Evidence for an Anomalous Microbial Oxidation of Acetophenone: New Access to Optically Active Tricarbonyliron Complexes

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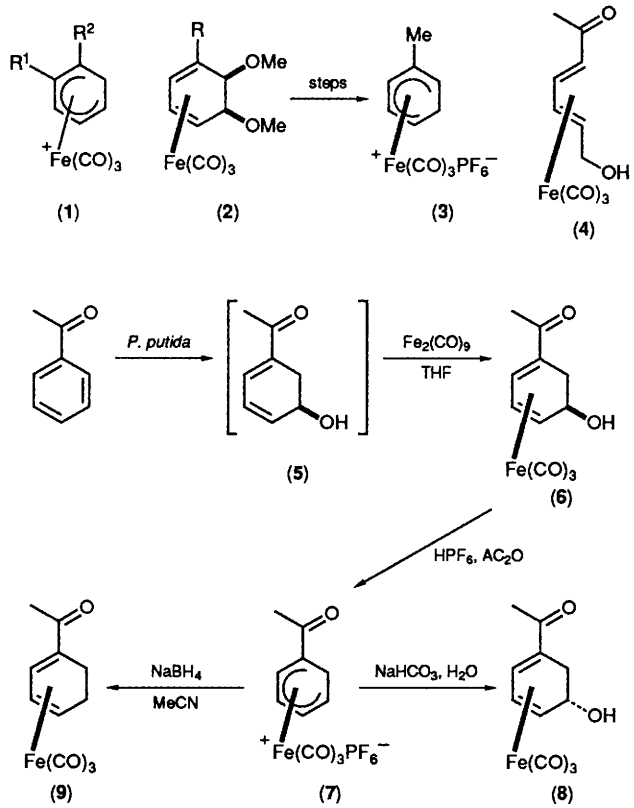
Oxidation of acetophenone by *Pseudomonas putida* afforded an unusual monohydroxy product, identified by conversion into the corresponding tricarbonyliron complex, and shown by circular dichroism (CD) investigations and chemical correlations to arise by addition of OH to the same enantioface of the arene ring as that susceptible to reaction in the normal dioxygenation pathway.

The dioxygenation of arenes by *Pseudomonas putida* has provided optically active *cis*-diol products that are valuable as starting materials for enantioselective organic synthesis.[1] In our own investigations in this area,[2,3] the chirality of the diol has been relayed to tricarbonyliron complexes; these are important as intermediates in asymmetric synthesis because, when converted into cationic η^5 complexes of type (**1**), alkylation reactions can be reliably performed with complete stereocontrol.[4] The diol complexation method has allowed the preparation of an optically pure sample of (**3**) from the diol



Scheme 1. Formation and absolute configuration of complexes obtained from a microbial mono-oxygenation product from acetophenone.

complex (**2**, R = Me),[3] and is now under investigation as a general route to homochiral tricarbonyliron complexes of types (**1**) and (**2**).

In the course of these studies, which have required an examination of the range of substrates suited to oxidation by *P. putida* (ICI strain 11767), we have found evidence for the anomalous formation of a monohydroxy product from acetophenone. The material obtained was somewhat unstable and was converted directly into the η^4-diene complex (**6**)† by reaction with $Fe_2(CO)_9$ in tetrahydrofuran (THF). This diene complex was formed as a single diastereoisomer, and was purified by chromatography.‡ Since *P. putida*-derived diols can be converted into tricarbonyliron derivatives without loss of the diol functionality,[2,3] this result provides an indication that mono-oxygenation, not dioxygenation, has been effected

† All new complexes have been characterised by NMR, IR, and mass spectrometry and by microanalysis or high resolution mass spectrometry.

‡ Oxidation of 36 g of acetophenone by a culture of *P. putida* (ICI strain 11767) at 27 °C for 4.5 h, followed by continuous extraction of the aqueous phase with chloroform, afforded a mixture of arenes and partially saturated oxidation products which were complexed directly with $Fe_2(CO)_9$. The monohydroxy complex (**6**) (1.44 g) was isolated by chromatography. Examination of the NMR spectrum of the crude complexation product indicated that a small quantity (<0.2 g) of the 5,6-diol complex had also been formed by complexation of the expected 1-acetylcyclohexa-1,3-diene-5,6-diol ligand which is the known product of *P. putida* oxidation of acetophenone: T. Gibson, B. Gschwendt, W. K. Yeh, and V. M. Kobal, *Biochemistry*, 1973, **12**, 1520.

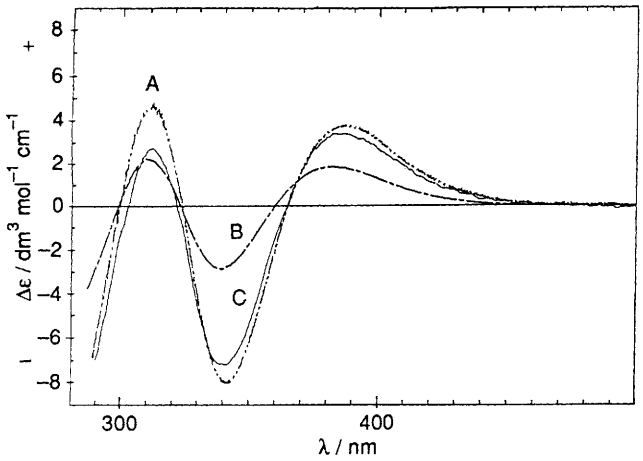


Figure 1. Circular dichroism spectra of complexes (**6**), (**8**), and (**9**), recorded in $CHCl_3$ at 25 °C. A, spectrum of complex (**6**) ($[\alpha]_D$ −47.2°) at *c* 0.9; B, spectrum of complex (**8**) ($[\alpha]_D$ −243.0 °) at *c* 1.0; C, spectrum of complex (**9**) ($[\alpha]_D$ −201.5°) at *c* 0.6.

in the case of the acetophenone substrate.§ Examination of the circular dichroism (CD) spectrum of this material revealed a similar spectrum to that of acyclic complexes of the related type (**4**), which have been the subject of a recent investigation.[5] This suggests that the planar chirality of the η^4 portion of (**6**) has the absolute configuration drawn in Scheme 1. NMR coupling constants provide a good guide to the relative stereochemistry between substituents at C-5 or C-6, and the face of cyclohexadienes bound to an $Fe(CO)_3$ group.[6] In the case of (**6**), examination of the ^1H NMR spectrum suggested that the *endo* isomer had been formed in the complexation step, as had been observed previously for dioxygenated products.[2,3]

In order to identify with certainty the relationship between enantioface selectivity in the mono-oxygenation and dioxygenation reactions effected by *P. putida*, steps were taken to confirm these stereochemical conclusions. The complex (**6**) was converted, by reaction with HPF_6 in acetic anhydride, into the cationic complex (**7**) ($[\alpha]_D^{25}$ −74.0°, *c* 0.4, MeCN) which was precipitated as a yellow powder in 86% yield by dilution of the reaction mixture with ether. Addition of HO^- to (**7**) by reaction with aqueous sodium hydrogen carbonate provided a sample of the alcohol (**8**), which showed the expected[6] NMR coupling constants for an *exo* isomer. The product (**8**) was formed in 52% yield. Comparison of (**8**) with (**6**) by ^1H NMR spectroscopy showed the two complexes to be diastereoisomers. Since the *exo* mode of addition of HO^- to tricarbonyl(η^5-cyclohexadienyl)iron cation complexes is well established,[7] this comparison confirms the *endo* relative stereochemistry of (**6**). Reduction of (**7**) with sodium borohydride

§ While with monocyclic substrates direct mono-oxygenation seems the most probable pathway, with bicyclic substrates an alternative overall mono-oxygenation could arise from dioxygenation/elimination reactions. The formation of bicyclic mono-oxygenation products with structures consistent with a route of this second type has recently been reported: D. R. Boyd, R. Austin, S. McMordie, N. D. Sharma, H. Dalton, P. Williams, and R. O. Jenkins, *J. Chem. Soc., Chem. Commun.*, 1989, 339; L. P. Wackett, L. D. Kwart, and D. T. Gibson, *Biochemistry*, 1988, **27**, 1360; D. B. Boyd, R. Austin, S. McMordie, H. P. Porter, H. Dalton, R. O. Jenkins, and O. W. Howarth, *J. Chem. Soc., Chem. Commun.*, 1987, 1722; S. E. Jones and H. Dalton, poster at 'Biotransformations' SCI symposium, Cambridge, April 1989.

proceeded by hydride addition at the far terminus of the dienyl system, forming the dihydroacetophenone complex (**9**)‡ in 74% yield after chromatography. The similarity between the CD spectra of (**6**), (**8**), and (**9**), shown in Figure 1, indicates that the chirality of attachment of the OH substituent does not exert a significant effect on the form of the CD curve. This observation further supports the assignment of the (1*R*,5*R*) absolute configuration to (**6**) by the comparison of our chiroptical data with the results reported[5] for acyclic complexes which lack a chiral centre in this position.

Our findings indicate that the original oxidation product (**5**) would have had the (*R*) configuration at C-5, and so establishes that the microbial oxidation of the aromatic ring leading to the formation of (**5**) occurs with the same enantioface selectivity as the more normal[8] dioxygenation process.

The mono-oxygenation mode of action offers potential advantages in the preparation of simple tricarbonyl(η^5-cyclohexadienyl)iron(1+) complexes of type (**1**), which will be valuable as stereocontrolled electrophilic reagents in enantioselective synthesis. While more extensive investigation is needed to develop general arene mono-oxygenation methods, our studies have demonstrated, at the present stage, that this type of biotransformation is possible, and that the absolute configuration of the monohydroxy product corresponds to that commonly arising in the normal dioxygenation series.

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