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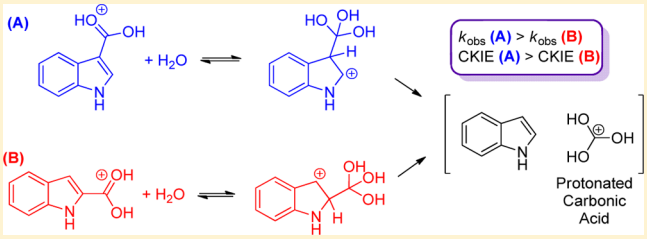
Carbon Kinetic Isotope Effects Reveal Variations in Reactivity of Intermediates in the Formation of Protonated Carbonic Acid

Adelle A. Vandersteen,[†] Scott O. C. Mundle,[‡] Georges Lacrampe-Couloume,[‡] Barbara Sherwood Lollar,[‡] and Ronald Kluger*,[†]

[†]Davenport Research Laboratories, Department of Chemistry, University of Toronto, 80 St. George Street, Toronto, Ontario, Canada M5S 3H6

[‡]Stable Isotope Laboratory, Department of Earth Sciences, University of Toronto, 22 Russell Street, Toronto, Ontario, Canada M5S 3B1

ABSTRACT: Kinetic evidence suggests that acid-catalyzed decarboxylation reactions of aromatic carboxylic acids can occur by a hydrolytic process that generates protonated carbonic acid (PCA) as the precursor of CO_2. Measurements of reaction rates and carbon kinetic isotope effects (CKIE) for decarboxylation of isomeric sets of heterocyclic carboxylic acids in acidic solutions reveal that C−C cleavage to form PCA is rate-determining with significant variation in the magnitude of the observed CKIE (1.018−1.043). Larger values are associated with the more reactive member in each isomeric pair. This variation is consistent with stepwise mechanisms in which C−C cleavage is competitive with C−O cleavage, leading to reversion to the protonated reactant to varying degrees with an invariant intrinsic CKIE for C−C cleavage. Thus, the relative barriers to reversion and formation of PCA control the magnitude of the observed CKIE in a predictable manner that correlates with reactivity. Application of the proposed overall mechanism reveals that carboxylation reactions in acidic solutions will proceed by way of initial formation of PCA.



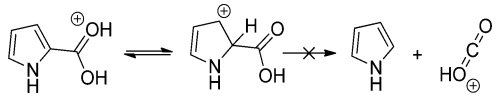
INTRODUCTION

The structure and properties of protonated carbonic acid ($C(OH)_3^+$, PCA) have been the subjects of spectroscopic and theoretical analysis.[1−5] We have recently reported that kinetic analysis of acid-catalyzed decarboxylation reactions of heterocyclic carboxylic acids implicates PCA as an obligatory reaction intermediate whose role is predicted to be similar in a wide range of reactions.[6−9] The overall process involves addition of the equivalent of a water molecule and a proton, in analogy to ester hydrolysis, hence the designation "hydrolytic decarboxylation".

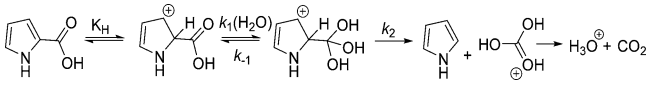
The rate law for specific-acid-catalyzed decarboxylation of a carboxylic acid requires that the rate-determining transition state arises from a species with the equivalent of one more proton than the neutral carboxylic acid. This is clearly inconsistent with C−C bond cleavage occurring via a simple dissociative process that produces CO_2. Furthermore, while the formation of the conjugate acid of CO_2 ($HOCO^+$) would be consistent with such a rate law, the exceedingly low proton affinity of CO_2 makes its conjugate acid an inaccessible reaction intermediate (Scheme 1).[10]

These results are instead consistent with an alternative pathway that produces PCA in the step that cleaves the C−C bond. This is achieved by the initial addition of water to the carboxyl group and protonation of the ring α to the carboxyl, leading to a highly reactive precursor of PCA (Scheme 2).[6−9]

Scheme 1. Prohibitive Decarboxylation Reaction of Pyrrole-2-carboxylic Acid Leading to Protonated Carbon Dioxide



Scheme 2. Acid-Catalyzed Decarboxylation of Pyrrole-2-carboxylic Acid via Addition of H^+ and H_2O



The overall process has been assessed and supported by independent computational studies that also tested various alternatives.[11,12]

In order to specify the role of PCA in the context of reaction intermediates, we evaluated the kinetic properties of two sets of isomeric heterocyclic carboxylic acids derived from pyrrole and indole. While they are expected to undergo hydration with similar energetics, the reactive intermediates that produce PCA are energetically distinct, leading to observable effects that define the nature of the transition states that produce PCA in competition with those that revert to the reactants.

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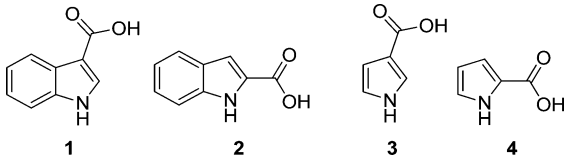
On the basis of the overall reaction patterns of these sets of reactants, we find that the transition state for the step that produces PCA is partially rate-determining to varying extents. This information is accessed via measurements that reveal the magnitudes of the observed carbon kinetic isotope effects (k^{12}/k^{13}, CKIE) for the reactions in the sets of related compounds under conditions where a defined single reaction mechanism is required by the observed rate law (plateau regions of the acidity-rate profiles). Comparing the results of observed rate constants and CKIE measurements in the acid-catalyzed decarboxylation reactions of compounds **1**−**4** reveals variations that affect the competing forward and reverse steps from the hydrated intermediates that are C-protonated (protonated at the position α to the hydrated carboxyl). The cations differ in energy depending on the extent to which stabilization by formation of iminium character is accessible. The range of observed CKIEs can be understood from considering a common intrinsic value for the isotope effect that is then attenuated by the extent to which the formation of PCA is competitive with reversion to the reactants from the steady-state reactive intermediate species.



RESULTS

As was reported for the acid-catalyzed decarboxylation reactions of pyrrole-2-carboxylic acid[6,7] and the indole-carboxylic acids,[9] the observed first-order rate constant for the decarboxylation of pyrrole-3-carboxylic acid increases with increasing acidity, reaching a plateau higher than that for the uncatalyzed reaction. The resulting dependence of the observed first-order rate constants on acidity (H_o) for all heterocyclic carboxylic acids fit the calculated titration curves for forming the conjugate acids of the reactants (Figure 1). In the plateau regions, there is necessarily a common mechanism for each species and for which the observed CKIEs were obtained.

While the equation-generated plots of acidity vs first-order rate constants are similar in shape for the various reactants, there are significant differences in the values of the high plateaus as well as for the lowest acidities at which the maximum values occur (presumably due to the pK_a of the conjugate acid). The fitted values for the maximum observed first-order rate constants are summarized in Table 1 along with the apparent macroscopic pK_a values that were used to fit the data to the Hammett acidity function, H_o. For the indole-carboxylic acids, the rate of decarboxylation is greater where the carboxyl group is positioned at C-3 of the indole ring, while carboxyl substitution at the C-2 position leads to a faster observed rate in the pyrrole-carboxylic acid series.

The variations of the observed maximum rate constants demonstrate a pattern that applies to the step leading to formation of PCA, with additional information provided by the differential magnitudes of the observed carbon kinetic isotope effects (CKIEs) (Table 2).

DISCUSSION

Previous studies with pyrrole-2-carboxylic acid[7] revealed a large CKIE for the acid-catalyzed decarboxylation which indicates

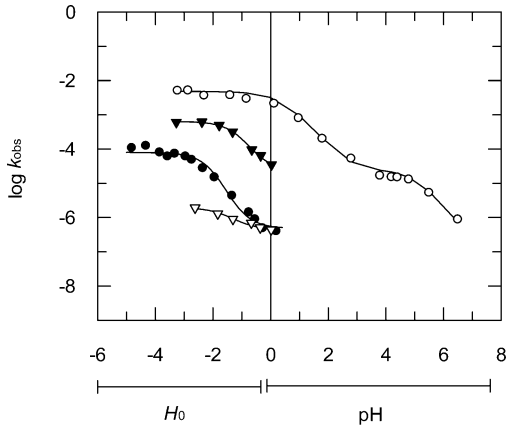


Figure 1. Logarithm of the first-order rate constants for the decarboxylation of (○) indole-3-carboxylic acid, (●) indole-2-carboxylic acid, (▼) pyrrole-2-carboxylic acid, (▽) pyrrole-3-carboxylic acid, as a function of pH and Hammett acidity constants. (▼/▽) $HClO_4$ solutions at 25 °C and (●/○) HCl solutions at 60 °C. For indole-3-carboxylic acid, data points higher than pH 1.0 are buffered solutions.[9]

Table 1. Maximum Observed First-Order Rate Constants for Decarboxylation

aromatic carboxylic acid	k_{obs}(max) (s^{-1})	pK_a[c]
pyrrole-2-carboxylic acid[a]	6.2×10^{-4}	−1.3
pyrrole-3-carboxylic acid	1.9×10^{-6}	−1.3
indole-2-carboxylic acid[b]	1.2×10^{-4}	−1.5
indole-3-carboxylic acid[b]	5.0×10^{-3}	0.4

[a]Reference 6. [b]Reference 9. [c]For monoprotonation of the carboxylic acids from titration using H_o-defined media.

Table 2. Carbon Kinetic Isotope Effects Observed for Acid-Catalyzed Decarboxylation Reactions

solution acidity (H_o or pH)	CKIE (±0.002)
Pyrrole-2-carboxylic Acid[a,b]	
H_o = −2.6	1.043
H_o = −0.7	1.027
Pyrrole-3-carboxylic Acid[b]	
H_o = −2.6	1.036
H_o = −0.4	1.028
Indole-2-carboxylic Acid[c]	
H_o = −3.8	1.018
H_o = −2.7	1.017
H_o = −1.3	1.002
Indole-3-carboxylic Acid[c]	
H_o = −3.2	1.030
H_o = 0.2	1.027
pH 4.4	1.003

[a]Reference 7. [b]Conditions: 25 °C, $HClO_4$. [c]Conditions: 60 °C, HCl.

that the rate-determining transition state involves cleavage of the C−C bond (forming PCA). However, the locations of the carboxyl group within a series of indole- and pyrrole-carboxylic acid positional isomers lead to differences in reactivity that affect the magnitudes of the observed CKIEs. It is likely that the step involving C−C cleavage is subject to a common intrinsic isotope effect, while the significant variations in the values of CKIEs arise from the extent to which the C−C cleavage step is rate-determining in competition with reversion to reactants. This is consistent with expectations from theoretical analysis of

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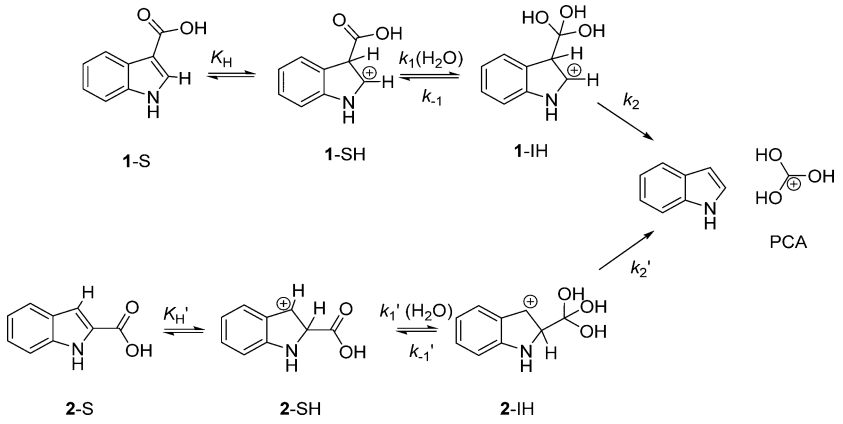
Scheme 3. Mechanisms for the Decarboxylation of Indole-3-carboxylic Acid (Upper) and Indole-2-carboxylic Acid (Lower)



carbon kinetic isotope effects from S_N2 and E2 reactions which reveal that, despite significant changes in the transition state (or bond order), intrinsic CKIEs are essentially invariant.[13−17]

An instructive example can be found in the heavy-atom isotope effects for decomposition of substituted benzenediazonium ions.[18] The electrons from the cleaved bond are transferred to nitrogen (forming N_2) rather than to the residual organic species as in decarboxylation reactions. While the electron transfer is in the opposite sense to that observed in the acid-catalyzed decarboxylation reactions, substituents also affect the bond-breaking event. This is apparent in the 100−1000-fold range in the observed rates for dediazoniation with various substituents.[18] This variation in reactivity might suggest that there are changes in the position of the transition state for the carbon−nitrogen cleavage step that lead to the variation in observed isotope effects; however, the measured nitrogen kinetic isotope effects are invariant for all derivatives.[19,20] Thus, variations in the observed kinetic isotope effects for breaking bonds to heavy atoms are not associated with the extent of bond breaking in the transition state for C−N bond cleavage.

An important study from Hilvert, O'Leary, and co-workers[21] determined the CKIEs for the decarboxylation of 5-nitro-3-carboxybenzisoxazole under a variety of conditions. In this one-step reaction coupled to decomposition, reversion to reactants is not possible. Therefore, the magnitude of the observed CKIE should be identical with the magnitude of the intrinsic isotope effect. The polarity of the reaction medium was varied (including the presence of a catalytic antibody), which produced a large variation in rate; the observed first-order rate constants vary by a factor as large as 2×10^4. However, a nearly constant magnitude for the CKIE was observed (~1.045). This value is a reasonable expectation for the intrinsic CKIE. In the present study there are significant changes in the magnitudes of CKIEs. This is clear evidence that variations in the CKIEs result from effects of variability in competition among partially rate determining steps in a multistep pathway. The invariance of the intrinsic CKIEs is the result of their arising only from differences in the ground state vibrational energies where the C−C bond is in place, whereas in the transition states that bond is broken and no new bond to either position is in the process of being formed (in contrast to the case for proton transfers between Brønsted acids and bases).

On the basis of the preceding discussion, we assume that the magnitudes of the observed CKIEs arise from a common intrinsic CKIE in the C−C bond-breaking steps. The observed CKIE for reaction of the conjugate acid of indole-3-carboxylic acid (**1**-SH; Scheme 3) is significantly larger than that for the conjugate acid of indole-2-carboxylic acid (**2**-SH) (1.030 vs 1.018). The extent to which the step competes with the reversion to the reactants controls the magnitude of the observed CKIE.

In Scheme 3 the magnitude of k_2/k_{-1} must be smaller than that for k_2'/k_{-1}' to give the observed differential CKIE values, as expressed in eqs 1−3. The magnitudes of the observed

$$k_{obs} = \frac{k_1 k_2}{k_{-1} + k_2} \tag{1}$$

$$CKIE_{obs} = \frac{k_2^{12}}{k_2^{13}} \cdot \frac{1 + \frac{k_2^{13}}{k_{-1}}}{1 + \frac{k_2^{12}}{k_{-1}}} \tag{3}$$

CKIEs (eq 3) depend on the relative values of k_2/k_{-1} for each reactant. The smaller the ratio, the larger the value of the observed CKIE, approaching the intrinsic CKIE for the unimolecular process as a limit. This follows the analysis from which Northrop identifies the nature of the effects of "commitment factors".[22,23]

$$CKIE_{obs} = \frac{k_{obs}^{12}}{k_{obs}^{13}} = \frac{\frac{k_2^{12}}{k_{-1} + k_2^{12}}}{\frac{k_2^{13}}{k_{-1} + k_2^{13}}} \tag{2}$$

In general, where the magnitudes of k_{-1} and k_2 are comparable, the CKIE that is observed depends on the ratio of the values of those rate constants. Where the rate constant for C−C cleavage is larger than that for the reversion process (loss of water and a proton), the observed CKIE will be smaller. For the less reactive substrate in each set (i.e., indole-2-carboxylic and pyrrole-3-carboxylic acids), protonation at a site that is more highly energetic than is the case for their paired isomers leads to a smaller observed rate constant within the pair of isomers. Therefore, reactions of the less reactive isomer occur from intermediates that are closer in energy to the transition state for formation of PCA than in the cases for the more reactive member. As a result, the larger observed CKIE is associated with the more reactive substrates, since the barrier

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giving the CKIE is higher relative to the reversion step than it is with the less reactive substrates.

The sources of these observations can be understood from mathematical models for these processes. The curves in Figure 2 were generated to illustrate the relationship between

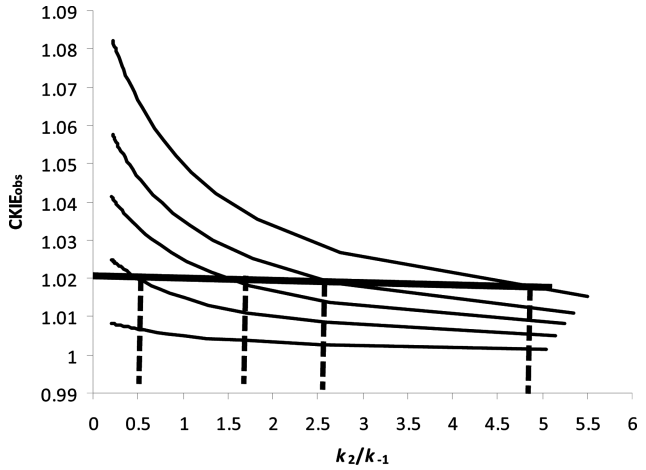
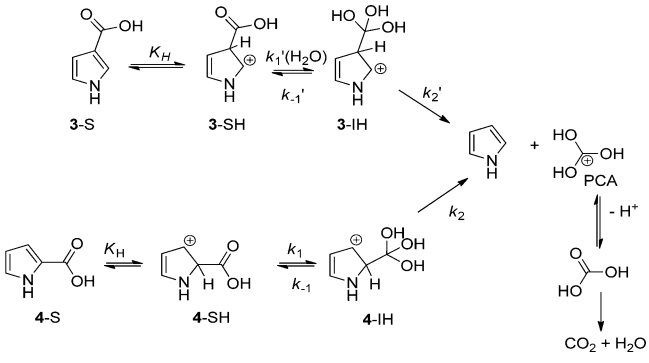


Figure 2. Theoretical model of observed CKIEs as a function of the ratio of the rate constants for C−C bond cleavage in comparison to reversal (k_2/k_{-1}) for a range of descending intrinsic CKIE values (1.10, 1.07, 1.05, 1.03, 1.01). Dashed lines indicate the relationship of commitment factor to observed CKIE as a function of a typical intrinsic CKIE for cleavage of a C−C bond.

commitment factors and observed CKIEs as derived from the rate law for the mechanism in Scheme 2. The *x* axis is the value of the rate constant subject to the intrinsic CKIE divided by the effective rate constant for conversion of the same intermediate to the protonated reactant. The *y* axis is the resulting observed CKIE. The value of the observed CKIE is reduced from the intrinsic CKIE (on the basis of a range of illustrative intrinsic values for a CKIE) as the rate constant for the C−C breaking step becomes larger relative to the rate constant for reversion (the latter increases and the former remains constant). As the barrier for reversion becomes larger relative to that for the C−C bond-breaking step, the value for the observed CKIE approaches a limiting value of 1.0.

Pathways for the decarboxylation of the isomeric pyrrole-carboxylic acids are presented in Scheme 4. These provide the necessary intermediates to understand the basis of the variation

Scheme 4. Mechanisms for the Decarboxylation of Pyrrole-3-carboxylic Acid (Upper) and Pyrrole-2-carboxylic Acid (Lower)

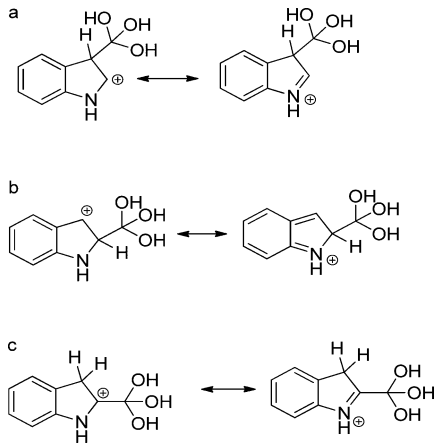


in CKIEs. The rapid decomposition of PCA leads to formation of CO_2, which is the detected product other than pyrrole, as the proton and water are catalytically cycled.

In all of the reactions that we have presented here, the species that precedes the cleavage of the C−C bond (to release PCA) contains a hydrated carboxyl group where protonation at the position α to the (hydrated) carboxyl carbon has taken place. These intermediates have a barrier to the transition state for decarboxylation lower than those in which the proton is added to the β position, although the latter in some cases are thermodynamically favored.[24−26]

The relative energies of the cationic intermediates depend on the effects of protonation on aromatic stabilization, as the positive charge is dispersed onto the nitrogen of the heterocycle, as shown in Scheme 5. In the case of the indole-

Scheme 5. Resonance Stabilization for α Protonation of (a) Indole-3-carboxylic Acid, (b) Indole-2-carboxylic Acid, and (c) β Protonation of Indole-2-carboxylic Acid



carboxylic acids, the pathway for the less reactive isomer (indole-2-carboxylic acid) proceeds through the structure shown in Scheme 5b. However, protonation to produce the structure in Scheme 5c gives a more stable intermediate.[25,26] In other words, protonation of the aromatic ring must occur at the less basic site in order for the system to be able to lose PCA in the next step. On the other hand, disruption of aromaticity is less significant for the pyrrole-carboxylic acid, as there is no benzenoid moiety as in the indole derivatives. As a result, this leads to a smaller distinction in the magnitudes observed for the CKIEs for the pyrrole-carboxylic acid isomers.

The difference between the pK_a values for protonation at the lower energy position vs that which is required for the reaction pathway of decarboxylation of pyrrole-3-carboxylic acid is about 2.0 pK_a units.[24] The observed rate constant for decarboxylation of pyrrole-3-carboxylic acid is about 300 times smaller than that for pyrrole-2-carboxylic acid (Table 1), suggesting that the rate differences arise principally from differences in the energies of the sites for protonation. In pyrrole-2-carboxylic acid, the required site of protonation for decarboxylation is also the lower energy site of protonation under the reaction conditions. The reaction patterns of the indole-carboxylic acids follow the same trend, leading to differences in the values for the rate maxima. However, in contrast to the pyrrole derivatives, it is the isomer with the carboxyl at the 3-position that is preferentially protonated and that also leads to C−C bond cleavage.

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In typical decarboxylation reactions in neutral solutions, formation of the residual carbanion is the key rate-controlling feature and the reaction falls within the realm of carbanion chemistry. We see from the present study that in acid-catalyzed decarboxylation reactions which produce PCA, the leaving group is derived from a cation that becomes neutral and aromatic upon passing through the transition state involving C−C cleavage. Since the other product, PCA, is a cation, we consider the step that produces it will be subject to factors that parallel the typical rate-determining step in substitution reactions that proceed by an S_N1 mechanism. Where the intermediate preceding PCA is higher in energy than in the case of its isomer, the leaving group is thereby activated, reducing the barrier to C−C cleavage. Although the barrier to that step is reduced, the overall reaction is slower than in the case where the pre-PCA intermediate is subject to greater stabilization. Thus, k_2 for the higher energy species will be greater (with a lower barrier) than k_2 for the lower energy species. On the other hand, in the competing process, loss of water to re-form the carboxyl group (k_{-1}) should be independent of the nature of the specific intermediate. As a result, k_2/k_{-1} is larger for the less reactive species, resulting in a lower value for the observed CKIE (see eq 3).

Our results show that the protonated carboxylic acid is more reactive toward decarboxylation than any other protonation state of the substrate. However, it does not directly produce CO_2 in the C−C cleavage step. This is not because PCA is formed more easily than CO_2; rather, it is due to the ring-protonated carbocation leaving group being formed to a much greater extent in acid, as compared to the zwitterion that forms in neutral solutions to produce CO_2.

Thus, we see the importance of PCA in defining the key intermediates in a major class of readily accessible decarboxylation reactions. Its formation occurs where an aromatic species acquires a proton to form a carbocation at the position β to a carboxyl group, even if the site of protonation is not the site that gives the intermediate that is lowest in energy. Rather, the key factor is that protonation must occur on the site that leads directly to the production of PCA. Significantly, the pattern of observed CKIEs provides the necessary context for arriving at this understanding and the results also provide insights into the factors leading to the observed value of a CKIE.

■ CONCLUSIONS

The mechanisms of acid-catalyzed decarboxylation reactions implicate the formation of PCA from a carbocationic intermediate that is generated by addition of water to the carboxyl group and a proton to the α position of the adjoining unsaturated species, regardless of the relative energy of protonation at that site. The variation in the observed CKIE is consistent with a common intrinsic value that depends on the extent to which hydration is also partially rate-determining. The key reactive intermediate is one that leads to the formation of PCA. The principle of microscopic reversibility suggests that electrophilic aromatic substitution based on PCA should be an accessible route to carboxylation of aromatic heterocycles.[27]

■ EXPERIMENTAL SECTION

Pyrrole-2-carboxylic acid, pyrrole-3-carboxylic acid, indole-2-carboxylic acid, and indole-3-carboxylic acid were obtained from commercial sources. All structures were verified spectroscopically, and the compounds were used without further purification. Acidic solutions were prepared from combinations of reagent-grade hydrochloric acid or perchloric acid with purified water.

Kinetics of Decarboxylation. The rates of decarboxylation for pyrrole-2-carboxylic acid, indole-2-carboxylic acid, and indole-3-carboxylic acid in acidic solutions have been previously reported.[6,7,9] The rate of decarboxylation of pyrrole-3-carboxylic acid was measured in solutions of perchloric acid of H_o-defined acidity. The reaction was followed by the decrease in absorbance at 255 nm with a UV−vis spectrometer at 25 °C, with the cell compartment kept within ±0.1 °C of the reported temperature. Data were collected with an interfaced computer, and the observed first-order rate constants were calculated by regression to the apparent first-order rate expression using the method of initial rates.

Measurement of Carbon Kinetic Isotope Effects. Reactions were carried out in 125 mL bottles sealed with butyl-blue stoppers. The acidic reaction solution (50 mL) was placed in the bottle, and the headspace was purged with helium to remove atmospheric CO_2. The carboxylic acid reactant (16 mg) was dissolved in degassed dimethyl sulfoxide (0.5 mL) and injected into the vessel to initiate the reaction. The reactions were maintained at 60.0 °C (indole-carboxylic acids) or 25.0 °C (pyrrole-carboxylic acids) in a circulating water bath. At specific reaction progress intervals, the bottle was cooled in ice and the reaction was quenched with 25 mL of degassed acetate buffer (1 M, pH 5) for reactions taking place in dilute acid solutions or by addition of 60 mL of degassed acetate buffer (5 M, pH 5) for reactions taking place in concentrated acid solutions in order to produce a dilute acid solution (~0.01 M) appropriate for headspace analysis. Solutions were kept at 0 °C prior to analysis. The headspace was sampled with a pressure-lock analytical syringe with a side-port taper needle.[7,28,29] Different reaction progress intervals were sampled from the headspace up to 50% conversion. Reaction progress was approximated by reaction time and by comparison with the peak area obtained from mass intensity scans on an isotope-ratio mass spectrometer coupled to a combustion oven and gas chromatograph (GC-C-IRMS). Samples of CO_2 from complete conversion of the reactants were taken after 10 half-lives for each reaction. As a control, the sequence was repeated without reactant. In these cases, CO_2 was not detected in the headspace.

The CKIEs were calculated from the measured data using an equation adapted from Bothner-By and Bigeleisen:[29,30]

$$k^{12}/k^{13} = \log(1 - f)/\log[1 - f(N_x/N_{x0})] \quad (4)$$

In eq 4, k^{12} and k^{13} are the observed first-order rate coefficients for reaction of the corresponding carbon isotopes and f denotes the fractional extent of the decarboxylation process, which varies from 0 at the start to 1 at completion. The originally defined terms "R" and "R_0" have been replaced with "N_x" and "N_{x0}" (where the ratio of abundance of $^{13}CO_2/^{12}CO_2$ from the IRMS has been converted to relative abundances).

■ AUTHOR INFORMATION

Corresponding Author

*E-mail for R.K.: rkluger@chem.utoronto.ca.

Notes

The authors declare no competing financial interest.

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■ ABBREVIATIONS

CKIE, carbon kinetic isotope effect; PCA, protonated carbonic acid

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