_{22}H_{17}NO$: C, 84.9; H, 5.47.
Found: C, 85.2; H, 5.28.

2-Acetyl-7-cinnamalaminofluorene. This compound was prepared by the reaction of cinnamaldehyde with a solution of 2-acetyl-7-aminofluorene in Methyl Cellosolve. Crystallization from Methyl Cellosolve gave an 89% yield of sulfur yellow plates, m.p. 203–204°.

Anal. Calc'd for $C_{24}H_{19}NO$: C, 85.46; H, 5.64.
Found: C, 85.23; H, 5.70.

2-Acetyl-7-(2'-hydroxybenzalamino)fluorene. The same procedure was followed as for the previous compound except that salicylaldehyde was used. Crystallization from alcohol gave a 95% yield of yellow-orange plates, m.p. 192–193°.

Anal. Calc'd for $C_{22}H_{17}NO_2$: C, 80.73; H, 5.19.
Found: C, 80.55; H, 5.32.

2-Acetyl-7-hydroxyfluorene. 2-Acetyl-7-aminofluorene (4.5 g.) was ground in a mortar with 6 ml. of conc'd hydrochloric acid. The product was washed into a 250-ml. flask with 130 ml. of water. The mixture was refluxed for a half hour and then cooled quickly to 0–10°. A solution of 1.5 g. of sodium nitrite in 9 ml. of water was added dropwise with stirring at the same temperature. The stirred mixture was warmed to 45° and kept at that temperature for 15 minutes. The filtered diazonium chloride solution was added dropwise through a funnel which extended below the surface of 360 ml. of boiling water containing 6 ml. of conc'd sulfuric acid. The mixture was boiled for 15 minutes after the addition and then cooled and filtered. The residue was extracted with boiling xylene (Darco). The xylene was evaporated to give 3.4 g. (76% yield) of crude product, m.p. 208–210°. Crystallization from aqueous ethanol gave 3.0 g. (67% yield) of cream-colored crystals, m.p. 213–214°. The compound has a blue fluorescence in alcohol solution under ultraviolet light.

Anal. Calc'd for $C_{15}H_{12}O_2$: C, 80.4; H, 5.36.
Found: C, 79.85; H, 5.51.

2-Acetyl-7-acetoxyfluorene. A solution of 0.22 g. of 2-acetyl-7-hydroxyfluorene and 0.11 g. of potassium acetate in 2 ml. of acetic anhydride was refluxed for 1 hour. The mixture was then poured into water. Crystallization from heptane gave 0.22 g. (85% yield) of cream-colored crystals, m.p. 126–127°.

Anal. Calc'd for $C_{17}H_{14}O_3$: C, 76.69; H, 5.26.
Found: C, 76.52; H, 5.24.

SUMMARY

The following new compounds of interest in chemical carcinogenesis studies have been prepared: 2-acetyl-7-aminofluorene, 2-acetyl-7-acetylaminofluorene, 2-acetyl-7-trichloroacetylaminofluorene, 2-acetyl-7-fluoroacetylaminofluorene, 2-

[4] Courtesy of Monsanto Chemical Co., St. Louis, Mo.

acetyl-7-diethylcarbamylaminofluorene, 2-acetyl-7-benzalaminofluorene, 2-acetyl-7-cinnamalaminofluorene, and 2-acetyl-7-(2′-hydroxybenzalamino)fluorene. The possibly estrogenic 2-acetyl-7-hydroxyfluorene and 2-acetyl-7-acetoxyfluorene have also been synthesized.

The ultraviolet absorption spectra of 2-nitro-7-aminofluorene, 2-acetyl-7-aminofluorene, and 2-acetyl-7-hydroxyfluorene and its anion were compared.

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