Synthesis and Mutagenicity of Dihydrodiol Metabolites of Benzo[*b*]fluoranthene[1,2]

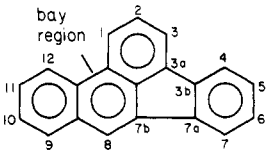
Shantu Amin,* Nalband Hussain, Harry Brielmann, and Stephen S. Hecht

Naylor Dana Institute for Disease Prevention, American Health Foundation, Valhalla, New York 10595

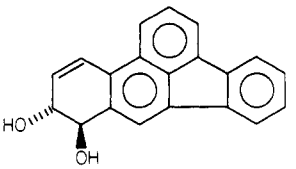
Received July 27, 1983

The syntheses of the two major hepatic microsomal dihydrodiol metabolites of the environmental carcinogen, benzo[*b*]fluoranthene, are described. 1,2-Dihydro-1,2-dihydroxybenzo[*b*]fluoranthene was prepared from 11*H*-benzo[*b*]fluorene-11-carboxylic acid. The key intermediate was 1-oxo-1,2,3,3a-tetrahydrobenzo[*b*]fluoranthene which was prepared by regiospecific cyclization of 11*H*-benzo[*b*]fluorene-11-propionic acid chloride. 11,12-Dihydro-11,12-dihydroxybenzo[*b*]fluoranthene was synthesized from 2-methylfluoranthene via 12-oxo-9,10,11,12-tetrahydrobenzo[*b*]fluoranthene. Both dihydrodiols were mutagenic toward *Salmonella typhimurium* TA 100, but their activities were less than that of benzo[*b*]fluoranthene.

Benzo[*b*]fluoranthene (**1**) is one of the most widely distributed of the carcinogenic polynuclear aromatic hydrocarbons.[3-5] Its tumor-initiating activity on mouse skin is less than that of benzo[*a*]pyrene, about equivalent to that of dibenz[*a*,*h*]anthracene, and greater than those of benz[*a*]anthracene and chrysene.[3,6-8] As a nonalternate hydrocarbon, benzo[*b*]fluoranthene might exhibit pathways of metabolic activation significantly different from those of benzo[*a*]pyrene and related alternate hydrocarbons. In previous studies, we have reported the synthesis, mutagenicity, and tumor-initiating activity of 9,10-dihydro-9,10-dihydroxybenzo[*b*]fluoranthene (**2**),



1

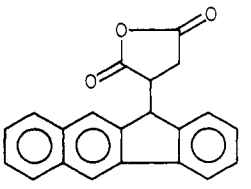


2

which could form a bay region dihydrodiol epoxide.[8-10] Compound **2** was about equipotent to benzo[*b*]fluoranthene as a mutagen and as a tumor initiator on mouse skin but has not yet been detected as a metabolite of benzo[*b*]fluoranthene.[8,9,11] The major dihydrodiol metabolites of benzo[*b*]fluoranthene, formed in vitro by rat liver 9000*g* supernatant, have been identified as 1,2-dihydro-1,2-dihydroxybenzo[*b*]fluoranthene (**3**) and 11,12-dihydro-11,12-dihydroxybenzo[*b*]fluoranthene (**4**).[11] In this report, we describe the syntheses of these metabolites and their mutagenic activities toward *S. typhimurium*.

Results and Discussion

The synthesis of **3** (Scheme I) began with 11-carboxy-11*H*-benzo[*b*]fluorene (**5**), which was esterified and then converted to the carboxyethyl derivative **7** in high yield. We used methyl acrylate instead of acrylonitrile in this reaction because the resulting diester is more easily hydrolyzed and decarboxylated to the desired acid **8** than is the nitrile derivative. Spectral data for **8** and the corresponding methyl ester **9** were consistent with the assigned structures. The key step in the synthesis of **3** was the regiospecific cyclization of **8**, as its acid chloride, to the cyclic ketone **10**. The structure of **10** was deduced from its NMR spectrum in which H_{12} appeared as a downfield multiplet due to the deshielding effect of the carbonyl group. No such deshielding would be expected if **8** had cyclized in the opposite direction, yielding 3-oxo-12b,1,2,3-tetrahydrobenzo[*k*]fluoranthene. Additional evidence for the structure of **10** was obtained by reduction, dehydration, and aromatization which yielded exclusively benzo[*b*]fluoranthene; no benzo[*k*]fluoranthene was detected. We are aware of only one previous report on the preferred cyclization of an 11*H*-benzo[*b*]fluorene derivative to a benzo[*b*]fluoranthene. In his book, Clar briefly noted the formation of benzo[*b*]fluoranthene upon treatment of **15** with $ZnCl_2$.[12]



15

Reduction of **10** to **11** was initially attempted with $LiAlH_4$ which we have used successfully for analogous reductions in the benzofluoranthene series.[10] However, we did not obtain **11** by this route, possibly due to the acidity of the 3a proton of **10**. In contrast, reduction of **10** with $NaBH_4$ proceeded smoothly to give **11** in good yield. Dehydration of **11** with *p*-toluenesulfonic acid produced the olefin **12** in 70% yield. Lower yields were obtained when we used H_2SO_4 and acetic acid, a system which was effective in the dehydration of related compounds.[10]

The dibenzoate **13** was prepared by the Prévost reaction, which is a standard method in the synthesis of *trans*-dihydrodiols of polynuclear aromatic hydrocarbons.[13,14]

(1) A Study of Chemical Carcinogenesis, 57.

(2) Supported by Grant No. ES-02030 from the National Institute of Environmental Health Sciences and Contract NO1-CP-15747 from the National Cancer Institute.

(3) "International Agency for Research on Cancer Monographs"; International Agency for Research on Cancer: Lyon, 1973; Vol 3, p 69.

(4) Hoffmann, D., Wynder, E. L. In "Air Pollution", 3rd ed.; Stern, A. C., Ed.; Academic Press: New York, 1977; pp 361–455.

(5) Grimmer, G.; Naujack, K. W.; Schneider, D. In "Polynuclear Aromatic Hydrocarbons. Chemistry and Biological Effects" Bjorseth, A., Dennis, A. J., Eds.; Battelle Press: Columbus, OH, 1980; pp 107–125.

(6) Wynder, E. L.; Hoffmann, D. *Cancer* **1959**, *12*, 1194–1199.

(7) Habs, M.; Schmähl, D.; Misfeld, J. *Arch. Geschwulstforsch.* **1980**, *50*, 266–274.

(8) LaVoie, E. J.; Amin, S.; Hecht, S. S.; Furuya, K.; Hoffmann, D. *Carcinogenesis* **1982**, *3*, 49–52.

(9) Hecht, S. S.; LaVoie, E.; Amin, S.; Bedenko, V.; Hoffmann, D. In "Polynuclear Aromatic Hydrocarbons: Chemistry and Biological Effects"; Bjorseth, A., Dennis, A. J., Eds.; Battelle Press: Columbus, OH, 1980; pp 417–433.

(10) Amin, S.; Bedenko, V.; LaVoie, E.; Hecht, S. S.; Hoffmann, D. *J. Org. Chem.* **1981**, *46*, 2573–2578.

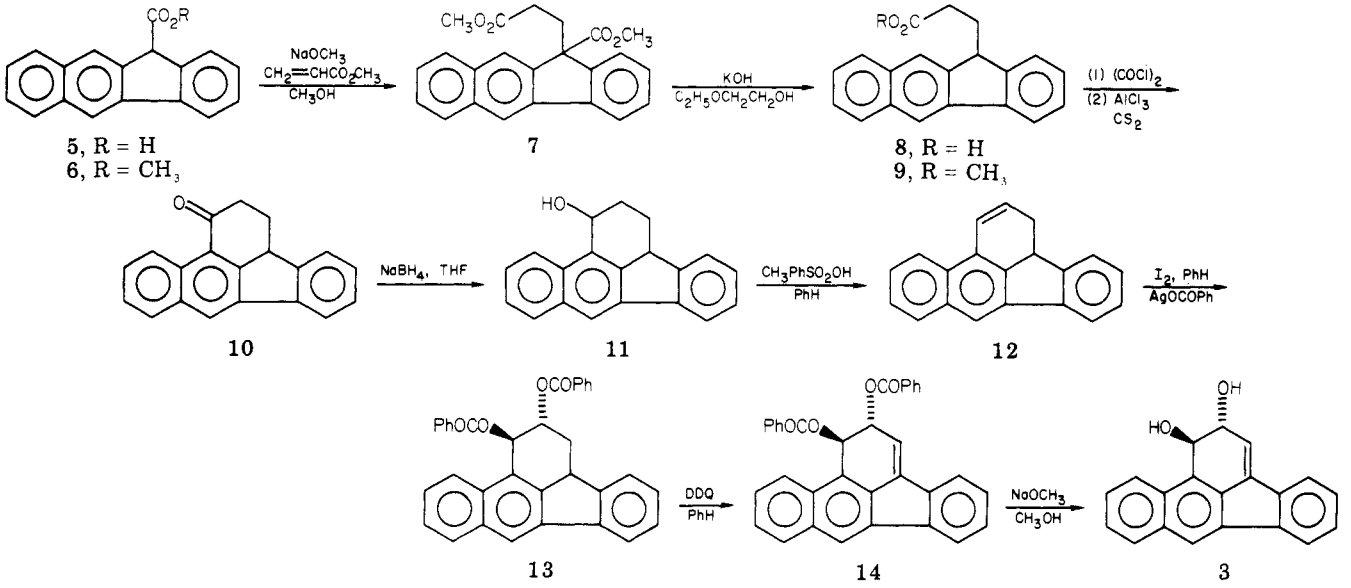
(11) Amin, S., LaVoie, E.; Hecht, S. S. *Carcinogenesis* **1982**, *3*, 171–174.

(12) Clar, E. "Polycyclic Hydrocarbons"; Academic Press: New York, 1964; Vol 1, p 191.

(13) McCaustland, D. J.; Engel, J. F. *Tetrahedron Lett.* **1975**, 2549–2552.

0022-3263/84/1949-1091$01.50/0

Scheme I



5, R = H
6, R = CH_3

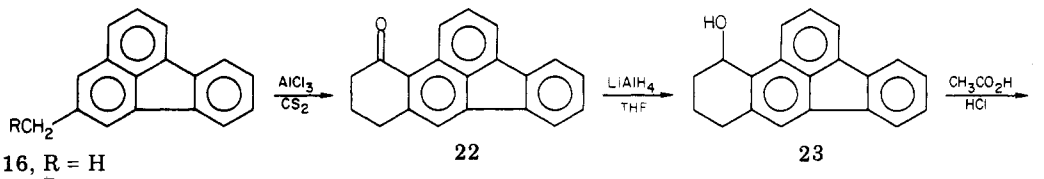
7

8, R = H
9, R = CH_3

10 11 12

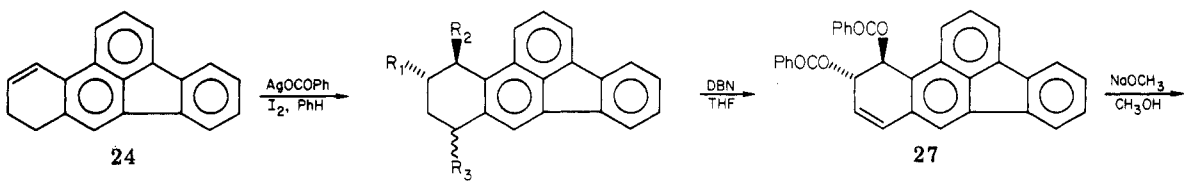
13 14 3

Scheme II



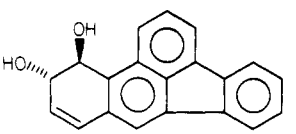
16, R = H
17, R = Br
18, R = $CH_2CH{=}CH_2$
19, R = $CH_2CH_2CH_2OH$
20, R = CH_2CH_2CHO
21, R = $CH_2CH_2CO_2H$

22 23



24 27

25, R_1 = R_2 = OCOPh; R_3 = H
26, R_1 = R_2 = OCOPh; R_3 = Br



4

Introduction of the double bond by the bromination–dehydrobromination procedure was not successful for **13**, giving aromatized products instead. However, treatment of **13** with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) gave **14** in 80% yield. Hydrolysis of **14** afforded the *trans*-dihydrodiol **3**.

The synthesis of 11,12-dihydro-11,12-dihydroxybenzo[*b*]fluoranthene (**4**) began with 2-methylfluoranthene (Scheme II). We employed the annelation procedure which we have previously used for the preparation of 9-oxo-9,10,11,12-tetrahydrobenzo[*j*]fluoranthene from 7-methylfluoranthene.[10] For the initial bromination step, it was necessary to use highly purified 2-methylfluoranthene. Small amounts of impurities led to erratic yields of **17**. Coupling, hydroboration, and a two-step oxidation procedure gave the desired butyric acid derivative **21**. Cyclization of the acid chloride of **21** proceeded specifically to ketone **22**, the structure of which was established by the presence in its NMR spectrum of the downfield H_1 proton and by conversion to benzo[*b*]fluoranthene. We did not observe any cyclization of **21** to the corresponding benzo[*a*]fluoranthene derivative.

Reduction of **22** with $LiAlH_4$ proceeded smoothly, in contrast to our results with **10**. Since **22** has no proton available at the 3a position, this supports our hypothesis that the relatively acidic 3a proton may have interfered with the reduction of **10**. Dehydration of **23** with HCl and acetic acid afforded **24** in good yield. Conversion of **24** to **25** by the Prévost reaction was also satisfactory. Introduction of the double bond by the bromination–dehydrobromination procedure gave **27** but only in 30% overall yield from **25**. The DDQ procedure, which was successful

(14) Harvey, R. G.; Fu, P. P. In "Polycyclic Hydrocarbons and Cancer"; Gelboin, H. V., T'so, P. O. P., Eds.; Academic Press: New York, 1978; Vol 1, pp 133–165.

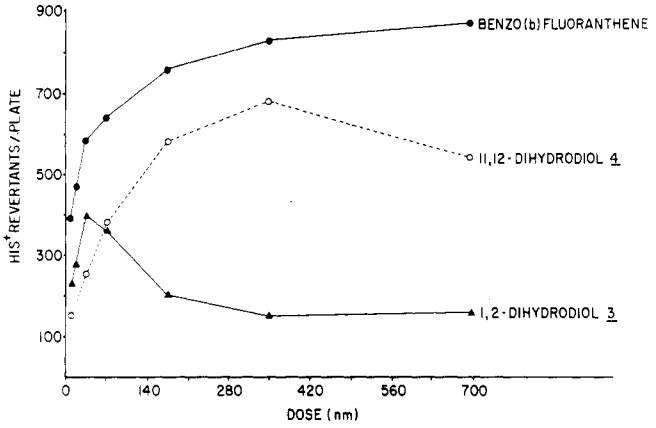


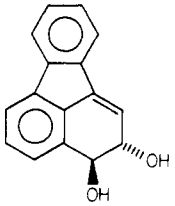
Figure 1. Mutagenicity toward *S. typhimurium* TA100 of benzo[*b*]fluoranthene and dihydrodiols **3** and **4** in the presence of 9000g supernatant from the livers of rats pretreated with Aroclor 1254. At doses above 70 nmol, **3** exhibited significant toxicity. See text for details.

for the conversion of **13** to **14**, failed to give satisfactory yields of **27**. This may have been due to relatively facile removal of the 3a hydride of **13**. Hydrolysis of **27** gave the *trans*-dihydrodiol **4**, contaminated with some *cis*-**4**. The latter was separated from *trans*-dihydrodiol **4** by column chromatography.

The NMR spectra of dihydrodiols **3** and **4** were in agreement with the assigned structures. The coupling constants $J_{1,2}$ for **3** and $J_{11,12}$ for **4** were small, as expected for the diequatorial protons of diaxial dihydrodiols in which the hydroxyl groups are situated in the bay region.[15]

Benzo[*b*]fluoranthene and dihydrodiols **3** and **4** were assayed for mutagenicity toward *S. typhimurium* strains TA 98 and TA 100, with activation by the 9000*g* supernatant from the liver of Aroclor pretreated rats. None of the compounds was active in TA 98. The results of the assays in TA 100 are illustrated in Figure 1. Both dihydrodiols were mutagenic although their activities were lower than that of benzo[*b*]fluoranthene. However under the conditions of the assay dihydrodiol **3** also exhibited bactericidal or bacteriostatic activity; at doses of 70 nmol and 350 nmol, bacterial survival rates were 65% and 8%, respectively. At the same doses, survival rates were 79% and 67% for the bacteria treated with **4**. No significant toxicity was observed for benzo[*b*]fluoranthene.

The mutagenicity of **3** appears to be lower than that of 2,3-dihydro-2,3-dihydroxyfluoranthene (**28**); 42 nmol of the latter induced 995 revertants/plate under similar conditions.[16] The mutagenicity of **28** can be attributed to



28

formation of the corresponding dihydrodiol epoxide, a process which is likely inhibited in **3** by the diaxial hydroxyl groups.[17,18] The lower mutagenic activities of **3** and **4** than of the parent hydrocarbon are typical of dihydrodiols in which the hydroxyl groups are situated in the bay region.[19] Since **4** is the major dihydrodiol metabolite of benzo[*b*]fluoranthene formed under the conditions of the mutagenicity assay,[11] it does contribute to the mutagenicity of benzo[*b*]fluoranthene. However, the levels of formation and mutagenic activities of dihydrodiols **2–4** do not seem to account for the mutagenicity of benzo[*b*]fluoranthene. This indicates that other metabolites such as the epoxide precursors to these dihydrodiols are likely to be involved.

Experimental Section

Infrared spectra were run on a Perkin-Elmer Model 267 grating infrared spectrophotometer. NMR spectra were determined with a Hitachi Perkin-Elmer R-24 spectrometer and a Jeol Model FX90Q spectrometer in $CDCl_3$ solution unless otherwise stated and are reported as parts per million downfield from Me_4Si as an internal reference. UV spectra were determined with a Cary Model 118 instrument. HPLC was carried out with a Waters Associates Model ALC/GPC-204 high-speed liquid chromatograph equipped with a Model 660 solvent programmer, a Model LC-25 UV/visible detector, and a 4.6 mm (ID) × 250 mm EM Lichrosorb RP-18, 10 μm column eluted from 50% CH_3OH in H_2O to 100% CH_3OH in 1 h at 2 mL/min. Mass spectra were run with a Hewlett-Packard Model 5982A dual source instrument using a membrane separator. Microanalyses were performed by Galbraith Laboratories.

Methyl 11*H*-Benzo[*b*]fluorene-11-carboxylate (6). Dry HCl gas was bubbled into a suspension of 11*H*-benzo[*b*]-fluorene-11-carboxylic acid[20] (**5**, 40 g, 0.151 mol) in 300 mL of MeOH until a clear solution was obtained; stirring was then continued at room temperature for 1 h. The product (41.4 g, 99%) separated as a white solid which was collected by filtration: mp 121–122 °C (MeOH) [lit.[20], mp 117–118 °C]; IR (Nujol) 1735 cm^{-1}; NMR δ 3.8 (s, 3 H), 5.1 (s, 1 H), 7.2–8.3 (m, 10 H); MS, *m/e* (relative intensity) 274 (M^+, 47), 215 (100).

Methyl 11-(Methoxycarbonyl)-11*H*-benzo[*b*]fluorene-11-propanoate (7). A mixture containing 5.0 g (0.018 mol) of **6**, 0.12 g (0.022 mol) of sodium methoxide, and 1.9 g (0.022 mol) of methyl acrylate in 250 mL of dry MeOH was stirred at 0 °C for 3 h, after which the solution became clear and then **7** separated as a white solid which was collected by filtration (6.0 g, 90%): mp 130–131 °C (MeOH); IR (Nujol) 1735 cm^{-1}; NMR δ 1.6–2.0 (m, 2 H), 2.7–3.0 (m, 2 H), 3.5 (s, 3 H), 3.7 (s, 3 H), 7.5–8.2 (m, 9 H), 8.7 (s, 1 H); MS, *m/e* (relative intensity) 360 (M^+, 100), 301 (87.4), 241 (97.2).

Anal. Calcd for $C_{23}H_{20}O_4$: C, 76.66; H, 5.55. Found: C, 76.47; H, 5.48.

11*H*-Benzo[*b*]fluorene-11-propionic Acid (8). Compound **7** (1 g, 0.0027 mol), ethoxyethanol (10 mL), and 3 mL of 30% aqueous KOH were heated under reflux for 3 h. The mixture was cooled to room temperature and extracted with Et_2O (2 × 100 mL). The Et_2O layer was washed with H_2O, and the aqueous phase was cooled and acidified with concentrated HCl. Extraction with CH_2Cl_2 and standard workup gave 0.76 g (97.5%) of **8** as a white solid: mp 173–174 °C (MeOH) NMR δ 1.8–2.2 (m, 2 H), 2.3–2.7 (m, 2 H) 4.1–4.3 (m, 1 H), 7.3–8.2 (m, 10 H); MS, *m/e* (relative intensity) 288 (M^+, 52.9), 228 (97.7), 215 (100).

Anal. Calcd for $C_{20}H_{16}O_2$: C, 83.33; H, 5.55. Found: C, 83.07; H, 5.54.

Methyl 11*H*-Benzo[*b*]fluorene-11-propanoate (9). A suspension of **8** (0.29 g, 0.001 mol) in absolute MeOH (5 mL) was stirred at 0 °C and HCl gas was bubbled through the suspension until a clear solution was obtained. After 20 min, the product

(15) Lehr, R. E.; Schaeffer-Ridder, M.; Jerina, D. M. *J. Org. Chem.* **1977**, *42*, 736–744.
(16) LaVoie, E. J.; Hecht, S. S.; Bedenko, V.; Hoffmann, D. *Carcinogenesis* **1982**, *3*, 841–846.
(17) Rastetter, W. H.; Nachbar, R. M., Jr.; Russo-Rodriguez, S.; Wattley, R. V.; Thilly, W. G.; Andon, B. M.; Jorgensen, W. L.; Ibrahim, M. *J. Org. Chem.* **1982**, *47*, 4873–4878.
(18) Fu, P. P.; Beland, F. A.; Yang, S. K. In "Polynuclear Aromatic Hydrocarbons: Chemical Analysis and Biological Fate"; Cooke, M., Dennis, A. J., Eds.; Battelle Press: Columbus, OH, 1981; pp 655–666.
(19) Wood, A. W.; Levin, W.; Chang, R. L.; Yagi, H.; Thakker, D. R.; Lehr, R. E.; Jerina, D. M.; Conney, A. H. In "Polynuclear Aromatic Hydrocarbons"; Jones, P. W., Leber, P., Eds.; Ann Arbor Science Publishers, Inc.: Ann Arbor, MI, 1978; pp 531–551.
(20) Collins, C. J., Burr, J. G., Jr.; Hess, D. N. *J. Am. Chem. Soc.* **1951**, *73*, 5176–5178.

(0.24 g, 80%) separated as a white crystalline material which was collected by filtration: mp 174–176 °C; IR (Nujol) 1740 cm^{-1}; NMR δ 1.6–2.2 (m, 2 H), 2.7–3.0 (m, 2 H), 4.2 (m, 1 H), 3.7 (s, 3 H), 7.5–8.2 (m, 9 H), 8.3 (s, 1 H); MS, *m/e* (relative intensity) 302 (M^+, 25.8), 228 (100), 215 (45.2).

1-Oxo-1,2,3,3a-tetrahydrobenzo[*b*]fluoranthene (10). Oxalyl chloride (8.3 g, 0.065 mol) was added dropwise to a suspension of the acid **8** (12.5 g, 0.043 mol) in dry CH_2Cl_2 (300 mL). The mixture was stirred for 3 h at room temperature and then the solvent was removed. The crude product was dissolved in 150 mL of CS_2 and the solution was cooled to 0 °C under an N_2 atmosphere. $AlCl_3$ (8.7 g, 0.065 mol) was added in portions, and then the mixture was stirred for 1 h at 0 °C and then heated under reflux for 30 min. The dark solution was then poured into ice and the complex was decomposed by adding 100 mL of dilute HCl. The resulting mixture was extracted with $CHCl_3$; the organic layer was washed with H_2O, dried ($MgSO_4$), and concentrated to give 8.7 g (75%) of **10**: mp 174–175 °C; NMR δ 1.5–2.1 (m, 1 H), 2.5–3.15 (m, 3 H), 3.9 (m, 1 H), 7.2–8.0 (m, 7 H), 8.1 (s, 1 H), 9.3–9.6 (m, 1 H); MS, *m/e* (relative intensity) 270 (M^+, 77.9), 242 (35.5), 228 (100).

Anal. Calcd for $C_{20}H_{14}O$: C, 88.88; H, 5.18. Found: C, 89.02; H, 5.12.

1-Hydroxy-1,2,3,3a-tetrahydrobenzo[*b*]fluoranthene (11). A solution of the ketone **10** (8.1 g, 0.03 mol) in THF (50 mL) and methanol (50 mL) was stirred with $NaBH_4$ (2 g, 0.06 mol) at ambient temperature for 30 min. The reaction mixture was then poured into ice. The mixture was extracted with CH_2Cl_2 (3 × 150 mL), washed with H_2O, and dried ($MgSO_4$). The usual workup gave **11** as a white solid (6.4 g, 79%), mp 171–172 °C. This was employed directly for the synthesis of **12**. MS, *m/e* (relative intensity) 272 (M^+, 100), 254 (100).

3,4-Dihydrobenzo[*b*]fluoranthene (12). A solution of **11** (5.5 g, 0.02 mol) and *p*-toluenesulfonic acid (10 mg) in benzene was heated at reflux for 30 min while using a Dean-Stark apparatus. Conventional workup followed by chromatography on silica gel and elution with hexane/CH_2Cl_2 (4/1) afforded **12** as a white solid (3.6 g, 70%): mp 118–119 °C; NMR δ 1.9–3.1 (m, 2 H), 4.1 (dd, 1 H, H_{3a}), 6.0–6.4 (m, 1 H), 7.1–8.1 (m, 10 H); MS, *m/e* (relative intensity) 254 (M^+, 100), 239 (35).

Anal. Calcd for $C_{20}H_{14}$: C, 94.48; H, 5.51. Found: C, 94.50; H, 5.42.

Aromatization of 12. A solution of olefin **12** (0.254 g, 0.001 mol) and DDQ (0.23 g, 0.001 mol) in dry benzene was heated at reflux for 10 min. The reaction mixture was worked up in the usual manner. The crude product was chromatographed on 20 g of silica gel with elution by Et_2O/hexane (1/1) to afford benzo(*b*)fluoranthene (0.2 g, 79%) as a white solid, mp 166 °C [lit.[21] 168 °C]. The UV of this compound was identical with that of a reference sample of benzo(*b*)fluoranthene.

***trans*-1,2-Bis(benzoyloxy)-1,2,3,3a-tetrahydrobenzo[*b*]fluoranthene (13).** A mixture of silver benzoate (5.5 g, 0.022 mol) and I_2 (2.65 g, 0.01 mol) in dry benzene (100 mL) was stirred under reflux until the red color disappeared. A solution of **12** (2.5 g, 0.01 mol) in 25 mL of dry benzene was added and the resulting mixture was heated under reflux for 3 h. The product was filtered hot and washed with benzene (3 × 25 mL). The filtrate was concentrated to give a solid which was chromatographed on silica gel. Elution with hexane gave a mixture of hydrocarbons (0.3 g). Further elution with hexane/CH_2Cl_2 (30/70) gave **13** (3.0 g, 62%): mp 184–185 °C; NMR δ 1.6–1.8 (m, 1 H), 3.0–3.4 (m, 1 H), 4.1–4.4 (dd, 1 H, H_{3a}), 6.1–6.3 (m, 1 H, H_2), 7.2 (dd, 1 H, H_1, $J_{1,2}$ = 4.4 Hz), 7.3–7.7 (m, 12 H), 7.8–8.2 (m, 6 H), 8.3 (s, 1 H, H_8); MS, *m/e* (relative intensity) 496 (20), 374 (30).

Anal. Calcd for $C_{34}H_{24}O_4$: C, 82.25; H, 4.83. Found: C, 82.23; H, 4.65.

***trans*-1,2-Bis(benzoyloxy)-1,2-dihydrobenzo[*b*]fluoranthene (14).** A suspension of **13** (0.5 g, 0.001 mol) and DDQ (0.23 g, 0.001 mol) in dry benzene was heated at reflux for 10 min. The reaction mixture was applied to a short column of neutral alumina and then eluted quickly with hexane followed by hexane/CH_2Cl_2 (50/50). Removal of the solvent left a white solid (0.4 g, 80%) mainly dihydro compound **14**: mp 132–134 °C; NMR δ 6.2–6.4 (m, 1 H), 6.5–6.8 (m, 1 H), 7.0–8.3 (m, 20 H).

***trans*-1,2-Dihydro-1,2-dihydroxybenzo[*b*]fluoranthene (3).** To a solution of **14** (0.5 g, 0.001 mol) in dry THF (10 mL) was added NaOMe (0.11 g, 0.002 mol) in MeOH (10 mL) and the resulting mixture was stirred at 5 °C for 1 h. EtOAc (10 mL) was added and the organic phase was washed with H_2O (2 × 50 mL), brine (2 × 50 mL), filtered, and concentrated to give **3** as an off-white solid (0.2 g). This crude diol was chromatographed on Florisil with elution by CH_2Cl_2 to remove some hydroxybenzo(*b*)fluoranthene, then by EtOAc/CH_2Cl_2 (30/70) which first gave a mixture of *cis*- and *trans*-**3** and then pure **3** (0.1 g, 35%): mp 186–187 °C; 300 MHz NMR δ 4.7 (m, 1 H, H_2), 5.1 (br s, 1 H, OH), 5.26 (br s, 1 H, OH), 5.38 (d, 1 H, H_1, J < 2 Hz), 6.7 (d, 1 H, H_3, $J_{2,3}$ = 4.8 Hz), 7.35–7.55 (m, 4 H, $H_{5,6,10,11}$), 7.82 (d, 1 H, H_4 or H_7 or H_9), 7.85 (d, 1 H, H_4 or H_7 or H_9) 7.91 (d, 1 H, H_4 or H_7 or H_9), 8.09 (s, 1 H, H_8), 8.3 (d, 1 H, H_{12}, $J_{11,12}$ = 8.3 Hz); MS, *m/e* (relative intensity) 286 (M^+, 30.3), 268 (100); UV (MeOH) λ max 340 nm (ϵ 5940), 297 (41580), 286 (39108), 273 (33168), 252 (88118), 244 (72772); HPLC retention times *trans*-**3**, 14.6 min; *cis*-**3**, 18.1 min.

Anal. Calcd for $C_{20}H_{14}O_2$: C, 83.91; H, 4.89. Found: C, 83.73; H, 5.08.

2-(Bromomethyl)fluoranthene (17). Warning: Allergic reactions were noted among those working with compounds **17–21**, even when stringent precautions were taken to avoid exposure.

A suspension of *N*-bromosuccinimide (NBS, 3.6 g, 0.02 mol), 2-methylfluoranthene[22] (**16**) (4.3 g, 0.02 mol), and benzoyl peroxide (20 mg) in CCl_4 (150 mL) was heated at reflux with a sun lamp for 1.5 h. Conventional workup provided **17** (5.3 g, 90%) as a yellow solid: mp 118–119 °C; NMR δ 4.5 (s, 2 H), 7.0–7.8 (m, 9 H); MS, *m/e* (relative intensity) 296 (M^+, 13.8), 294 (13.8) 215 (100).

4-(2-Fluoranthenyl)-1-butene (18). Allyl magnesium bromide (1.88 g, 10 mL of 1.3 M, Ventron Corp) was added dropwise to a solution of **17** (2.8 g, 0.0095 mol) in 300 mL of Et_2O. After 8 h, the mixture was worked up as usual to yield 1.85 g of **18**, which was used directly in the next step: NMR δ 2.2–2.5 (m, 2 H), 2.6–2.8 (m, 2 H), 4.7–5.0 (m, 2 H), 5.6–5.8 (m, 1 H), 7.0–7.8 (m, 9 H); MS, *m/e* (relative intensity) 256 (M^+, 80), 215 (100).

4-(2-Fluoranthenyl)-1-butanol (19). To a solution of 11.0 g of **18** (0.044 mol) in 500 mL of dry THF was added 15.2 mL of a 1 M solution of diborane in THF over a 20-min period. The reaction mixture was stirred at room temperature for 12 h, cooled to 5 °C, and treated with 10 mL of H_2O dropwise. Sodium hydroxide (200 mL of a 10% solution) was added over 30 min followed by dropwise addition of 160 mL of 30% H_2O_2. The resulting solution was stirred for 2 h, then refluxed for 8 h, and then worked up in the usual manner to give 9.2 g of crude product, which was chromatographed on silica gel with elution by CH_2Cl_2 yielding 7.2 g (60%) of **19** as an oil: NMR δ 1.5–1.8 (m, 4 H), 2.0 (br s, 1 H), 2.5–2.8 (m, 2 H), 3.3–3.6 (m, 2 H), 7.0–7.8 (m, 9 H); MS, *m/e* (relative intensity) 274 (M^+, 48.7), 215 (100).

4-(2-Fluoranthenyl)butyraldehyde (20). Alcohol **19** (5.4 g, 0.02 mol) was dissolved in 150 mL of dry CH_2Cl_2 and added dropwise to a suspension of pyridinium chlorochromate (PCC, 8.4 g, 0.04 mol) in dry CH_2Cl_2 (100 mL). The mixture was stirred for 3 h and then filtered through Celite. The Celite was washed with CH_2Cl_2 (2 × 50 mL) and the combined filtrates were washed with 3 N HCl (2 × 100 mL) and H_2O (2 × 100 mL) and dried ($MgSO_4$). The solvent was removed to give aldehyde **20** (4.5 g, 82%) as a liquid, which was used without further purification: NMR δ 1.8–2.4 (m, 4 H), 2.5–2.8 (m, 2 H), 7.0–7.8 (m, 9 H), 9.5 (br s, 1 H); MS, *m/e* (relative intensity) 272 (M^+, 50.5), 215 (100).

4-(2-Fluoranthenyl)butyric Acid (21). To a solution of **20** (2.72 g, 0.01 mol) in 30 mL of THF/H_2O (3/1) was added 2.6 g of $AgNO_3$ in 100 mL of H_2O. The mixture was stirred at room temperature and 50 mL of 10% aqueous NaOH was added dropwise. The resulting mixture was stirred for 2 h, filtered, and washed several times with H_2O. The mixture was extracted with Et_2O and the aqueous phase was cooled and acidified with HCl. Extraction with CH_2Cl_2 followed by standard workup gave 1.8 g (62%) of **21** as a yellow solid: mp 168–169 °C; NMR δ 1.95–2.6 (m, 4 H), 2.7–3.1 (m, 2 H), 7.1–8.1 (m, 9 H); MS, *m/e* (relative intensity) 288 (M^+, 28), 244 (30), 215 (100).

(21) Buu-Hoï, N.; Lamy, J. *J. Chem. Soc.* **1959**, 1845–1849.

(22) Tucker, S. K. *J. Chem. Soc.* **1952**, 803–807.

Anal. Calcd for $C_{20}H_{16}O_2$: C, 83.33; H, 5.55. Found: C, 83.07; H, 5.54.

12-Oxo-9,10,11,12-tetrahydrobenzo[*b*]fluoranthene (22). Oxalyl chloride (1.9 g, 0.015 mol) was added dropwise to a stirred suspension of 1.8 g of **21** in 300 mL of CH_2Cl_2. After 3 h stirring at room temperature, a conventional workup gave 1.8 g of acid chloride which was used without further purification. Treatment of the acid chloride with $AlCl_3$ (1.0 g, 0.0075 mol) as described for **8** afforded crude **22**, which was chromatographed on silica gel using CH_2Cl_2/hexane (30/70) to give pure **22** as a yellow solid (1.0 g, 65%): mp 156–157 °C; NMR δ 1.6–2.1 (m, 2 H, H_{10}), 2.3–2.8 (m, 4 H, H_9 and H_{11}), 7.0–7.9 (m, 7 H), 8.8 (dd, 1 H, H_1, $J_{1,2}$ = 7 Hz, $J_{1,3}$ = 2 Hz); MS, *m/e* (relative intensity) 270 (M^+, 100), 242 (32.1), 214 (69.4).

Anal. Calcd for $C_{20}H_{14}O$: C, 88.88; H, 5.18. Found: C, 88.72; H, 5.30.

12-Hydroxy-9,10,11,12-tetrahydrobenzo[*b*]fluoranthene (23). A solution of **22** (0.8 g, 0.003 mol) in 30 mL of anhydrous THF was added dropwise during 10 min to a stirred suspension of 0.1 g (0.003 mol) of $LiAlH_4$ in 30 mL of THF. The mixture was stirred for 1 h at room temperature, diluted with H_2O, and extracted with EtOAc. The EtOAc solution was dried ($MgSO_4$) and concentrated to afford 0.8 g of **23**. The crude alcohol was used directly in the next step: NMR δ 1.6–1.8 (m, 4 H, H_{10} and H_{11}), 2.0–2.4 (m, 2 H, H_9), 2.9 (br s, 1 H, OH), 4.8 (br s, 1 H, H_{12}), 6.9–7.7 (m, 8 H); MS, *m/e* (relative intensity) 272 (M^+, 40), 254 (100).

9,10-Dihydrobenzo[*b*]fluoranthene (24). A solution of the alcohol **23** (2.72 g, 0.01 mol) in 150 mL of acetic acid containing 1 drop of concentrated HCl was heated at 90 °C under N_2 for 1 h. The solution was cooled and diluted with 500 mL of H_2O. The product was removed by filtration, washed with H_2O, dissolved in CH_2Cl_2, and chromatographed on silica gel. Elution with hexane–CH_2Cl_2 gave pure **24** (1.8 g, 72%): mp 127–129 °C; NMR δ 2.3–2.7 (m, 2 H), 2.8–3.2 (m, 2 H), 6.1–6.4 (m, 1 H), 7.1–7.9 (m, 9 H); MS, *m/e* (relative intensity) 254 (M^+, 100), 239 (10).

Anal. Calcd for $C_{20}H_{14}$: C, 94.48; H, 5.51. Found: C, 94.50; H, 5.42.

***trans*-11,12-Bis(benzoyloxy)-9,10,11,12-tetrahydrobenzo-[*b*]fluoranthene (25).** The reaction of **24** (2.5 g, 0.01 mol), silver benzoate (4.6 g, 0.02 mol), and I_2 (2.5 g, 0.01 mol) in benzene (100 mL) was effected as described for **13**. Crystallization of the crude product from CH_2Cl_2/hexane gave **25** (3.0 g, 60%) as a yellow solid: mp 213–214 °C; NMR δ 1.9–2.4 (m, 2 H), 2.6–3.1 (m, 2 H), 5.8–6.1 (m, 1 H, H_{11}), 6.9–7.9 (m, 19 H).

Anal. Calcd for $C_{34}H_{24}O_4$: C, 82.25; H, 4.83. Found: C, 82.31; H, 4.99.

9-Bromo-11β,12α-Bis(benzoyloxy)-9,10,11,12-tetrahydrobenzo[*b*]fluoranthene (26). The bromodibenzoate was prepared from **25** (2.0 g, 0.004 mol) and NBS (0.71 g, 0.004 mol) under conditions as described above for **17**: yield, 1.4 g (60%); mp 118–120 °C. This was used directly in the synthesis of **27**. NMR δ 1.6–3.2 (m, 2 H, H_{10}), 5.6–5.9 (m, 2 H, H_9 and H_{11}), 7.0–8.2 (m, 19 H).

***trans*-11,12-Bis(benzoyloxy)-11,12-dihydrobenzo[*b*]-fluoranthene (27).** To a solution of **26** (1.1 g, 0.002 mol) in THF under N_2 at 0 °C was added 1,5-diazabicyclo[4.3.0]non-5-ene (DBN, 4.8 g, 0.2 mol). The resulting mixture was stirred at 0 °C for 3 h. The reaction mixture was extracted with EtOAc and the EtOAc layer was washed with H_2O, dilute HCl, and H_2O, dried ($MgSO_4$), and concentrated to give **27** (0.8 g) as a dark yellow compound. The crude product was chromatographed on neutral alumina. Elution with CH_2Cl_2/hexane (30/70) gave pure **27** (0.5 g, 50%): mp 106–108 °C; NMR 5.9–6.2 (m, 1 H), 6.4–6.8 (m, 1 H) 7.0–8.3 (m, 20 H).

***trans*-11,12-Dihydro-11,12-dihydroxybenzo[*b*]fluoranthene (4).** The conversion of **27** (0.5 g, 0.001 mol) to the corresponding dihydrodiol was carried out as described for preparation of **3**. The crude product was chromatographed on Florisil with elution by CH_2Cl_2 and EtOAc/CH_2Cl_2 (30/70) to give first a mixture of *cis*-**4** and *trans*-**4** and then *trans*-**4** (0.09 g, 32%): mp 151–152 °C; 300 MHz NMR δ 4.3 (m, 1 H, H_{11}), 5.3 (d, 1 H, H_{12}, $J < 3$ Hz), 6.2 (m, 1 H, H_{10}), 6.8 (d, 1 H, H_9, $J_{9,10}$ = 9.6 Hz), 7.2–7.4 (m, 2 H), 7.5–7.8 (m, 2 H), 7.85–8.1 (m + s, 3 H), 8.2 (d, 1 H); MS, *m/e* (relative intensity) 286 (M^+, 29.3), 268 (100), 239 (61.9); UV (MeOH) λ max 370 nm (ϵ 8928), 352 (8928), 300 (10357), 288 (25357), 278 (47857), 270 (44285), 260 (44285), 236 (27857); HPLC retention times *trans*-**4**, 22.5 min; *cis*-**4**, 28.3 min.

Anal. Calcd for $C_{20}H_{14}O_2$: C, 83.91; H, 4.89. Found: C, 83.69; H, 5.03.

Mutagenicity Assays. *S. typhimurium* strains were kindly provided by Dr. Bruce Ames of the University of California, Berkeley, CA. Assays were carried out as described previously.[16] Positive controls included *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine and quinoline. Percent survivors for all compounds assayed were determined by employing dilutions of bacterial broth under identical conditions, except that excess histidine was added to the top agar.

Acknowledgment. We thank Dr. Edmond J. LaVoie for carrying out the mutagenicity assays. 300-MHz NMR spectra were obtained at the Rockefeller University on a Nicolet Magnetics NTC 300 Wide Bore Spectrometer, purchased with funds from The National Science Foundation (PCM-7912083), The Camille and Henry Dreyfus Foundation, and The Fleischmann Foundation.

Registry No. 1, 205-99-2; *cis*-**3**, 88746-57-0; *trans*-**3**, 88746-47-8; **4**, 81824-11-5; **5**, 88746-48-9; **6**, 88746-49-0; **7**, 88746-50-3; **8**, 88746-51-4; **9**, 88766-63-6; **10**, 88746-52-5; **11**, 88746-53-6; **12**, 88746-54-7; **13**, 88746-55-8; **14**, 88746-56-9; **16**, 33543-31-6; **17**, 88746-58-1; **18**, 88746-59-2; **19**, 88746-60-5; **20**, 88746-61-6; **21**, 88746-62-7; **22**, 88746-63-8; **23**, 88746-64-9; **24**, 88746-65-0; **25**, 88746-66-1; **26**, 88746-67-2; **27**, 88746-68-3; methyl acrylate, 96-33-3.