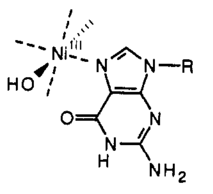
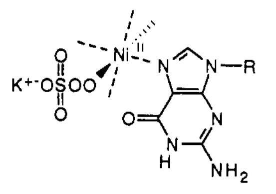
be demonstrated. A second possible role for Ni^{II} is as a Lewis acid for activation of a peracid toward oxidative attack at a guanine. In this case then, Ni^{II} might serve as a template for coordination of both substrate and reactant as shown in structure B. The successful ligands, L_1, L_3, and L_4, are those that provide an intermediate ligand field strength, allowing for formation of either square-planar or octahedral species. Thus, the important criteria for intrinsic reactivity of Ni^{II} complexes are (i) availability of vacant coordination sites through a square-planar geometry, (ii) overall positive charge on the complex, and (iii) a relatively high reduction potential of the Ni^{III} state. Further verification of these hypotheses through a systematic study of ligand effects is in progress.



A



B

In support of a nickel–guanine complex, oxidation is specific for only freely accessible residues. When 1 was hybridized to its complement and then subjected to oxidation, only a single G reacted, the 3′-terminal guanine (data not shown). This reagent should therefore prove to be quite useful as a probe for unusual DNA structures.[17]

Acknowledgment. Support of this work through a Seed Grant from the Stony Brook Center for Biotechnology sponsored by the New York State Science and Technology Foundation (to S.E.R. and C.J.B.) and by grants (to C.J.B.) from the National Science Foundation (CHE-9006684) and National Institutes of Health (GM-34841) is gratefully acknowledged.

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Olefin Formation in the Oxidative Deformylation of Aldehydes by Cytochrome P-450. Mechanistic Implications for Catalysis by Oxygen-Derived Peroxide

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Received January 22, 1991

We describe the cytochrome P-450 mediated oxidative deformylation of a xenobiotic aldehyde with introduction of unsaturation into the residual carbon framework. The reaction with cyclohexanecarboxaldehyde is a useful model for the demethylation reactions catalyzed by the steroidogenic P-450s, aromatase and lanosterol demethylase, where formic acid and an olefinic product are also formed.[1]

The active oxidant in P-450 catalyzed reactions is generally thought to be a pentavalent oxoiron species, or "iron oxene".[2] This concept fails, however, to account for the oxidative carbon–carbon bond cleavage step in steroid demethylation reactions. Alternatively, various investigators have suggested a role for an O_2-derived peroxide in the P-450 catalyzed cleavage of the oxysteroid intermediate.[3] However, no direct evidence for the role of peroxide in these reactions has been provided. H_2O_2 and organic peroxy compounds can be substituted for O_2 and NADPH in many P-450 catalyzed reactions,[2,4] but in the deformylation herein reported

Table I. Component Requirements and Effects of Catalase and Superoxide Dismutase on the Formation of Cyclohexene from Cyclohexanecarboxaldehyde

system[a]	act., (nmol/min)/(nmol of P-450)
Experiment 1	
complete	0.30 ± 0.04
NADPH omitted	0.02 ± 0.00
O_2 concn reduced (4.0 μM)	0.05 ± 0.00
reductase omitted	0.02 ± 0.00
P-450 LM_2 omitted	0.00 ± 0.00
DLPC omitted	0.07 ± 0.00
Experiment 2	
complete	0.26 ± 0.02
catalase added (240 units)	0.29 ± 0.02
catalase added (960 units)	0.27 ± 0.01
superoxide dismutase added (60 units)	0.28 ± 0.00
superoxide dismutase added (360 units)	0.29 ± 0.00

[a] The complete system contained 0.25 nmol each of the reductase and P-450 LM_2, 30 μg of DLPC, 50 μmol of potassium phosphate buffer, pH 7.4, 1.0 μmol of cyclohexanecarboxaldehyde, and 2.0 μmol of NADPH as the final addition in a 1.0-mL reaction volume. The vessel was sealed with a rubber septum and incubated at 37 °C for 10 min. The reactions were quenched by the addition of 100 μL of 30% perchloric acid, and the cyclohexene was quantitated by gas chromatography. Each experiment was carried out in triplicate and corrected for a blank in which the enzymes had been heat-denatured prior to addition.

Table II. Effectiveness of Other Oxidants in the Cytochrome P-450 Catalyzed Formation of Cyclohexene from Cyclohexanecarboxaldehyde

oxidant added[a]	concn, mM	cyclohexene formed, nmol
hydrogen peroxide	0.10	0.19 ± 0.06
hydrogen peroxide	0.50	0.91 ± 0.05
iodosobenzene	0.01	nd[b]
iodosobenzene	0.05	nd
m-chloroperbenzoic acid	0.01	nd
m-chloroperbenzoic acid	0.05	nd
cumyl hydroperoxide	0.10	nd
cumyl hydroperoxide	0.50	nd

[a] The reactions were as described in Table I except that the reductase, NADPH, and phospholipid were omitted. Reactions were initiated by the addition of a 10 mM aqueous solution of the oxidant or, in the case of iodosobenzene, a methanolic solution. The volume of methanol used was known not to affect the formation of cyclohexene in the complete system as described in Table I. After incubation for 3 min, reactions were quenched by the addition of 100 μL of saturated aqueous sodium thiosulfate. With H_2O_2 the reaction is linear with time for 3 min. In other experiments the inactivity of the three organic oxidants was shown not to be due to P-450 destruction. [b] Not detected (limit of detection, 50 pmol).

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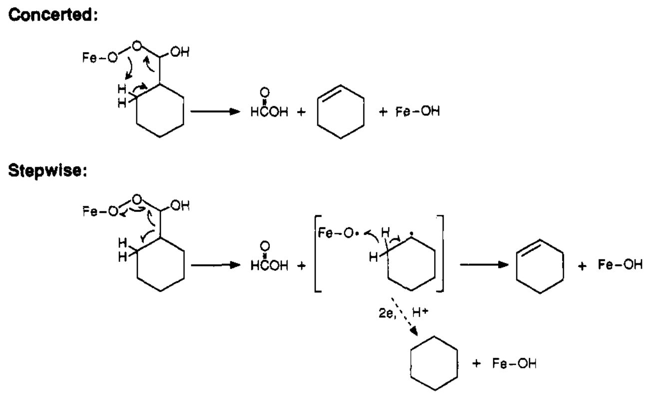
Scheme I



only H_2O_2 was active. We conclude that "iron oxene" is not the active oxidant in this reaction and propose that a peroxyhemiacetal-like adduct generated from the aldehyde and O_2-derived peroxide is a transient enzyme-bound intermediate. Rearrangement of this peroxy intermediate by either a concerted or sequential β-scission mechanism then yields formic acid and the olefin.

Cyclohexene was identified by gas chromatography/mass spectroscopy in the head-space gas of a reaction mixture containing cyclohexanecarboxaldehyde, NADPH, P-450 LM_2,[5,7] reductase,[8] and DLPC. Cyclohexanol, cyclohexyl formate, and cyclohexanecarboxylic acid (which is formed in the reaction) did not yield cyclohexene when substituted for the aldehyde. With substrate labeled with ^{14}C at the aldehyde carbon, formate is formed in about an equimolar amount with respect to cyclohexene.

When either of the enzymes or NADPH was omitted, the amount of cyclohexene formed was insignificant (Table I). A small amount of cyclohexane (about 10% of the amount of cyclohexene) was detected with the complete system. At low O_2 concentration (4.0 μM), an 83% decrease in activity was observed, and omission of phospholipid[9] gave a 77% loss. Added catalase and superoxide dismutase had no effect.[10]

Hydrogen peroxide supports the cytochrome P-450 dependent oxidative deformylation of cyclohexanecarboxaldehyde to cyclohexene (Table II). In experiments not shown, formate was found to be produced in the H_2O_2-supported reaction in about an equimolar amount with respect to cyclohexene. The inability of iodosobenzene to support the reaction suggests that the generally accepted pentavalent "iron oxene"[4f] is not the oxidant. Furthermore, the results with *m*-chloroperbenzoic acid and cumyl hydroperoxide indicate that deformylation by H_2O_2 is mechanistically distinct from hydroxylation reactions supported by these oxidants.[4c-e]

In Scheme I, we propose that an O_2-derived, heme iron bound peroxide reacts with the electrophilic aldehyde carbonyl group to form an enzyme-bound peroxyhemiacetal-like intermediate,[3a] rearrangement of which yields the olefin and formic acid by a concerted or a sequential β-scission mechanism. The small amount of cyclohexane formed is accounted for by reduction of the carbon radical, as in the β-scission of hydroperoxides to yield alkanes.[11]

(5) Abbreviations: P-450 LM_2, phenobarbital-inducible rabbit liver microsomal cytochrome P-450, also designated as P-450 IIB4;[6] DLPC, dilauroylglycero-3-phosphocholine; reductase, NADPH-cytochrome P-450 reductase.

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In experiments not presented, oxidative deformylation to yield olefins was shown to occur with other cyclic and acyclic aldehydes in the presence of P-450 LM_2. Of eight purified P-450 isozymes examined, four were catalytically active in deformylation of cyclohexanecarboxaldehyde to cyclohexene. Oxidative deformylation may be a commonly encountered pathway in the metabolism of xenobiotic aldehydes.

Acknowledgment. This investigation was supported by Grant DK-10339 from the National Institutes of Health. E.S.R. was a Predoctoral Trainee, Pharmacological Sciences Training Grant T32 GM-07767, National Institutes of Health.

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Substituted 2,2′:5′,2″:5″,2‴:5‴,2′′′′:5′′′′,2′′′′′:5′′′′′,2′′′′′′:5′′′′′′,2′′′′′′′:5′′′′′′′,2′′′′′′′′:5′′′′′′′′,2′′′′′′′′′:5′′′′′′′′′,2′′′′′′′′′′-Undecithiophenes: The Longest Characterized Oligothiophenes

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Received October 29, 1990

Polythiophenes figure prominently in current research on conducting polymers[1] and are of interest as nonlinear optical materials.[2] In both cases the properties depend critically on the effective conjugation length that can be realized in the polymers, i.e., on their structure and conformation. It is generally assumed that the polymerization of thiophenes leads to regular polymers in which the thiophene units are linked at the α-positions.[3] However, little is known about the degree to which deviations from this ideal behavior occurs. For unsubstituted polythiophenes containing seven or more thiophene units, spectral characterization is virtually impossible due to their insolubility.[4] Solubility in regular organic solvents is obtained when polymers derived from 3-alkylthiophenes are prepared.[5] However, adjacent alkyl substituents give rise to steric hindrance and, hence, to a nonplanar conformation.[6] Moreover, the degree of regularity depends

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