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AROMATIC BIOTRANSFORMATIONS 2: PRODUCTION OF NOVEL CHIRAL FLUORINATED 3,5-CYCLOHEXADIENE-*CIS*-1,2-DIOL-1-CARBOXYLATES

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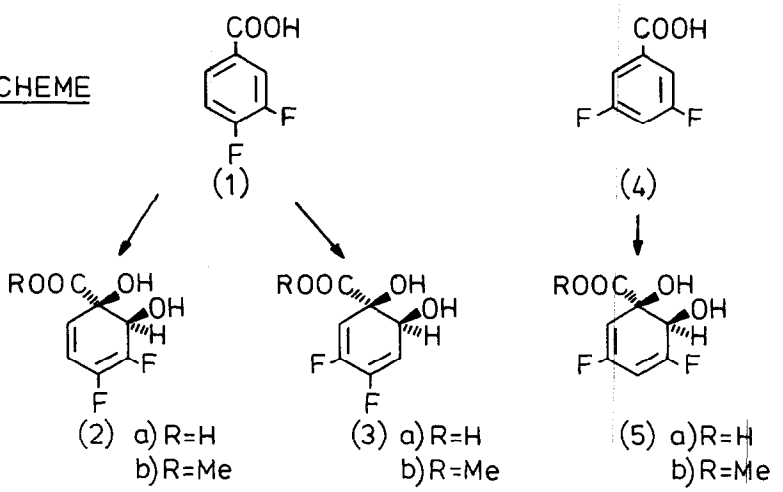
Summary 3,4-, 4,5-and 3,5-Difluoro-3,5-cyclohexadiene-*cis*-1,2-diol-1-carboxylates were prepared from 3,4-and 3,5-difluorobenzoate by microbial oxidation with a mutant strain of *Pseudomonas putida* JT 103.

As the demand for efficient chiral synthesis increases so does the requirement for new chiral building blocks[1]. The microbial oxidation of simple aromatic systems can provide such building blocks as was recently shown in a six step synthesis of (±) - pinitol from prochiral *cis*-1,2-dihydroxyclohexa-3,5-diene, a bacterial oxidation product of benzene [2]. Similar carboxylated chiral diols are formed in high yield by bacterial mutants from benzoate and several analogues [3,4,5,6]. All four isomers of monofluorinated 3,5-cyclohexadiene-*cis*-1,2-diol- 1-carboxylate have been obtained from the three isomers of fluorobenzoate. Here we report the formation of three optically active novel difluorinated 3,5-cyclohexadiene-*cis*-1,2-diol-1-carboxylates from difluorobenzoates with a mutant strain of *Pseudomonas putida* JT 103 (Scheme).

SCHEME



3,4-Difluorobenzoate (1) was quantitatively oxidised by strain JT 103 to give two regioisomers, 3,4- and 4,5- difluoro-3,5- cyclohexadiene-cis-1,2-diol-1-carboxylate (2a,3a) in the ratio of 3:1 while 3,5-difluorobenzoate (4) gave 3,5-difluoro-3,5-cyclohexadiene-cis-1,2-diol-1-carboxylate (5a)[7]. The free acids were characterised as their methyl esters (2b,3b,5b) and the cis stereochemistry confirmed by preparation of the acetonides[8]. The structures of the methyl esters were confirmed by ^{1}H-n.m.r., COSY and heteronuclear ^{1}H-^{19}F COSY. (2b), δ (360MHz) 6.07 (1H,dt,J=10.2,7.8Hz,H_5) 5.58 (1H,ddd,J=10.1,4.7,1.9Hz,H_6) 4.79 (1H,dd,J=7.4,2.4 Hz,H_2) 3.87 (3H,s,Me) 3.88 (1H,br s,OH) 3.20 (1H,br s,OH); (3b) 5.45 (2H,m,H_6,H_3) 4.94 (1H,dd,J=6.6,2.0Hz,H_2) 3.90 (3H,s,Me) 3.82 (1H,br s,OH) 2.85 (1H,br s,OH); (5b) 5.57 (1H,ddt,J=8.5,7.3,2.0Hz,H_4) 5.17 (1H,dquintet,J=10.8,1.1Hz,H_6) 4.95 (1H,m,H_2) 3.89 (3H,s,Me) 3.77 (1H,br s,OH) 2.79 (1H,br s,OH). $[\alpha]_D^{25}$ (2b)= -228.9^{o} (C= 1.24;$CHCl_3$), (3b)= -66.3^{o} (C=0.54,$CHCl_3$), (5b)= -79.8^{o} (C=0.420;$CHCl_3$).

These results further demonstrate the power of microbial hydroxylations with mutant strains in producing otherwise inaccessible fluorinated chiral compounds in high yields for use as chiral starting analogues of a variety of compounds with biological activity.

Footnotes.

All new compounds gave satisfactory n.m.r. and mass spectral data (including accurate mass)

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7 Cells of strain JT 103 were grown up in a litre of culture medium[9] containing sodium benzoate (5mM, induces the benzoate pathway), harvested and resuspended in medium containing phosphate buffer (pH 7.0,0.1M) and 1-5g of the appropriate difluorobenzoate. The free acids were isolated as previously described[3]. The two regioisomers 2b and 3b were separated on a Waters μBONDAPAK C_{18} preparative column using a mobile phase containing 90% water and 10% methanol.

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