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HYDROXYLATION OF BENZENES INVOLVING THE NIH SHIFT UTILIZING ELECTRONIC MODEL COMPOUND FOR MONOOXYGENASES

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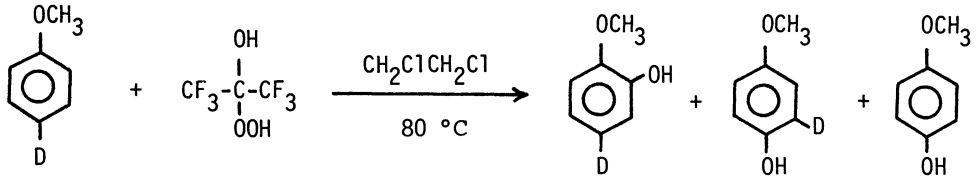
The reaction of anisole-4-^{2}H or toluene-4-^{2}H with 2-hydroperoxyhexafluoro-2-propanol gave corresponding phenols involving the NIH shift, the extent of which was 57% and 51% respectively. These values were very close to those of cytochrome P450.

Intramolecular migration of aryl ring substituents during hydroxylation (the NIH shift)[1] is observed in reactions catalyzed by various types of monooxygenases.[2] The NIH shift provides evidence for the formation of arene oxide intermediates during enzymatic hydroxylation of aromatic compounds.[1] In marked contrast to aliphatic olefins, epoxidation of benzenes is energetically very unfavourable because of their large resonance energy and thus the NIH shift may be the most characteristic process among reactions of monooxygenases. Recently, many interesting reagents[3,9] mimicking the function of oxygenases are advanced for epoxidation of simple olefins but examples of hydroxylation of aromatic compounds involving the NIH shift are limited.[4,5,7]

Since the early work concerning the NIH shift,[5] anisole-4-^{2}H, (1) has been utilized as a model substrate. It exhibits a high migration and retention (60%) of deuterium during microsomal para-hydroxylation[6] and two reagents (molybdenum hexacarbonyl with t-butyl hydroperoxide[7a] and photochemically excited pyrazine-N-oxide[7b]) are reported to cause a comparable retention with the microsomal enzyme. Elaboration of chemical oxidants which exhibit the NIH shift

should permit further insight into the electronic requirement for the "active oxygen" of monooxygenases. In this paper, we wish to report another system to cause the NIH shift in close similarity to cytochrome P450.

Ethylene dichloride solution of 1 (1.0 g) and 2-hydroperoxyhexafluoro-2-propanol, (2), prepared[8,9] in situ from hexafluoroacetone (200 mg) and hydrogen peroxide (30% aqueous solution, 0.5 ml) was stirred vigorously at 80 °C for 16 h (see the equation below). Purification of the products[10] with silica gel chromatography afforded 2-hydroxy-4-deuterio-anisole (3, 80 mg) and 4-hydroxyanisole (4 + 5, 30 mg) with unreacted 1 (770 mg). The retention of deuterium in 4-hydroxyanisole was found to be 57% (mean value of three runs) on the basis of NMR spectrum. The position of deuterium was established utilizing the shift reagent ($Eu(DPM)_3$). Toluene-4-^2H was also subjected to a similar reaction and investigation of NMR spectrum of the resulting p-cresol revealed the retention of deuterium to be 51% (mean value of three runs). Again this value is very close to that of microsomes (54%).[6] These results support that 2 is an excellent model for the electronic state of "activated oxygen" of monooxygenases to cause the NIH shift.



1 2 3 4 5

Recently, 2 was suggested as a structural analogue of flavin hydroperoxide,[9] which was implicated to be an active species in flavin dependent monooxygenases.[11] Although known flavoprotein monooxygenases do not catalyze

a reaction, in which the NIH shift can be examined,[2] a closely related cofactor, i.e., tetrahydrobiopterin dependent monooxygenases such as phenylalanine hydroxylase[12] and tryptophan hydroxylase[13] cause the NIH shift in high yield. In these enzymes, dioxygen reacts with the reduced state of cofactor (dihydroflavin or tetrahydrobiopterin) and finally utilizes two electrons to become the activated form.[2] Therefore, it is isoelectronic with hydrogen peroxide somehow bound to the oxidized state of cofactor, which is apparently electron-withdrawal on the contrary to the reduced coenzymes. In this point, it is noticeable that 2, which is assumed to be an electron-deficient hydrogen peroxide adduct, mimicks the diagnostic reaction of monooxygenases.

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