Oxygen Exchange as a Function of Epimerization of 15(*R*)- and 15(*S*)-Methylprostaglandin E_2. Evidence for Asymmetric Carbonium Ion Intermediates

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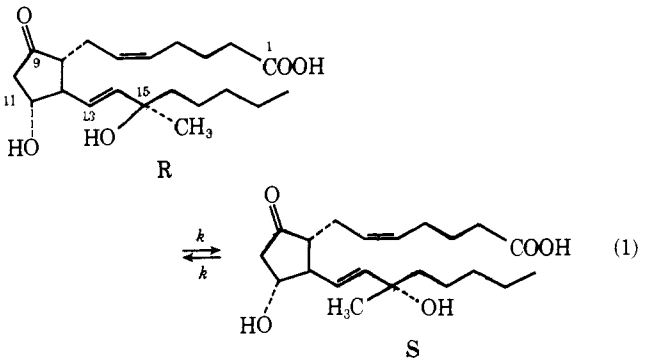
Abstract: The rate of oxygen exchange of each epimer as a function of epimerization of 15(*R*)-methylprostaglandin E_2 (R) and 15(*S*)-methylprostaglandin E_2 (S) (tertiary alcohols) in ^{18}O-enriched water has been determined by LC and GC–MS methods. The relative rates indicate that exchange of the C-15 hydroxyl group is more rapid than the change in configuration at C-15. These data are consistent with a mechanism for epimerization in which the initially formed carbonium ion formally retains the configuration of starting material. A significant percentage of the initially formed epimerized product does not exchange with the ^{18}O-enriched water. This result indicates that, in some cases, the departing water molecule ultimately bonds the carbonium ion from the opposite face. A reaction scheme is proposed in which the waters of solvation exchange with the departing water more rapidly than the initially formed carbonium ion epimerizes.

Introduction

We have reported on the kinetics of the epimerization of 15(*R*)-methylprostaglandin E_2 (R) and 15(*S*)-methylprostaglandin E_2 (S) in aqueous solution.[1] The equilibrium constant was found to be 1.02 ± 0.02. The rate of epimerization, k_{epim}, exhibited a first-order dependence on both hydrogen ion and substrate concentration. This dependence is typical of allylic alcohols in which racemization, in the case of optically active alcohols, proceeds through a common carbonium ion intermediate.



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The aspect of the epimerization of R and S atypical of allylic alcohols was the complete absence of rearrangement products. In general, the acid-catalyzed rearrangement of a tertiary α-allylic alcohol to a primary or secondary alcohol is quite facile. We suggested that the large steric crowding at C-13 was a major factor in preventing allylic rearrangement during epimerization.[1]

The absence of reactions competing with epimerization and our ability to separate the diastereomers R and S afforded us a unique opportunity to examine the intimate details of the change in configuration at the α carbon in a tertiary alcohol. We report here the rate constants for the following processes as a function of epimerization and the mechanistic interpretation of these results:

$$ROH + H_2{}^{18}O \xrightarrow{k_{R-ex}} R{}^{18}OH \quad (2)$$

$$ROH + H_2{}^{18}O \xrightarrow{k_{R-ep+ex}} S{}^{18}OH \quad (3)$$

$$SOH + H_2{}^{18}O \xrightarrow{k_{S-ex}} S{}^{18}OH \quad (4)$$

$$SOH + H_2{}^{18}O \xrightarrow{k_{S-ep+ex}} R{}^{18}OH \quad (5)$$

ROH and $R^{18}OH$ symbolize R with oxygen in natural abundance and enriched in ^{18}O at C-15, respectively. SOH and $S^{18}OH$ symbolize S with oxygen in natural abundance and enriched in ^{18}O at C-15, respectively.

Studies of the exchange of ^{18}O between water and optically active alcohols have provided much of the experimental basis for our current understanding of nucleophilic substitution and of the structure of carbonium ion intermediates. Previous studies of oxygen exchange in alcohols as a function of racemization have measured only the total exchange of the unresolved mixture of diastereomers.[2-6] These results are, to our knowledge, the first report of the rate constants for oxygen exchange for each member of the pair of compounds diastereomeric at the α carbon of an optically active alcohol as a function of racemization at this center.

Experimental Section

Chemicals. The prostaglandins were obtained from the research laboratories of The Upjohn Co. Water enriched in ^{18}O (ca. 30 atom % enrichment) was purchased from Miles Biochemicals, Elkhart, Ind. Methoxyamine hydrochloride and bis(trimethylsilyl)trifluoroacetamide (BSTFA) were obtained from Applied Science and Regis, respectively.

Equipment. All chromatography was performed on a Varian 8500 dual pump liquid chromatograph with a Model LC-55 Perkin-Elmer UV detector equipped with a 25-cm silver ion loaded microparticulate cation exchange resin column as previously described.[1] The ^{18}O content of the epimers was determined by mass spectrometry using a fully computerized GC–MS method with an LKB-9000 interfaced to an IBM-1800 computer.[7] The GC column used throughout this study was a 2-ft 3% SE-30 on Gas Chrom Q, 60–80 mesh, operated at 280 °C.

Samples for kinetic runs were thermostated in a water bath with the temperature controlled to 37.20 ± 0.01 °C with a Haake Model E52 temperature controller calibrated with an NBS thermometer.

Procedures. The data reported here are the results of two kinetic runs in which the rates of epimerization and ^{18}O incorporation into each epimer were determined. R was the starting material in one run; S was the starting material in the second.

The rate of epimerization was determined by the previously reported LC procedure[1] modified to determine the ^{18}O content of each epimer for each kinetic point. Accurately weighed samples of prostaglandin and Na_2SO_4 were dissolved in ^{18}O-enriched water to give concentrations of 0.500 mg/mL and 0.033 M, respectively. After the solution reached thermal equilibrium in the 37.20 °C bath, the kinetic run was initiated by addition of 5 N HCl (14 μL/7.00 mL) to give a pH of 2.2–2.4. At predetermined times, aliquots were removed, quenched, and extracted. The prostaglandins were then converted to their *p*-nitrophenacyl esters. The epimeric esters were separated and collected via liquid chromatography on a silver-ion column. The relative con-

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Table I. Epimerization of R and S. ^{18}O Exchange Data

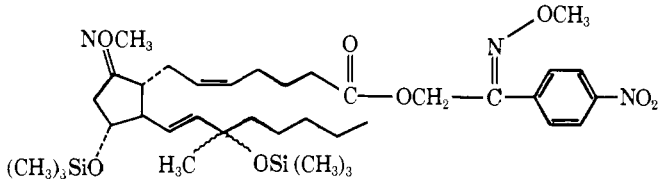
kinetic run	starting material	time, min	%[a] R	%[a] S	ΔI_t[b] R	ΔI_t[b] S	% enrichment[c] R	% enrichment[c] S	% epimer[d] $R^{18}OH$	% epimer[d] $S^{18}OH$
1	R	5	89.3	10.7	67.3	465.3	12.3	85.5	11.0	9.1
		10	84.5	15.5	105.4	476.4	19.3	87.5	16.3	13.6
		15	80.4	19.6	144.4	493.8	26.5	90.8	21.3	17.8
		25	73.2	26.8	212.7	499.8	39.0	91.9	28.5	24.6
		35	67.9	32.1	277.4	501.4	50.9	92.1	34.6	29.6
		45	64.5	35.5	331.0	505.4	60.7	92.9	39.2	33.0
		55	60.7	39.3	378.3	510.0	69.5	93.8	42.2	36.9
		65	58.8	41.2	411.0	513.8	75.4	94.4	44.3	39.0
		300	50.0	50.0	544.9 ± 9.0		100	100	50.0	50.0
2	S	5	4.8	95.2	370.1	29.2	79.5	6.3	3.8	6.0
		10	9.5	90.5	404.1	66.9	86.9	14.4	8.2	6.0
		15	13.3	86.7	418.7	93.8	88.9	20.2	12.0	17.5
		25	19.8	80.1	422.7	150.1	90.8	32.3	18.0	25.9
		35	25.5	74.4	423.7	202.2	91.1	43.5	23.2	32.4
		45	29.9	70.1	431.7	247.9	92.8	53.3	27.6	37.4
		55	33.8	66.1	431.1	287.7	92.6	61.8	31.3	40.9
		65	37.0	62.9	437.4	307.2	94.0	66.0	34.8	41.6
		300	50.0	50.0	465.3 ± 7.5		100	100	50.0	50.0

[a] % of prostaglandin present as total R ($R^{18}OH$ + ROH) or total S ($S^{18}OH$ + SOH) from LC data. [b] ΔI_t = difference in intensity of peak of *m/z* 489, normalized with respect to that of *m/z* 487, and that of the corresponding R or S standard. [c] % enrichment = $\Delta I_t/\Delta I_{t=300}$. [d] % $R^{18}OH = (\Delta I_t/\Delta I_{t=300})(R_t/(R_t + S_t))100\%$; % $S^{18}OH = (\Delta I_t/\Delta I_{t=300})(S_t/(R_t + S_t))100\%$.

centration of each epimer was determined from the chromatographic data. The pseudo-first-order rate constants for epimerization were calculated as before. For each kinetic run, eight points were taken in the interval from 5 to 65 min (ca. $2\tau_{1/2}$); an additional point was taken at 300 min ($10\tau_{1/2}$).

Volatile derivatives of each sample of the LC-separated *p*-nitrophenacyl esters of R and S were prepared for GC–MS analysis of the ^{18}O content as follows. The chromatographic mobile phase was removed with a stream of nitrogen and the sample dissolved in a measured volume of chloroform. An aliquot of this solution equivalent to 100 μg of prostaglandin, based on 100% recovery from the chromatographic separation, was transferred to a 1-mL Reactivial. Following nitrogen removal of the chloroform, 200 μL of a pyridine solution of methoxyamine hydrochloride (15 mg/mL) was added to the sample. The sample was sealed and allowed to react overnight. Following addition of 100 μL of BSTFA, the sample was heated at 65 °C for 4 h. Solvent and excess reagent were removed with a stream of nitrogen. The residue was dissolved in a volume of hexane to give a final concentration equivalent to ca. 0.5 μg/μL of prostaglandin. Unlabeled standards were prepared from the *p*-nitrophenacyl esters of R and S in the same fashion.

Mass spectra of these derivatives exhibited a parent peak at *m/z* 731, consistent with the molecular structure **1**.



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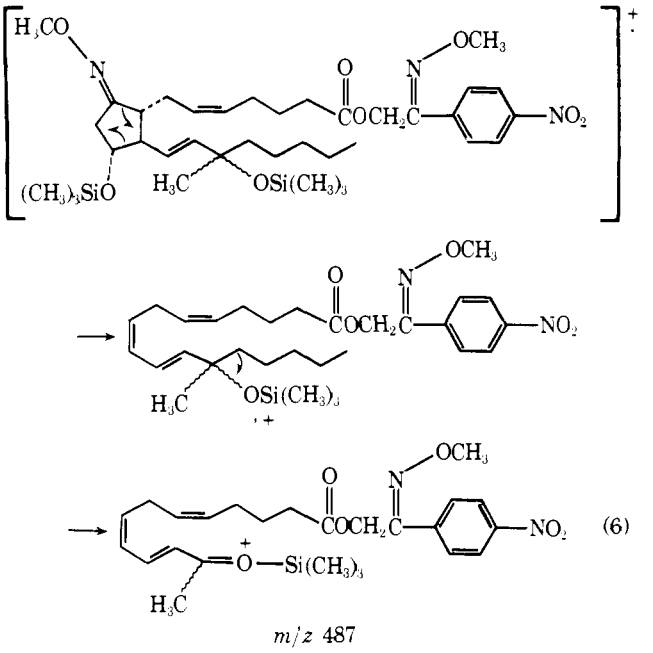
For ^{18}O analysis, our attention focused on a major fragment ion of *m/z* 487. The origin of this ion has been studied previously in the case of the methoxime-tris(trimethylsilyl) derivative of PGF_1.[8] It arises via fragmentation of the cyclopentane ring and loss of the C_5H_{11} side chain as shown in eq 6. This ion contains only one exchangeable oxygen, that at C-15.

The GC column, used to introduce the derivatives **1** of R and S into the mass spectrometer, separated these materials from the liquid ion exchanger used in the extraction procedure, excess reagent, and solvents. The epimeric derivatives 1 of R and S yielded a single GC peak with identical retention times under the chromatographic conditions used. The ^{18}O content of the derivatives was derived from the intensity of the peak at *m/z* 489, relative to that at *m/z* 487. The relative intensities of these peaks were obtained by repetitively scanning the *m/z* range from 487 to 491 over the width of the GC peak arising from **1**. The ion intensities of 20 scans/injection were averaged. The measurement of these intensities for each kinetic sample was bracketed by an identical measurement of **1** derivatives of the corresponding unlabeled R or S standard. The difference in these relative ion intensities (*m/z* 489 vs. 487) between the kinetic sample and those of the standard was used to calculate the ^{18}O enrichment. This procedure minimized errors arising from small changes in the resolution of the mass spectrometer. Duplicate measurements of each sample were made. Approximately 7 nmol of prostaglandin was required for each determination. It was assumed that the samples equilibrated for 300 min had undergone complete oxygen exchange. The percent oxygen exchange for all other samples in that kinetic run was calculated from the ^{18}O content of R and S at 300 min.



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m/z 487

II

Results

Table I contains the GC–MS data used to calculate the ^{18}O enrichment of each epimer and, hence, the rate of oxygen exchange. The percent enrichment at any time *t* is given by the expression

$$(\%\ \text{enrichment})_t = (\Delta I_t/\Delta I_{t=300})100\% \qquad (7)$$

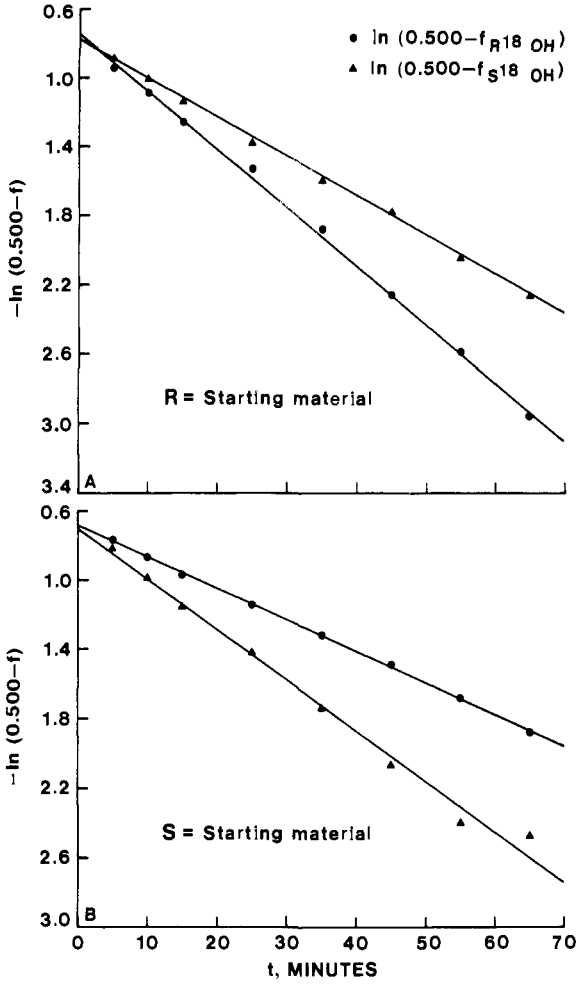


Figure 1. Linear least-squares plots of ln (0.500 − *f*) vs. time for oxygen exchange reaction. The top half of the figure (A) contains the data for the kinetic run in which R was the starting material. The bottom half (B) contains the data for the kinetic run in which S was the starting material.

ΔI_t is the difference in intensity of the peak at *m/z* 489, normalized with respect to that at *m/z* 487, between the derivative of the epimer in the kinetic sample and that of the corresponding standard of R or S. $\Delta I_{t=300}$ is the same parameter at t = 300 min.

The percent ^{18}O-labeled epimer at any time t was calculated from the percent enrichment at that time and the fraction of the starting material present as that epimer. The following expression was used to calculate the % R^{18}OH at time = t:

$$\%\ R^{18}OH = \frac{\Delta I_t}{\Delta I_{t=300}}\left(\frac{R_t}{R_t + S_t}\right)100 \qquad (8)$$

$$R_t = (\text{total R})_t = (ROH + R^{18}OH)_t$$

$$S_t = (\text{total S})_t = (SOH + S^{18}OH)_t$$

A similar expression was used to calculate the percent of prostaglandin present as S^{18}OH. The value for $R_t/(R_t + S_t)$ was taken directly from the LC data at time t. The rate constants for epimerization, $k_{epim} = 2k$ (eq 1), were determined from the LC data as previously described and are reported in Table II. The slopes of the linear least-squares plots of ln ($|\%\ ^{18}\text{O epimer}_t - \%\ ^{18}\text{O epimer}_{t=300}|$) vs. time yielded the rate constants for the pseudo-first-order processes represented in eq 2-5. The plots are shown in Figure 1. In addition, the rate constants for the processes formally represented in eq 9 and 10 were also determined in an identical fashion from plots of

Table II. Pseudo-First-Order Rate Constants of Epimerization[a] and ^{18}O Exchange at 37.2 °C, pH 2.3[b]

	rate constant × 10^2, min^{-1}, of starting material	
	R	S
k_{epim}	2.51 ± 0.03[c]	2.07 ± 0.01[c]
$k_{total\ ex}$	2.51 ± 0.01	2.27 ± 0.04
k_{ex}	3.32 ± 0.06	2.91 ± 0.12
k_{ep+ex}	2.20 ± 0.04	1.82 ± 0.02

[a] $k_{epim} = k_{epimerization}$. k_{ex} = rate constant for ^{18}O exchange with starting material; i.e., with R as the starting material $k_{ex} = k_{R\text{-}ex}$ in eq 2. $k_{total\ ex}$ = rate constant for ^{18}O exchange with both R and S. k_{ep+ex} = rate constant for ^{18}O exchange with product epimer; i.e., with R as the starting material $k_{ex} = k_{R\text{-}ep+ex}$ in eq 3. [b] The precision is reported as the standard deviation. [c] The previously obtained value for $k_{epim} = 2k$ was ($1.92 \pm 0.12 \times 10^{-2}$) min^{-1} at pH 2.35 and 37.2 °C (ref 1). The value for k_{epim} with R as the starting material differs significantly both from that value and from that obtained with S as the starting material in this study. This difference is most probably attributable to a slightly more acidic pH for this run; the rate corresponds to that predicted for pH 2.25.

Table III. Comparison of Rate of Epimerization with Rates of ^{18}O Exchange[a]

	starting material	
	R	S
k_{ex}/k_{epim}	1.32 ± 0.02	1.41 ± 0.04
$k_{total\ ex}/k_{epim}$	1.00 ± 0.01	1.10 ± 0.02
k_{ep+ex}/k_{epim}	0.88 ± 0.02	0.88 ± 0.01
k_{ex}/k_{ep+ex}	1.50 ± 0.02	1.60 ± 0.04

[a] Error expressed as standard deviation.

ln $|\%\ (R^{18}OH + S^{18}OH)_t - \%\ (R^{18}OH + S^{18}OH)_{t=300}|$ vs. time.

$$ROH \xrightarrow{k_{R-\text{total ex}}} R^{18}OH + S^{18}OH \qquad (9)$$

$$SOH \xrightarrow{k_{S-\text{total ex}}} R^{18}OH + S^{18}OH \qquad (10)$$

The minimum correlation coefficient was |0.9952|. These rate constants are contained in Table II. The ratios of the rate constants for epimerization to those for oxygen exchange are compiled in Table III. Since the rate constants for oxygen exchange are derived, in part, from the data used to determine k_{epim}, they are not independent of k_{epim}. The absolute values of k_{ex} and k_{ep+ex} for the two different kinetic runs are not directly comparable, whereas the ratios of rate constants contained in Table III are directly comparable.

We have previously discussed the use of liquid chromatography to monitor the rate of epimerization of R and S.[1] In that study we found the equilibrium constant for the epimerization shown in eq 1 to be 1.02 ± 0.02. In the present work, we found the ratios of R/S at t = 300 min (ca. $10\tau_{1/2}$) to be 1.03 ± 0.02 and 1.04 ± 0.2 for the kinetic runs starting with R and S, respectively. The rate constants of epimerization reported here (Table II) are in reasonable agreement with those obtained in the previous study. The differences in rate constants obtained using each of the epimers as the starting materials are ascribable to small differences in pH.

The fully computerized GC-MS method used to determine the extent of oxygen exchange was originally developed for quantitative determination of small amounts of drugs.[7] Isotope ratio measuring techniques of this sort have been called selected ion monitoring gas chromatography mass spectrometry (SIM-GCMS).[9] This method is based upon the accurate measurement of the intensities of selected ions of a drug and its deuterated analogues. As little as 200-300 pg of drug have

been quantitatively determined from this ratio. This computerized SIM-GCMS system seemed ideally suited for measuring the oxygen-18 content of the LC-isolated *p*-nitrophenacyl esters of R and S kinetic samples. Preliminary direct probe mass spectral examination of the *p*-nitrophenacyl ester of R, isolated after equilibration for 90 min in acidic ^{18}O-enriched water, indicated that oxygen exchange occurred at more than one site. The mass spectrum of this material, in comparison with the *p*-nitrophenacyl ester of a standard, exhibited intense P + 2 and P + 4 peaks fragment ions; the presence of the P + 4 ions demonstrated the exchange of two oxygens. Examination of known exchange rate constants indicated that ^{18}O could be incorporated into the ketone at C-9 as well as into the tertiary alcohol at C-15, but not into the secondary alcohol at C-11 nor the carboxylic acid at C-1 under the conditions of this experiment.[10] Consequently, we sought a derivative of the *p*-nitrophenacyl esters of R and S that would allow us to isolate the exchange reaction at C-15 from that at C-9 as well as to provide sufficient volatility for GC introduction into the mass spectrometer. The dimethoxime di(trimethylsilyl) ether derivatives **1** fulfulled both these requirements. Formation of the methoxime at C-9 eliminated this exchangeable site; the fragment of *m/z* 487 contains only one exchangeable oxygen, that at C-15. It should be noted that the GC column served only as a device to introduce the LC-separated R and S derivatives into the mass spectrometer. Liquid chromatographic examination of selected samples indicated that the epimeric purity of the separated esters was at least 98%.

Our derivation of the rate constants for oxygen exchange is based upon the assumption that the oxygen at the C-15 position has completely equilibrated with ^{18}O water at 300 min. Comparison of the measured isotopic composition of the prostaglandins with that of the water indicated the validity of this assumption. The isotopic composition of the ^{18}O-enriched water was determined via exchange with carbon dioxide and mass spectral examination of this gas.[11,12] For kinetic run 1 (Table I), the ^{18}O atom percent enrichment in the water was found to be 32.7 ± 0.5% vs. 35.2 ± 0.5% calculated from ΔI of derivatives **1** at 300 min. Correspondingly, for kinetic run 2, the ^{18}O atom percent enrichment in the water was found to be 31.7 ± 0.7% vs. 31.7% ± 0.3% calculated from $\Delta I_{t=300}$ for derivatives **1**. Repetitive examination of the derivatized samples of R and S obtained at 300 min in the kinetic runs indicated a high precision in our assay of ^{18}O composition (see Table I). The relative intensities of the *m/z* 489 peak vs. that at *m/z* 487 of each of the two epimers isolated at 300 min were averaged; the relative standard deviation for each run was 1.6%. Of the experimental parameters used in deriving the rate constants for oxygen exchange, an undetected determinant error in $\Delta I_{t=300}$ would produce the largest error. Any increase in $\Delta I_{t=300}$ decreases the ratio of both k_{ex}/k_{epim} and k_{ex+ep}/k_{epim}. For example, if we let $\Delta I_{t=300}$ increase 5%, then k_{ex}/k_{epim} = 1.10 ± 0.03 and k_{ex+ep}/k_{epim} = 0.79 ± 0.03 for the kinetic run 1. The value of $\Delta I_{t=300}$ must be increased by 10% for k_{ex}/k_{epim} to equal unity. Decreases in $\Delta I_{t=300}$ increase the ratio of both k_{ex}/k_{epim} and k_{ex+ep}/k_{epim}. Assuming that exchange is complete in 65 min (an unreasonable assumption since $\Delta I_{t=300} > I_{t=65}$) and letting $\Delta I_{\infty} = I_{t=65}$, we find that k_{ex}/k_{epim} = 1.63 ± 0.03 and k_{ex+ep}/k_{epim} = 1.05 ± 0.04 for the kinetic run using R as the starting material.

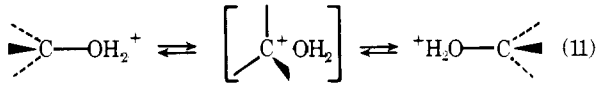
Examination of the relative intensities of the mass spectral peaks from the **1** derivatives of the unlabeled R and S standards allowed us to estimate the absolute error in the intensity data. The calculated intensity of the *m/z* 489 peak for the natural abundance ion II ($C_{25}H_{35}N_2O_6Si$) is 103 (intensity of *m/z* 487 = 1000). The measured intensity of the *m/z* 489 peak for the standards examined simultaneously with the samples from *t* = 300 min was 103.4 ± 3.01. Similarly, the mean intensity of this peak of the standards run simultaneously with the remaining kinetic samples ranged from 105.3 ± 2.2 for the data used to determine k_{ex} with R as the starting material to 108.0 ± 2.0 for the data used to calculate k_{ex+ep} starting with S. The 2-5% difference between the measured and the expected intensity probably results from incomplete resolution of the mass spectral peaks. To minimize the effects of changing resolution, we calculated the ^{18}O enrichment from the difference between the relative ion intensities (*m/z* 489 vs. 487) of each kinetic sample and those of two standards bracketing that sample. Consequently, the error in the ^{18}O enrichment is probably less than ±5%.

It is of interest to compare the SIM-GC method with other techniques for measuring isotopic composition of organic compounds. Matthews and Hayes have recently reported a new technique for measuring isotope ratios of submicromolar quantities of organic compounds.[13] In this technique, which they call isotope-ratio-monitoring gas chromatography-mass spectrometry (IRM-GCMS), a combustion furnace is inserted between the gas chromatograph and a GC-MS interface of an isotope ratio mass spectrometer to monitor the CO_2 ion currents due to the combustion products. The intensities of $^{13}CO_2$ current relative to those of $^{12}CO_2$ are used to provide the isotopic analysis of organic compounds enriched in ^{13}C. The combustion of the derivatives **1** (molecular formula $C_{37}H_{61}N_3O_8Si_2 + \Delta \rightarrow C_{37}O_{74}$), completely exchanged with 30% ^{18}O-enriched water, would yield an enrichment in ^{18}O of 0.4%, whereas the enrichment of the kinetic samples would be between 0.025 and 0.30%. The former value, 0.025%, is comparable to the standard deviation of the IRM-GCMS method. In contrast, the values of ΔI obtained in the kinetic run using S as the starting material ranged from ca. 4 to 40 times the standard deviation ($29.2 < \Delta I_t < 307.2$ vs. a standard deviation of 7.5). Consequently, for the assay of the ^{18}O content in R and S, the particular SIM-GCMS technique used here appears to be as good as the IRM-GCMS method in terms of precision. The latter method is, however, capable of more accurate determination of absolute isotopic enrichment.

It should be noted that the sensitivity of our analytical methods has permitted the use of much lower substrate concentrations (3×10^{-4} M) than those of earlier studies of oxygen exchange of optically active alcohols ($0.4\text{-}9 \times 10^{-1}$ M).[2-6] Less than 1.0 mg of prostaglandin was used in each kinetic run. Earlier studies of oxygen exchanges in alcohols relied on the conversion of much larger amounts (two to three orders of magnitude) of isolated compounds to CO_2; the ^{18}O content of CO_2 was determined using isotope ratio mass spectrometers.

Discussion

Studies of the rate of exchange of ^{18}O between water and optically active alcohols as a function of racemization have provided evidence for the intermediacy of ion pairs, or, more correctly, ion dipole pairs in these processes.[2-6] For the process involving the formation of a single carbonium ion as the intermediate, such as **2** that is shown in eq 11, $k_{exchange}$ =



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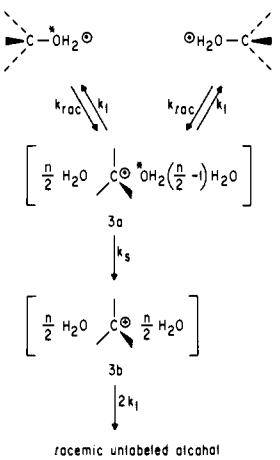
$k_{racemization}$ (or $k_{epimerization}$ in this case). Examination of only the ratio $k_{total\ ex}/k_{epim}$ for R and S (Table III) suggests that the change in configuration proceeds by the simple S_N1 mechanism involving a single carbonium ion shown in eq 11. The relative rate constants for oxygen exchange of the individual epimers reveal, however, a more complicated process: $k_{ex}/k_{epim} > 1$ and $k_{ep+ex}/k_{epim} < 1$ (Table III). The unique and most surprising result from this study is that k_{ex}/k_{epim} =

1.32 ± 0.02 and 1.41 ± 0.04 with R and S as the starting material, respectively. These values suggest that the initially formed carbonium ion retains its original asymmetry. In earlier studies of ^{18}O exchange in optically active alcohols, it had been assumed that $k_{ionization} = k_{racemization}$. In other words, the molecule racemizes with formation of a carbonium ion. Our data indicate, however, that, in the case of R and S, the rate of carbonium ion formation is faster than the rate of epimerization, i.e., $k_{ex}/k_{epim} > 1$. Implicit in this statement is the assumption that the rate of carbonium formation equals the rate of ionization.

The fact that $k_{ex+ep}/k_{epim} < 1$ means that some of the product is formed by attack of the departing water on the opposite face of the carbonium ion. In other words, the departing water becomes part of the solvation sphere and is available for forming the epimeric product. This phenomenon is most apparent at early times in the kinetic run. For example, the % ^{18}O enrichment in R, with S as the starting material, is only 79.5% at 5 min; 20.5% of the product contains the original water. A similar result was noted in the irreversible rearrangement of ^{18}O-labeled γ-phenylallyl alcohol to give α-phenylallyl alcohol containing a slightly greater ^{18}O abundance than the water in the solvent.[5] This excess indicated that some of the product resulted from the recombination of the partners formed in the dissociation. An additional precedent is found in the study of the racemization of optically active ^{18}O-enriched 1-phenylethyl alcohol. Grunwald, Heller, and Klein found that the ratio of rate constants for oxygen exchange to that for racemization, k_{ex}/k_{rac}, was 0.82 ± 0.04.[3] The observed ratio of k_{ex}/k_{rac} less than unity was attributed to shielding by the departing $-OH_2$ group. By analogy to Winstein's ion pair scheme for solvolysis,[14] they suggested that the racemization occurs through a solvated carbonium ion in which the solvation sphere contains six water molecules ($n = 6$) capable of reacting with the intermediate as shown in eq 12. The labeled atom (different oxygen isotope than that of the solvent water) is indicated with an asterisk. In this scheme, **3a** and **3b** are analogous to the intimate and solvent-separated ion pairs, respectively, of the solvolysis scheme.

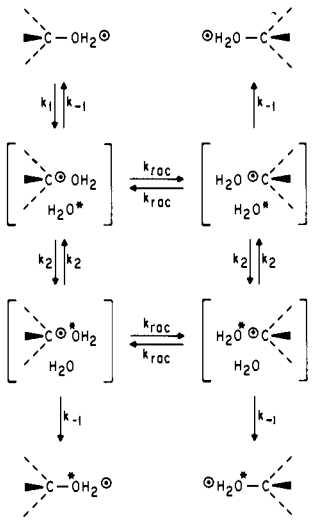


(12)

Our kinetic data cannot be fitted to the mechanistic scheme shown in eq 12; no values of the rate constants can result in a ratio of $k_{ex}/k_{epim} > 1$. A variety of similar schemes were examined in which the intermediate planar carbonium ions differed in the number of molecules in the solvation sphere and in the relative rate constants for exchange of these waters and for bond formation (k_s and k_1, respectively, in eq 12). None of these schemes was consistent with the value of k_{ex}/k_{epim} exceeding unity. Mechanistic schemes in which the initially formed carbonium ion formally retains its geometry can, however, yield a ratio of $k_{ex}/k_{epim} > 1$. A more convenient way of comparing our results with various models is to express the kinetic data as the ratio of k_{ex}/k_{ex+ep}. If one assumes that $k_{-1} > k_1$ and that the solvation sphere contains only two molecules which do not exchange with external water, it can be shown that

$$k_{ex}/k_{ex+ep} = \frac{(k_{-1} + k_{rac} + k_2)^2 - k_2^2 + k_{rac}^2}{2(k_{-1} + k_{rac} + k_2)k_{rac}} \quad (13)$$

for the reaction scheme illustrated in eq 14. This simple model



(14)

is consistent with our experimental data; i.e., $k_{ex}/k_{ex+ep} = 1.4$–1.6 (Table II), if either racemization or exchange of water is the rate-limiting step.

For the case in which racemization is the rate-limiting step, i.e., $(k_{-1} + k_2) \gg k_{rac}$, then

$$k_{ex}/k_{ex+ep} = \frac{k_{-1}(k_{-1} + 2k_2)}{2k_{rac}(k_{-1} + k_2)} > 1 \quad (15)$$

If exchange of the positions of the water molecules within the solvation sphere is the rate-limiting step, i.e., $(k_{-1} + k_{rac}) \gg k_2$, then

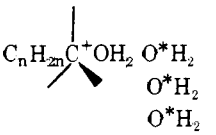
$$k_{ex}/k_{ex+ep} = \left[\frac{k_{-1}^2}{2k_{rac}(k_{-1} + k_{rac})}\right] + 1 > 1 \quad (16)$$

This model indicates that bond formation is not the rate-determining process. If the bond-forming step, k_{-1}, were the rate-limiting step, i.e., $(k_{rac} + k_2) \gg k_{-1}$, then

$$k_{ex}/k_{ex+ep} = \frac{2k_{rac}(k_{rac} + k_2)}{2k_{rac}(k_{rac} + k_2)} = 1 \quad (17)$$

These qualitative conclusions regarding the epimerization process are not altered by allowing the intermediates to exchange their waters of solvation with the surrounding medium and/or increasing the number of water molecules within the solvation sphere.

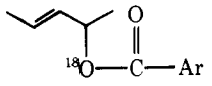
The retained asymmetry in the initially formed carbonium ion can be most readily explained via asymmetric solvation. Crystallographic data of E_2 prostaglandins indicate that the two chains are in close proximity to yield a hairpin-like structure.[19] The combination of these interacting chains and the nonplanar ring can yield an intermediate carbonium ion from either R or S with a hydrophobic side and a hydrophilic side which is formally asymmetric as shown in **4**. Examination of



4

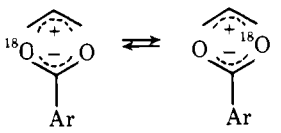
molecular models reveals that the proposed structure of the initial intermediate is not unreasonable.

Other studies have also indicated the presence of asymmetric carbonium ion intermediates in allylic systems. The nature of ion pairs in allylic systems has been extensively studied by Goering and his associates via determination of the extent of ^{18}O scrambling in the solvolysis of ^{18}O-labeled *p*-nitrobenzoate and *p*-toluenesulfonate esters. Of particular interest here are the results from the partial solvolysis of optically active esters such as **5**.[15-17] Comparison of the rates of ^{18}O exchange with



5

the rate of racemization in the recovered ester led to the conclusion that, in some cases, the intermediate ion pair in solvolysis is made asymmetric by the position of the anion rather than by the structure of the cation. For example, for **5**, the ion-pair intermediate was concluded to be represented by the interconverting species **6**.



6

Wiberg and Nakahira have examined the extent of racemization and deuterium scrambling during the solvolysis of the 3,5-nitrobenzoate esters of optically active cycloalkenols labeled with deuterium at C-1.[18] The apparent retention of configuration of the starting material on ionization in the epimerization of R and S is analogous to the results obtained from the larger cycloalkenols. With cyclooct-2-enyl 3,5-dinitrobenzoate, more than 90% of product was obtained with retention of configuration and with no deuterium scrambling. They interpreted this result as being consistent with minimal allyl participation in the solvolysis mechanism and with steric interactions restricting the attack at the backside of the carbon bearing the leaving group. An alternative way of expressing this latter conclusion is to state that the intermediate carbonium ion retains, to a degree, its original asymmetry.

In summary, we have used modern analytical techniques to reveal three competing processes in the change of configuration at the α carbon of a nonrearranging tertiary allylic alcohol: (1) substitution (^{18}O exchange) with epimeric equilibration, (2) substitution (^{18}O exchange) with retention, and (3) epimerization without substitution. Comparison of the relative rate constants for the first two processes has led us to propose a mechanistic scheme in which the initially formed intermediate carbonium ion is asymmetric by virtue of its solvation. Although it is quite possible that the asymmetric intermediates revealed by this study are unique to the prostaglandins, it may be that they are representative of the general case of tertiary alcohol racemization, undetected by methods used in previous kinetic studies. For the general case, the proposed mechanistic scheme predicts that asymmetric intermediates should be most apparent in those cases in which the carbonium ions are short lived.

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