Synthesis of Cyclopenta[*cd*]pyrene 3,4-Epoxide, the Ultimate Mutagenic Metabolite of the Environmental Carcinogen, Cyclopenta[*cd*]pyrene

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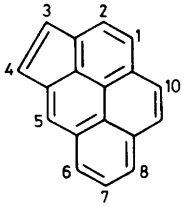
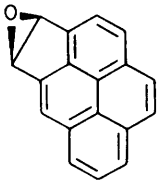
Summary Cyclopenta[*cd*]pyrene 3,4-epoxide, a potent bacterial mutagen, has been synthesized *via* the bromohydrin obtained from the addition of hypobromous acid to the ethylenic bridge of cyclopenta[*cd*]pyrene.

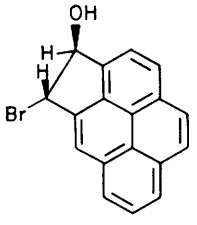
THE non-alternant, non-bay region structure of the highly mutagenic polycyclic aromatic hydrocarbon cyclopenta[*cd*]-pyrene (**1**) has stimulated interest in this compound.[1-5] The 3,4-epoxide (**2**) was predicted to be the ultimate mutagenic metabolite.[1] We report the synthesis of (**2**) *via* the bromohydrin formed directly from (**1**) by addition of HOBr generated from *N*-bromosuccinimide (NBS) in wet dimethyl sulphoxide.[6] This reaction should be generally applicable to epoxidation of arene double bonds to which electrophilic addition is favoured. We also confirm that (**2**) is a potent mutagen in the absence of liver enzymes.



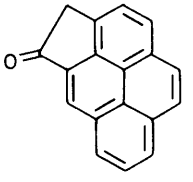
(1)



(2)



(3)



(4)

Since PMO considerations[7] pointed to the ethylenic bridge of both (**1**) and acenaphthylene as the favoured site for electrophilic addition, acenaphthylene was selected as a model polycyclic compound for the addition with HOBr. Reaction of acenaphthylene with 2 equiv. of NBS in Me_2SO containing 7% H_2O at 15 °C yielded (40%), the bromohydrin as colourless needles following reverse phase h.p.l.c.: m.p. 199—201 °C; δ (60 MHz, $CDCl_3$) 5·49 and 5·81 (each 1H, br. s), and 7·35—7·90 (6H, m, ArH); λ_{max} (CH_2Cl_2) (log ε), 285 (3·34) nm; mass spectrum, M^+ 250 and 248 with major fragment at *m/e* 169 (M^+ — HBr). The small coupling of the bridge protons is consistent with a *trans* configuration.[8]

Similar treatment of (**1**) yielded (66%) the bromohydrin as pale yellow needles following reverse phase h.p.l.c.: m.p. (sealed capillary) 118—119 °C (decomp.); δ(270 MHz, CD_3COCD_3 5·80 and 6·14 (each 1H, br. s) and 8·08—8·39 (8H, m, ArH); λ_{max} (CH_2Cl_2) (log ε), 345 (3·92), 328 (3·79), 314 (3·46), 296 (3·11), 277 (4·08), 267 (3·81), 255 (3·52), 246 (4·26), and 233 (4·01) nm; mass spectrum, M^+ 324 and 322, major fragments at *m/e* 306 and 304 ($M^+ - H_2O$). The pyrene-like appearance of the u.v. spectrum and the small coupling of the benzylic protons in the n.m.r. spectrum indicate *trans* addition to the ethylenic bridge of (**1**). Resolution of only one bromohydrin by h.p.l.c. and the n.m.r. evidence are consistent with the prediction by PMO theory of Br^+ addition at C(4). On this basis, structure (**3**) (*trans*-4-bromo-3-hydroxy-3,4-dihydrocyclopenta[*cd*]pyrene has been tentatively assigned.

A solution of (**3**) in dry tetrahydrofuran (THF) was added to a 10-fold molar excess of sodium methoxide and stirred at room temperature for 10 min. Percolation of the mixture through a small amount of basic alumina (activity IV) and concentration of the solvent at 0 °C with a stream of nitrogen yielded (33%) (**2**), as colourless spars, m.p. (sealed capillary) 207—209 °C (decomp.); δ (270 MHz, CD_3COCD_3) 5·18, (2H, br. s) and 8·08—8·39 (8H, m, ArH); λ_{max} (CH_2Cl_2) (log ε), 374 (3·63), 366 (3·39), 353 (4·12), 347 (4·49), 330 (4·36), 315 (4·01), 275 (4·54), 264 (4·34), 246 (4·68), and 232 (4·64) nm; mass spectrum M^+ 242·07210 (calc. for $C_{18}H_{10}O$ 242·07316), major fragments at *m/e* 214 and 213; i.r. (KBr) 12·1 and 12·4 μm. The chemical shift and near-degeneracy of the oxiran protons in the n.m.r. spectrum are in accord with data[9] for other asymmetric arene oxides.

The direct mutagenicity of (**2**) was confirmed by exposing 2×10^8 bacteria (*Salmonella typhimurium* strain TA100, a histidine auxotroph),[10] to different amounts of the purified epoxide in 0·5 ml of 0·1 M sodium phosphate, pH 7·4. After 5 min at room temperature, the mixture was plated on selective minimal plates and the number of his^+ revertants was determined after 48 h at 37 °C. When bacteria were exposed to 5, 10, 20, 50, or 100 ng of the epoxide, the yield of induced his^+ revertants per plate was 21, 51, 111, 303, and 735, respectively. Clearly, the epoxide is potently mutagenic in the absence of liver microsomes and the yield of mutants varies linearly with dose.

T.l.c. over silica or stirring with HCl in THF isomerized (**2**) cleanly and completely to the ketone (**4**), which can be distinguished from the previously described 3-oxo derivative[2-4] by u.v. and i.r. spectroscopy and its m.p. (220—222 °C). Isomerization exclusively to (**4**) is consistent with the predicted preference for carbonium ion formation[1] at C(3) and lends support to the assignment of structure (**3**) for the bromohydrin.

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