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Scheme 4. a) iBuLi, THF, 90 min, -78 °C; Bu2NF, THF, 1 h, 0 °C; 85%; b) iBuLi, CICO2Et, THF, 1 h, -30 °C, 85%; c) morpholine, THF, 2 h, reflux; 60% CH3CO2H, 2 h, 50 °C, 75%; d) TBDPSCl, imidazole, DMF, 7 d, -60 °C, 96%; e) TBDPSCl, imidazole, DMF, 7 d, -60 °C, 96%; f) Ac2O, pyridine, 4-dimethylaminopyridine, 30 min, 0 °C, 98%; g) HCl(O+Me)2, CH2Cl2, MeOH, 4 d, 20 °C, 99%; h) Ti(O+Pr)4, 2-(trimethylsilyl)ethanol, 3 d, 100 °C, 90%; i) CsF, DMF, 1 d, 30 °C, 98%; j) 1-(trimethylsilyl)ethanol (TDMSCl), CH2Cl2, triethylamine, 30 min, 20 °C; (CH3)2C=CH2, CICO2H, 1 h, 0 °C, 91%; k) triethylamine, acetone, H+O, 15 min, RT; Cs2CO3, DMF, 1 d, 20 °C, 50%; l) tetrabutylammonium fluoride, THF, 4 h, 0 °C, 1 M HCl, THF, 1 d, 20 °C, 95%; m) 2 equiv of potassium 2,6-diisopropylphenoxide, DMF, 1 d, 20 °C; m) Me$_2$S$_2$, DMF, molecular sieves (4 Å), 30 min, 0 °C; HCl, THF, 12 h, 20 °C, 90%.

Scheme 4 was quenched with an excess of methyl iodide within 30 min at 0 °C. Subsequent cyclization to the hemiketal in acidic medium gave soraphen A$_1$ (4a) in 70% yield.

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[7] M. T. Keen, Angew. Chem. 1984, 96, 542; Angew. Chem. Int. Ed. Engl. 1984, 23, 556. When using the magnesium salt of acetylene only the desired trans-glycero isomer 11 was detected. The formation of the l-glycero isomer, however, was favored in a ratio of 8:1 when using the trisopropylsilyloxymethyl salt.

Further details of the crystal structure investigation are available on request from the Director of the Cambridge Crystallographic Data Centre, 12 Union Road, CB2 1EZ (UK), on quoting the full journal citation.

[8] Further details of the crystal structure investigation are available on request from the Director of the Cambridge Crystallographic Data Centre, 12 Union Road, CB2 1EZ (UK), on quoting the full journal citation.

[16] Upon treatment of soraphen A$_1$ (la) [2] with base (thermodynamic conditions), no change of configuration occurs at the methylated carbon a to the lactone. The crystal structure of 1a shows that the mesityl group of the tetrahydropyran ring and the methyl group a to the lactone are in a sterically favorable position relative to each other. In the corresponding epimer form, however, these methyl and methoxy groups are subject to 1,3-diaxial interactions. This might also be the reason why a possible dimethylation of norsoraphen 1b can be avoided under appropriate reaction conditions.

Novel Bimetallic Model System for Cytochrome P$_{450}$: Effect of Membrane Environment on the Catalytic Oxidation**

Albertus P. H. J. Schenning, Dominicus H. W. Hubert, Jan H. van Esch, Martinus C. Feiters, and Roeland J. M. Nolte*

Cytochrome P$_{450}$ catalyzes a variety of oxidation reactions, including the hydroxylation of alkenes and the epoxidation of alkenes.1,2 The active site of this membrane-bound enzyme contains a heme function and a thiolate as axial ligand. The catalytic cycle involves the binding of a substrate, reduction of iron(I) to iron(III) and hydroxide, and reductive cleavage of molecular oxygen to generate what is formally an oxoiron(v) complex, according to the mechanism proposed by Ruston.3-5

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which transfers its oxygen atom to the bound substrate. The iron center accepts the electrons from NADPH, through mediation of a flavoprotein. Because of its biological importance—this enzyme plays a crucial role in the metabolism of endogenous chemicals and xenobiotic compounds—and also because this archetype oxidation catalyst may serve as a model for a new generation of synthetic catalysts, a great deal of research is currently focused on mimicking the action of cytochrome P450. Until now only a few models have been described that incorporate the important features of the natural system, namely molecular oxygen as the oxidant, a metalloporphyrin as catalyst, an electron donor, and a membrane system holding these components. All models have the disadvantage of displaying very low catalytic activity.

During the course of our studies on novel supramolecular catalytic systems, we found that the rhodium complex [Rh(H\(\text{C}_2\text{O})(\text{bpy})\text{Cl}_2)] (\(\text{C}_2\text{O} = \text{pentamethylcyclopentadienyl}, \text{bpy} = 2,2'\text{-bipyridine}\)) is an efficient catalyst for the reduction of manganese(II) porphyrins by sodium formate. We report here on a membrane-bound cytochrome P450 mimic that epoxidizes alkenes with good turnover numbers. The system is composed of vesicles containing an \(\alpha\)-(aceto)-[5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrinato]manganese(III) catalyst ([MnIII-ta\(\text{c}_5\text{Cl}_2\text{Cl}^-\text{P})\text{Cl}_2\]) and a membrane system. EPR studies, using [Cu(t2\(\text{Cl}_2\text{PP})]\) instead of the oxidant, a metalloporphyrin as catalyst, an electron donor, and a membrane system holding these components. All models have the disadvantage of displaying very low catalytic activity.

The manganese porphyrin I and the rhodium complex 2 were incorporated into positively charged dimethyl-dioctadecylammonium chloride (DODAC) and negatively charged dihexadecylphosphate (DHP) vesicles by the ethanol injection method. Gel permeation chromatography and electron microscopy were used to show that the porphyrin and the rhodium complex were bound to the bilayers of the vesicles and that the vesicle structure was not destroyed by the incorporation procedure. The diameters of the vesicles of both amphiphiles amounted to about 4000 Å. The fluorescence spectra of H\(_2\text{t}_{12}\text{a}_5\text{Cl}^-\text{CIPP}\) in both DODAC and DHP vesicles (\(T = 70 \text{ °C}\)) showed no effects of self-quenching when the porphyrin to lipid ratio was less than 0.005. This indicates that below these ratios the porphyrin molecules are not aggregated. The addition of water-soluble quenchers (NaI and CuSO\(_4\)) in the case of DODAC and DHP vesicles, respectively, to the system did not change the fluorescence spectrum of the vesicle-bound H\(_2\text{t}_{12}\text{a}_5\text{Cl}^-\text{CIPP}\). Most likely the porphyrin is situated in the inner part of the vesicle bilayer. EPR studies, using [Cu(t2\(\text{Cl}_2\text{PP})]\) instead of I revealed that for both vesicle systems the orientation of the porphyrin molecules was parallel to the vesicle surface. The structure of the model system is depicted in Figure 1.

First we investigated the influence of the membrane matrix on the reduction of the manganese(III) porphyrin. To this end the decrease in the absorption at 660 nm (MnIII porphyrin) and the increase in absorption at 448 nm (MnII porphyrin) in the UV/VIS spectrum was followed as a function of time, both under an argon atmosphere and in air. The rhodium complex manganese-porphyrin ratio was varied from 0.5 to 10. Under argon in DODAC as well as in DHP vesicles, the MnIII porphyrin was reduced at all Rh/Mn ratios tested. The reduction rate was found to increase linearly with the RhIII concentration. At a fixed Rh/Mn ratio the reduction was faster in DODAC vesicles than in DHP vesicles ([Rh]/[Mn] = 1; \(k_0\) (DODAC) > 100 mmol L\(^{-1}\) s\(^{-1}\), \(k_0\) (DHP) = 20 ± 2 mmol L\(^{-1}\) s\(^{-1}\)). The reduction of MnIII in DODAC vesicles also took place when the reactions were carried out in an air atmosphere. A similar result was obtained for DHP except when the Rh/Mn ratio was equal or less than 1, under which conditions reoxidation of MnIII was observed. In the absence of the RhIII complex or formate no reduction took place. These results indicate that the rhodium-formate system is capable of reducing the membrane-bound MnIII porphyrin and presumably, as observed previously, the rhodium complex acts as a redox-active phase transfer catalyst in this process. The reduction of MnIII is faster in the positively charged vesicles than in the negatively charged ones because the formate concentration is higher at the bilayer/water interface of the former aggregates. The fact that in DODAC vesicles in general and in DHP vesicles with a Rh/Mn ratio higher than 1 no reoxidation of MnIII takes place, indicates that the reduction of manganese is much faster than its reoxidation.

In a second series of experiments we investigated whether our membrane-bound cytochrome P450 mimic was able to epoxidize alkenes. The results are listed in Table 1. All substrates tested were epoxidized by the catalytic system based on DHP vesicles. The turnover numbers are higher than those obtained with the two-phase system previously published by us. They are in the same range as those observed for the natural system (1 nmol product per nmol P450 per min). Remarkable is the high stability of the catalyst during the reaction, which is in contrast to the two-phase system. The effect of the membrane environment on the catalytic epoxidation became clear when the DHP vesicles were replaced by the DODAC vesicles. In the latter membrane system no epoxidation of alkenes was observed (last two entries of Table 1). Presumably, the concentration of protons is too low to allow the formation of the catalytically active oxomanganese(v) species at the positively charged interface. When the Rh/Mn ratio was increased from 1 to 10 the turnover number of the reaction decreased considerably (Table 1, entry 5). This phenomenon is possibly caused by the fact that more elec-
### Table 3. Epoxidation of alkenes by the membrane-bound cytochrome P₄₅₀ mimic

<table>
<thead>
<tr>
<th>Entry</th>
<th>Surfactant</th>
<th>Substrate</th>
<th>Product [b]</th>
<th>Turnover number [c]</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>DHP</td>
<td>α-pinene</td>
<td>DHP</td>
<td>45</td>
</tr>
<tr>
<td>2</td>
<td>DHP</td>
<td>α-pinene</td>
<td>a-pinene oxide</td>
<td>45</td>
</tr>
<tr>
<td>3</td>
<td>DHP</td>
<td>limonene</td>
<td>DHP</td>
<td>45</td>
</tr>
<tr>
<td>4</td>
<td>DHP[c]</td>
<td>styrene</td>
<td>styrene oxide</td>
<td>[d]</td>
</tr>
<tr>
<td>5</td>
<td>DHP[c]</td>
<td>styrene</td>
<td>DHP</td>
<td>0</td>
</tr>
<tr>
<td>6</td>
<td>DODAC</td>
<td>styrene</td>
<td>styrene oxide</td>
<td>0</td>
</tr>
<tr>
<td>7</td>
<td>DODAC[c]</td>
<td>styrene</td>
<td>DHP</td>
<td>0</td>
</tr>
</tbody>
</table>

[a] For reaction conditions see Experimental Procedure. The reaction was followed by GLC for 1 h. No destruction of the catalyst was found after that period. The only product of the reaction was epoxide, which was gradually decomposed into other products. The decomposition processes were independent of the Rh/Mn ratio. No epoxide was formed without vesicles or when any of the components of the catalytic system were omitted. [c] Turnover number [epoxide] 1 h, calculated from the initial part of the conversion/time plot. α-Pinene oxide was converted (>90%) into pinocamphone within a period of one hour. Limonene oxide decomposed (± 50%) into as yet unidentified products. [d] Styrene oxide and stilbene oxide were stable under the experimental conditions. [e] Rh/Mn molar ratio = 10.0.

**Experimental Procedure**

Reduction experiments: The desired amounts of stock solutions of 1, 2, N-methylimidazole, and DHP or DODAC in chloroform were mixed in a test tube. The solvent was evaporated under a stream of nitrogen to leave a homogeneous film. This film was solubilized in 100 μL ethanol/tetrahydrofuran (1:1, v/v) and injected in 1.25 mL water at 75°C. The suspension was purged with argon for 30 min and injected in a cuvette containing 1.25 mL of an ethylmorpholine/sodium formate buffer at 75°C. Final conditions: 2.4 μM of 1, 2.4 × 10⁻⁵ M of 2 (in Rh/Mn molar ratio), 0.1 μM DODAC or DHP in ethylmorpholine (50 mM)/sodium formate (250 mM) buffer (pH = 7.0), T = 70°C. Epoxidation experiments: Final conditions as above, except Rh/Mn molar ratio = 1 and [substrate] = 200 μM. Now the 100 μL solution was directly injected in a 2.5 mL buffered solution at 75°C. Substrate was added and the reaction mixture was analyzed from time to time by taking a 0.2 mL aliquot to which was added 0.1 mL diethyl ether containing mesitylene as an internal standard. This mixture was shaken vigorously (vortex apparatus) and centrifuged. After phase separation, a 5 μL sample was taken from the diethyl ether layer and analyzed by GLC (columns Chrompack, WCOT/CP-SILCB, temperature program 70°C (2 min), 10 K min⁻¹ to 200°C (2 min)).

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**Scheme 1. Cycloaromatization of enediynes.**

**Scheme 2. Cycloaromatization of cumulene-enzymes.**

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1,4-Didehydrobutadiene: The Intermediate in a New Class of Thermally Induced Intramolecular Alkyne Couplings

Rolf Gleiter* and Joachim Ritter

Dedicated to Professor Heinz Dörr on the occasion of his 60th birthday

More than twenty years ago Bergman and co-workers discovered that enediynes such as (Z)-hexadiyn-3-ene (1) cyclize to give the corresponding 1,4-didehydrobenzenes (2) as short-lived intermediates. This reaction occurs at 200°C with an activation energy of 28 kcal mol⁻¹ (Scheme 1). The recent discovery of the natural antitumor antibiotics calicheamicin, esperamycin, and dynemicin, which contain the cyclic enediyne unit 3, led to an enormous increase in publications on enediyne chemistry in the last seven years. A common feature in the mechanism of action of these antibiotics is the cycloaromatization of the enediyne unit 3 to give the bicyclic 1,4-didehydrobenzene 4. In the case of neocarzinostatin, the electrocyclization of an initially generated cyclic (Z)-cumulene-enzyme 7 is proposed as the key step. In related work Myers et al. found that the surprisingly facile cyclization of the open-chain cumulene 5 gives 1,3-didehydrodiene 6 and is an example of a new type of a 1,4-biradical generating reaction (Scheme 2).

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**References**