$Fe(CO)_4$ unit which is linked only to the apical Bi(1), with Bi(1)–Fe(4) = 2.752 (6) Å. Atoms Fe(1), Fe(2), and Fe(3) are each in a distorted-octahedral coordination environment, whereas Fe(4) has a trigonal-bipyramidal geometry. The $[Bi_4Fe_4(CO)_{13}]^{2-}$ cluster as a whole has approximate C_{3v} symmetry.

As shown in Figure 2, there are weak Bi···Bi interactions between the dianions. These secondary contacts are reminiscent of the interactions observed in such solid state Zintl phases as $Ca_{11}Bi_{10}$.[7]

The two Bi–Bi distances observed for I (ca. 3.16 and 3.46 Å) are comparable with the two closest Bi–Bi contacts in the pure crystalline element (3.07 and 3.53 Å).[8] Compound I may be compared to the Bi_4^{2-} anion, which has been crystallographically charterized.[9] In this molecule, a square-planar array of Bi atoms is observed with two unique Bi–Bi distances of 2.936 (2) and 2.941 (2) Å. These distances are noticeably shorter than for I, and this may arise via π interactions in the square-planar molecule since it is a 6π-electron system. Unfortuntely, the Bi–Bi distance cannot be directly compared to that in tetrahedral $Sn_2Bi_2^{2-}$ since in that molecule the Sn and Bi atoms are equally disordered over all sites.[10] The bonding in this molecule will be discussed elsewhere.

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Registry No. $[Et_4N][BiFe_3(CO)_{10}]$, 92786-73-7; $[Et_4N]_2[Bi_4Fe_4(CO)_{13}]$, 94483-21-3; $Fe(CO)_4PPh_3$, 35679-07-3; $Fe(CO)_3(PPh_3)_2$, 21255-52-7; $Fe(CO)_5$, 13463-40-6; Bi, 7440-69-9; Fe, 7439-89-6.

Supplementary Material Available: Tables of fractional coordinates and anisotropic thermal parameters (2 pages). Ordering information is given on any current masthead page.

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Kinetically Stable Conformers of 3,4,5,6-Tetramethyl-9,10-dihydroxy-9,10-dihydrophenanthrene as Probes of the Conformer Specificity of UDPglucuronosyltransferase

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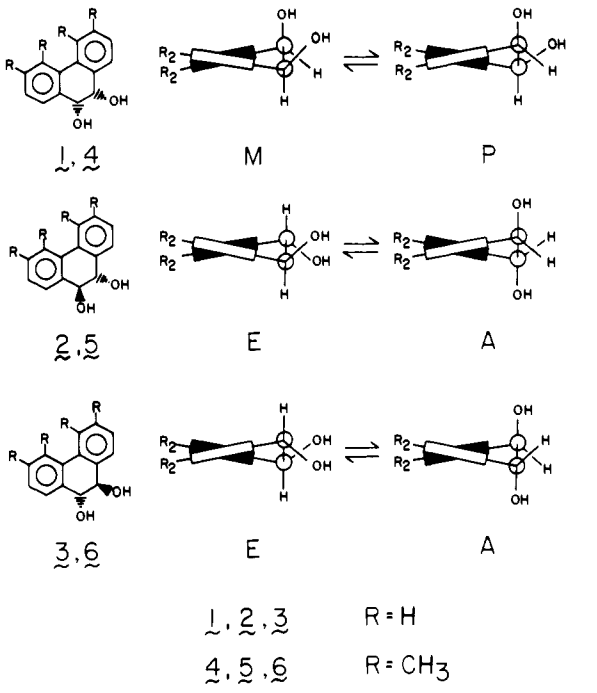
Microsomal UDPglucuronosyltransferase (EC 2.4.1.17) participates in the mammalian metabolism of polycyclic aromatic hydrocarbons by catalyzing the glucuronidation of phenolic and *trans*-dihydro diol metabolites.[1] The enzyme has been shown to discriminate between stereochemically distinct carbinol groups of several dihydro diols, e.g., the 9,10-dihydroxy-9,10-dihydrophenanthrenes **1–3**.[2] A salient feature of the stereochemical recognition of dihydro diols by UDPglucuronosyltransferase is the potential for conformer specificity (Scheme I). Unfortunately,

Scheme I



the kinetic lability of the conformers of **1–3** precludes their use in obtaining this information. In this paper we report the synthesis and use of the six kinetically stable, stereoisomeric conformers of 3,4,5,6-tetramethyl-9,10-dihydroxy-9,10-dihydrophenanthrene to ascertain the conformer specificity of UDPglucuronosyltransferase.

Oxidation of 3,4,5,6-tetramethylphenanthrene with OsO_4 followed by workup with $NaHSO_3$ gave the racemate **4**.[3] Further oxidation of **4** to the orthoquinone with DDQ[4] followed by the KBH_4 reduction in the presence of O_2[5] gave, stereoselectively, a mixture consisting of racemic trans diequatorial isomers **5E** + **6E** (98%) and cis isomers **4** (2%). Mutarotation of **5E** + **6E** (16 h, 90 °C, 25% CH_3OH, 75% H_2O) gave a 20% yield of **5A** + **6A** after separation of the equilibrium mixture by silica chromatography. Structures of the three racemates were based on proton NMR spectra of the corresponding diacetates, where vicinal coupling constants $^3J_{9,10}$ for the benzylic protons are particularly diagnostic of the relative configuration. Benzylic protons of diacetyl-**4** were magnetically nonequivalent, δ 5.81 (d, 1 H), 5.93 (d, 1 H), $^3J_{9,10}$ = 3.0 Hz. Coupling constants for the magnetically equivalent benzylic protons of the trans isomers were obtained from the natural-abundance ^{13}C-satellite resonances located 77 Hz upfield and downfield from the singlet resonance of the ^{12}C isotopomers.[6] Thus for diacetyl-(**5E** + **6E**) δ 5.91 (s, 2 H), $^1J_{^1H,^{13}C}$ = 155 Hz, $^3J_{9,10}$ = 11.2 Hz, and for diacetyl-(**5A** + **6A**) δ 5.80 (s, 2 H), $^1J_{^1H,^{13}C}$ = 155 Hz, $^3J_{9,10}$ = 3.1 Hz. Further confirmation of the structures of the *trans*-diacetates was obtained by X-ray crystallography as shown in Figure 1.

The *cis*-antipodes **4M** and **4P** were resolved on a preparative scale via synthesis, resolution, and hydrolysis of the diastereomeric bis[(−)-α-methoxy-α-(trifluoromethyl)phenylacetates.[7] Enantiomers **5A** and **6A** were readily resolved by HPLC using a chiral stationary phase.[8] The diequatorial isomers **5E** and **6E** were

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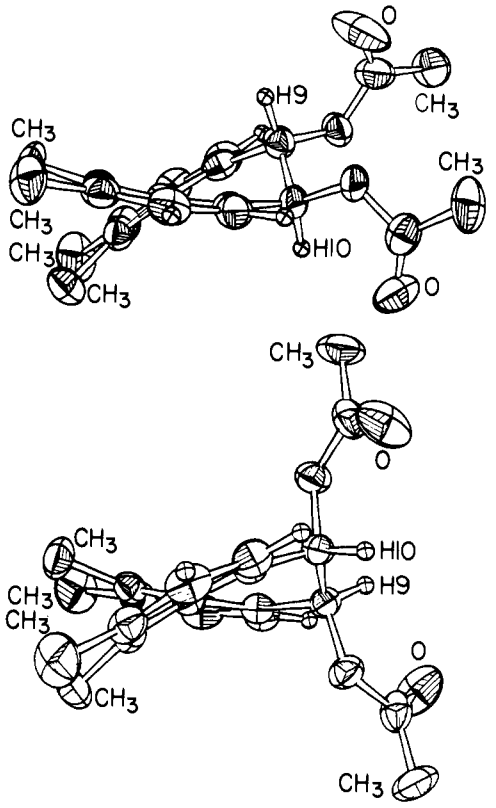

Figure 1. ORTEP representations of the diactate of **5E** (top) and **5A** (bottom) viewed down the biphenyl axis. Twist angle of the biphenyl is 42°. Final R ($\sum|F_o - F_c|\sum F_o$) values were 0.092 and 0.080 for **5E** + **6E** and **5A** + **6A**, respectively.

Table I. Kinetics of the Enzyme-Catalyzed Glucuronidation of the Six Stereoisomeric 3,4,5,6-Tetramethyl-9,10-dihydroxy-9,10-dihydrophenanthrenes[a]

substrate	$\Delta\epsilon_{dis}$,[b] M^{-1} cm^{-1}	abs config[c]	k_c, s^{-1}	k_c/K_{mapp}, M^{-1} s^{-1}
4P	+143	*R,S,P*	0.020[d]	
4M	−143	*R,S,M*	0.0058[d]	
5A	+144	*S,S,P*	<0.0005[d,e]	
6A	−143	*R,R,M*	0.0069[d]	
5E	−120	*S,S,M*	0.41 ± 0.01	980 ± 100
6E	+116	*R,R,P*	0.20 ± 0.004	910 ± 60

[a] Reactions were run as described in ref 2. [b] Circular dichroic extinction coefficients for the dissymmetry transition at 233 (**4M** and **4P**), 232 (**5A** and **6A**), and 230 nm (**5E** and **6E**) were determined in 2-propanol. [c] Designations *R* and *S* described absolute configuration of the carbinol carbons. *M* and *P* designate the helicity of the biphenyl axis. [d] Turnover numbers were estimated by using saturating 1.5–2.2 mM substrate concentrations. Reactions were too slow for accurate determination of k_c/K_{mapp} due to lability of the enzyme at 25 °C. [e] No product detected.

resolved by synthesis and resolution of the diastereomeric mono[(−)-α-methoxy-α-(trifluoromethyl)phenylacetates]. All pairs of enantiomers gave mirror image CD spectra (supplementary material, Figures 2–4) with an intense dissymmetry transition at ~232 nm characteristic of the 2,2′-bridged biphenyl chromophore. Absolute configurations of the six stereoisomers were determined from the sign of this transition (Table I), which reports the helicity of the biphenyl axis.[6,9] The conformers are quite stable at room temperature. For example the half-life for racemization of **4** is 2.0 years at 25 °C.[3]

Enzyme-catalyzed reactions of the six conformationally locked substrates were monitored by reversed-phase HPLC.[10] Three interesting observations can be made. First, only one product from each of the two *cis*-antipodes is evident by HPLC even at long reaction times, suggesting that only one of the two topochemically distinct carbinol groups of **4M** or **4P** is glucuronidated. Second, the trans diaxial stereoisomer **5A** is not a substrate for the enzyme. Finally, the most interesting feature of the enzyme-catalyzed reaction is the striking kinetic discrimination between the six stereoisomers by UDPglucuronosyltransferase (Table I). In particular, the trans diequatorial isomers **5E** and **6E** are turned over 30 to >800 times more rapidly than their corresponding conformational diastereomers with axial hydroxyl groups. It therefore seems likely that the preferred conformation of kinetically labile *trans*-dihydro diols such as **2** or **3** in a productive enzyme–substrate complex is that with diequatorial hydroxyl groups. In view of the conformer specificity of the enzyme toward the trans isomers it appears probable that only the equatorial hydroxyl group in **4M** and **4P** is recognized by the enzyme. This suggests that the two diasteromeric glucuronides formed from **1**[2] arise via reaction of the equatorial hydroxyl groups of the two conformational enantiomers **1M** and **1P** in the enzyme–substrate complex. Work is in progress to clarify this point.

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Supplementary Material Available: Description of synthesis and resolution of **4M**, **4P**, **5A**, **6A**, **5E**, and **6E**, complete NMR data for the racemic diacetates, circular dichroism spectra of all six stereoisomeric dihydro diols (Figures 2–4), and crystallographic data including atomic coordinates and temperature factors (29 pages). Ordering information is given on any current masthead page.

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(10) Retention times (min) for substrates and products: **4M** and **4P**, 42.6; **4M** glucuronide, 36.0; **4P** glucuronide, 36.5; **5A** and **6A**, 29.6; **6A** glucuronide, 17.6; **5E** and **6E**, 45.0; **5E** glucuronide, 42.2; **6E** glucuronide, 40.9, found by using a Rainin Microsorb C18 column (4.6 mm × 25 cm) eluted at 0.5 mL/min with 50% CH_3OH in 0.1 M acetic acid for 10 min then a gradient of 1%/min to 80% CH_3OH. Products were identified by their CD spectra.

Calculation of Substrate Dissociation Constants from Steady-State Isotope Effects in Enzyme-Catalyzed Reactions

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Isotopic probes of enzyme reactions provide a measure of the extent to which bond cleavage steps limit catalysis.[1-7] Further, when the magnitude of the intrinsic isotope effect on isolated bond

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