# KINETIC ISOTOPE EFFECTS IN THE DECARBOXYLATION OF SOME SUBSTITUTED BENZOIC ACIDS

by

John Buccini

A Thesis Submitted to the

Faculty of Graduate Studies and Research

of the University of Manitoba

In Partial Fulfillment of the Requirements

For the Degree of Master of Science

May 1967

DEDICATED

TO MY WIFE, CAROLE

ACKNOWLEDGEMENTS

The author would like to thank Dr. G. E. Dunn for his interest and for his numerous helpful suggestions concerning this work. Thanks are also due to Dr. A. N. Bourns and Mr. Jan Monster of McMaster University for the mass spectrometric analyses.

The financial support of the Shell Oil Co. Ltd. and the National Research Council is gratefully acknowledged.

ABSTRACT OF M.Sc. THESIS

submitted by John Buccini

Kinetic Isotope Effects in the Decarboxylation of Some Substituted Benzoic Acids.

(A) The carboxyl-$C^{13}$ kinetic isotope effects were measured for the decarboxylations of 4-methylsalicylic acid in solutions of quinoline and of .02 M quinoline in nitrobenzene. The observed isotope effects at 195°C were 0.7 $\pm$ .1 % and 2.2 $\pm$ .1 %, for the quinoline-nitrobenzene and pure quinoline solutions, respectively. These results indicate that the mechanism of this reaction involves the equilibrium formation of ion pairs, followed by the formation of a reaction intermediate which can decompose to either products, or an ion pair.

(B) The carboxyl-$C^{13}$ kinetic isotope effects were measured for the decarboxylation of 4-methoxyanthranilic acid in aqueous solutions of different pH and constant ionic strength (0.5). The isotope effects observed at 60°C were 4.2 $\pm$ .1%, 1.4 $\pm$ .1% and 0 for solutions of pH = -.3, 1.3, and 4.0, respectively. These results are interpreted in terms of a mechanism in which the acid (HA), the acid anion ($A^-$) and zwitterion (Z) are all protonated to form non-Bjerrum intermediates, ($H_2A^*$, $HA^*$ and $HZ^*$, respectively) of which only $HA^*$ can decarboxylate.

TABLE OF CONTENTS

TABLE OF CONTENTS (cont'd)

# KINETIC ISOTOPE EFFECTS IN THE DECARBOXYLATION OF SOME SUBSTITUTED BENZOIC ACIDS

## INTRODUCTION

In order to predict the effects of isotopic substitution on reaction rates, one must first have a theory of chemical reactions. The most successful theory to date has been the theory of absolute reaction rates as put forward by Eyring (13), Polanyi (12) and others (15). This theory assumes that a chemical reaction can be represented by a three dimensional surface, the shape of which is determined by the reaction coordinate, and the potential energy of the reacting system. Such a surface is called a potential energy surface. It is assumed that there is but one potential energy surface along which a reaction takes place. On this surface, one part of coordinate space represents the reactants, and another, the products. Separating these two regions there is an energy barrier. In order for a chemical reaction to occur, the reactants must approach one another and pass over the energy barrier. The molecular species corresponding to the top of the energy barrier, for the energetically easiest path from reactants to products, is called the "activated complex". It is proposed that there is always a concentration of activated complex in equilibrium with the reactants, and that the rate of reaction is determined by the rate of decomposition of the complex.

When a mechanism is proposed for a particular reaction, we are, at present, usually satisfied if we have a general idea of the structure of the activated complex. One way in which information may be gained about the activated complex is from the studies of isotopic substitution. As most reactions involve only a small part of a given reactant molecule, we may learn the fate of a particular atom in this part of the molecule by conducting "tracer" studies; i.e. we can substitute an isotope b* for a given atom b in the reactive part of the molecule, and then locate the isotope b* in the reaction products. Such experiments

will lead to the conclusion that certain bonds are broken, made or unaffected. These results will enable us to postulate one, or more, possible activated complexes. These models can then be experimentally tested to see if they are correct.

One way of testing a possible complex involves isotopic substitution within the reaction center, and the measurement of the relative rates of reaction of the two isotopic isomers. If the two rates are different, there is said to be a kinetic isotope effect.

Often, there is more than one possible activated complex. However, these different complexes frequently give rise to different predictions as to the existence of a kinetic isotope effect. Thus the determination of the kinetic isotope effect can be used as a powerful diagnostic tool.

In some cases, a measurable kinetic isotope effect can be observed when the labelled atom is not at the actual reaction site. This is referred to as a secondary isotope effect. The following discussion will be limited to primary kinetic isotope effects, i.e. those cases in which the labelled atom is at the actual reaction site.

## THE THEORETICAL PREDICTION OF ISOTOPE EFFECTS

When an atom b of a molecule is replaced by its isotope b*, a number of relatively small effects come into play, all of which must be taken into consideration in order to understand the over-all effect. Using the theory of absolute reaction rates, Bigeleisen (3) has arrived at an expression for predicting the kinetic isotope effect in the following type of reaction.

$$A_1 + B + C + \ldots \xrightarrow{k_1} (\text{products})_1 \qquad \ldots.(1)$$

$$A_2 + B + C + \ldots \xrightarrow{k_2} (\text{products})_2 \qquad \ldots.(2)$$

$A_1$ and $A_2$ are isotopic isomers.

$k_1$ and $k_2$ are the specific rate constants for the reactions expressed by equations (1) and (2), respectively.

B, C, etc. may or may not be present; if they are present, they may be other A molecules, or different species.

The subscripts (1) and (2) usually refer to the light and heavy isotopes, respectively.

For reactions involving the isotopes of carbon and heavier elements, Bigeleisen (3) proposes the following equation for the theoretical calculation of the kinetic isotope effect.

$$(s_2/s_1)(s_1^{\ddagger}/s_2^{\ddagger})(k_1/k_2) =$$

$$(K_1/K_2)(\nu_{L1}^{\ddagger}/\nu_{L2}^{\ddagger})\left[1 + \sum_i^{3n-6} G(u_{i2})\Delta u_i - \sum_i^{3n^{\ddagger}-7} G(u_{i2}^{\ddagger})\Delta u_i^{\ddagger}\right](1 + \Delta|u_L^{\ddagger}|^2/24) \quad ..(3)$$

The various symbols and terms in equation (3) will now be defined and discussed.

In any term, the superscript (‡) refers to a property of the activated complex, and the subscripts (1) and (2) refer to the light and heavy isotopes, respectively.

The ratio $k_1/k_2$ gives the relative rate of reaction of the two isotopic isomers, $A_1$ and $A_2$. This ratio is thus a measure of the kinetic

isotope effect in a particular reaction.

The classical symmetry number (s) is the number of indistinguishable positions that a molecule (or activated complex) can take in space. In effect, the symmetry numbers convert the calculated ratio of rates of reaction to the ratio of rates per equivalent position in the reactant molecule.

For example, if ABA and ABA* are isotopic isomers which decompose according to the following reactions,

$$ABA \xrightarrow{k} A + BA$$

$$ABA^* \xrightarrow{k'} A + BA^*$$

then, in calculating $k/k'$ we must account for the fact that even if there is no kinetic isotope effect due to the substitution of A* for A, the value of $k'$ will be one half that of k. This occurs because the probability of the rupture of the A-B bond in ABA is twice that in ABA*. Thus, for the above reactions, assuming an activated complex of the form A---B-A, we will have $s_1 = 2$, $s_2 = 1$, $s_1^{\ddagger} = 1$, and $s_2^{\ddagger} = 1$. Therefore the left hand side of equation (3) would become $k/2k'$.

The transmission coefficient (K) is the fraction of the activated complexes which decomposes in the direction corresponding to the completed reaction. Thus, the transmission coefficient accounts for the activated complexes which decompose to reactants rather than to products. In order to be able to calculate K, the potential energy surface for a reaction must be known, and even then a rigorous calculation is not necessarily possible. Because the potential energy surface is not generally known, K cannot be evaluated. However, some theoretical calculations (20) have shown that in cases of experimental interest above room temperature, the ratio of the transmission coefficients can be taken as unity. It should be noted that this assumption is one of the weakest

points in the theory. It would be most desirable to be able to calculate the values of the two transmission coefficients.

The frequency $\nu_L^{\ddagger}$ corresponds to the stretching vibration along the coordinate of decomposition for the bond in the activated complex which is about to be broken. This frequency is an imaginary number. It has been shown (6) that the ratio $\nu_{L1}^{\ddagger}/\nu_{L2}^{\ddagger}$ can be replaced by $(m_2^*/m_1^*)^{\frac{1}{2}}$, where m* is the effective (or reduced) mass of the activated complex along the coordinate of decomposition. In the earlier literature, m* was assumed to be the reduced mass of the two atoms which form the bond about to be broken. Now, however, m* is usually taken to be the reduced mass of the two molecular fragments created by the cleavage of the bond(+). If M' and M'' are the two separating molecular fragments, then

$$\frac{\nu_{L1}^{\ddagger}}{\nu_{L2}^{\ddagger}} = \left(\frac{m_2^*}{m_1^*}\right)^{\frac{1}{2}} = \left(\frac{1/M_1' + 1/M_1''}{1/M_2' + 1/M_2''}\right)^{\frac{1}{2}} \qquad \ldots.(4)$$

Thus, the term $\nu_{L1}^{\ddagger}/\nu_{L2}^{\ddagger}$ is dependent only on the reduced masses of the sets of molecular fragments formed by the two isotopic isomers. For this reason, this term is called the reduced mass factor, or the temperature independent factor.

In the last term of equation (3),

$$\Delta|u_L^{\ddagger}|^2 = |u_{L1}^{\ddagger}|^2 - |u_{L2}^{\ddagger}|^2$$

and

$$u_L^{\ddagger} = h\nu_L^{\ddagger}/kT$$

where h is Planck's constant, k is the Boltzmann constant and T is the absolute temperature of the reaction.

---

(+) In fact, this value is correct "only if the centers of mass of the two fragments lie on the extensions of the line corresponding to the broken bond". Otherwise the mass fragment value is only an approximation (40).

The last term of equation (3) gives the correction for the effect of quantum mechanical "tunneling" on the rate of reaction. Quantum mechanically there is a possibility that reactants can pass into products without passing over the usual energy barrier. This is accomplished by "tunneling", or leakage through the energy barrier. The net effect is a slight increase in the rate of reaction. A calculation of the tunnel effect would require a thorough knowledge of the potential energy surface for the reaction. As this is generally unknown, the effect of tunneling on the rate of a reaction cannot be calculated. However, from general experience it is found that the tunnel correction is negligible for the isotopes of carbon and the heavier elements. It is therefore assumed that the correction is negligible in the consideration of isotope effects.

Thus, omitting the symmetry numbers, and neglecting the ratio of the transmission coefficients and the tunnel correction, equation (3) reduces to equation (5).

$$(k_1/k_2) = (\mathcal{V}^{\ddagger}_{L1}/\mathcal{V}^{\ddagger}_{L2})\left[1 + \sum_{i}^{3n-6} G(u_{i2})_{\Delta}u_i - \sum_{i}^{3n^{\ddagger}-7} G(u^{\ddagger}_{i2})_{\Delta}u^{\ddagger}_i\right] \qquad \ldots(5)$$

The reduced mass (or temperature independent) term and the other remaining term (which will be referred to as the zero-point energy term) are the two factors which will determine whether or not there will be a kinetic isotope effect. While both terms are necessary for a quantitative theoretical prediction, the zero-point energy term alone can be employed in a very useful qualitative discussion of the prediction of kinetic isotope effects.

In the theory of the electronic structure of the hydrogen atom, the nuclear mass (M) enters the calculations only in the form of the reduced mass of the system ($\mu$). If m is the mass of the electron, then $1/\mu = 1/m + 1/M$. As the minimum value of M is about 1850m, a change in

M from unity to infinity will not noticeably affect the value of $\mu$. Thus, a change in the nuclear mass of the hydrogen atom will have a negligable effect on the electronic structure of the atom. Extending this idea to the heavier elements, the electronic structures of two isotopes should be the same to a very high degree of approximation.

Consider now the replacement of an atom in a complex molecule by one of its isotopes. The interatomic forces will not be affected by the change in nuclear mass, as these forces are determined by the electronic structure. Thus, isotopic isomers should have the same interatomic distances, vibrational force constants and also the same potential energy surface for any reaction, to a very high degree of approximation.

When the translational motion of an atom is considered, it is the total mass, m + M, and therefore M which is of importance. The translational energy of an atom is then governed by the nuclear mass. Isotopic substitution should, therefore, affect the vibrational frequencies of the bonds to the labelled atom, as these vibrations involve acceleration of the nuclear mass. In fact, it is found that the substitution of the heavier isotope reduces the frequency of vibration associated with a particular bond to the labelled atom.

In the zero-point energy term, n is the number of atoms in a particular species, and

$$G(u_i) = 1/2 - 1/u_i + 1/(e^{u_i} - 1)$$

also, $$\Delta u_i = u_{i1} - u_{i2}$$

where $$u_i = h\nu_i/kT$$

and $\nu_i$ is the vibrational frequency associated with the $i^{\underline{th}}$ normal mode of vibration. In the reactant, there are 3n-6 normal modes of vibration,

while there are only $3n^{\ddagger}-7$ normal modes in the activated complex. The missing mode of vibration in the activated complex corresponds to $\nu_L^{\ddagger}$, and has already been taken into consideration in the reduced mass factor.

When all the oscillators in a molecule are in their lowest energy states, the total vibrational energy of the molecule is not zero, but a minimum value ($V_o$), called the zero-point energy. If the minimum in the potential energy curve for the normal molecule is taken as the energy origin, then it is easily shown (25) that

$$V_o = \sum_i^{3n-6} \tfrac{1}{2}h\nu_i$$

As previously stated, the substitution of a heavier isotope in a molecule will result in a decrease in the vibrational frequencies of the bonds to the labelled atom. Assuming that the vibrational frequencies of the bonds to other atoms are not appreciably affected, one would expect the molecule with the heavier isotope to have a lower zero-point energy than the one with the light isotope, i.e. we would expect

$$\sum_i^{3n-6} \tfrac{1}{2}h\nu_{i1} > \sum_i^{3n-6} \tfrac{1}{2}h\nu_{i2}$$

or,
$$V_{o1} > V_{o2}$$

If the top of the energy barrier is taken as the energy origin for the activated complex, a similar argument shows that the zero-point energy ($V_o^{\ddagger}$) is given by

$$V_o^{\ddagger} = \sum_i^{3n^{\ddagger}-7} \tfrac{1}{2}h\nu_i^{\ddagger}$$

It follows, therefore, that

$$\sum_i^{3n^{\ddagger}-7} \tfrac{1}{2}h\nu_{i1}^{\ddagger} > \sum_i^{3n^{\ddagger}-7} \tfrac{1}{2}h\nu_{i2}^{\ddagger}$$

or,
$$V_{o1}^{\ddagger} > V_{o2}^{\ddagger}$$

This situation is shown graphically in Figure 1.

Let $\quad \Delta V = V_{o1} - V_{o2}$

and $\quad \Delta V^{\ddagger} = V^{\ddagger}_{o1} - V^{\ddagger}_{o2}$

The summation $\sum_{i}^{3n-6} G(u_{i2})\Delta u_i$ is related to $\Delta V$, the difference between the zero-point energies of the isotopic reactants. The summation $\sum_{i}^{3n^{\ddagger}-7} G(u^{\ddagger}_{i2})\Delta u^{\ddagger}_i$ is related to $\Delta V^{\ddagger}$, the difference between the zero-point energies of the isotopic activated complexes. From equation (5), it is seen that the ratio $k_1/k_2$ is proportional to

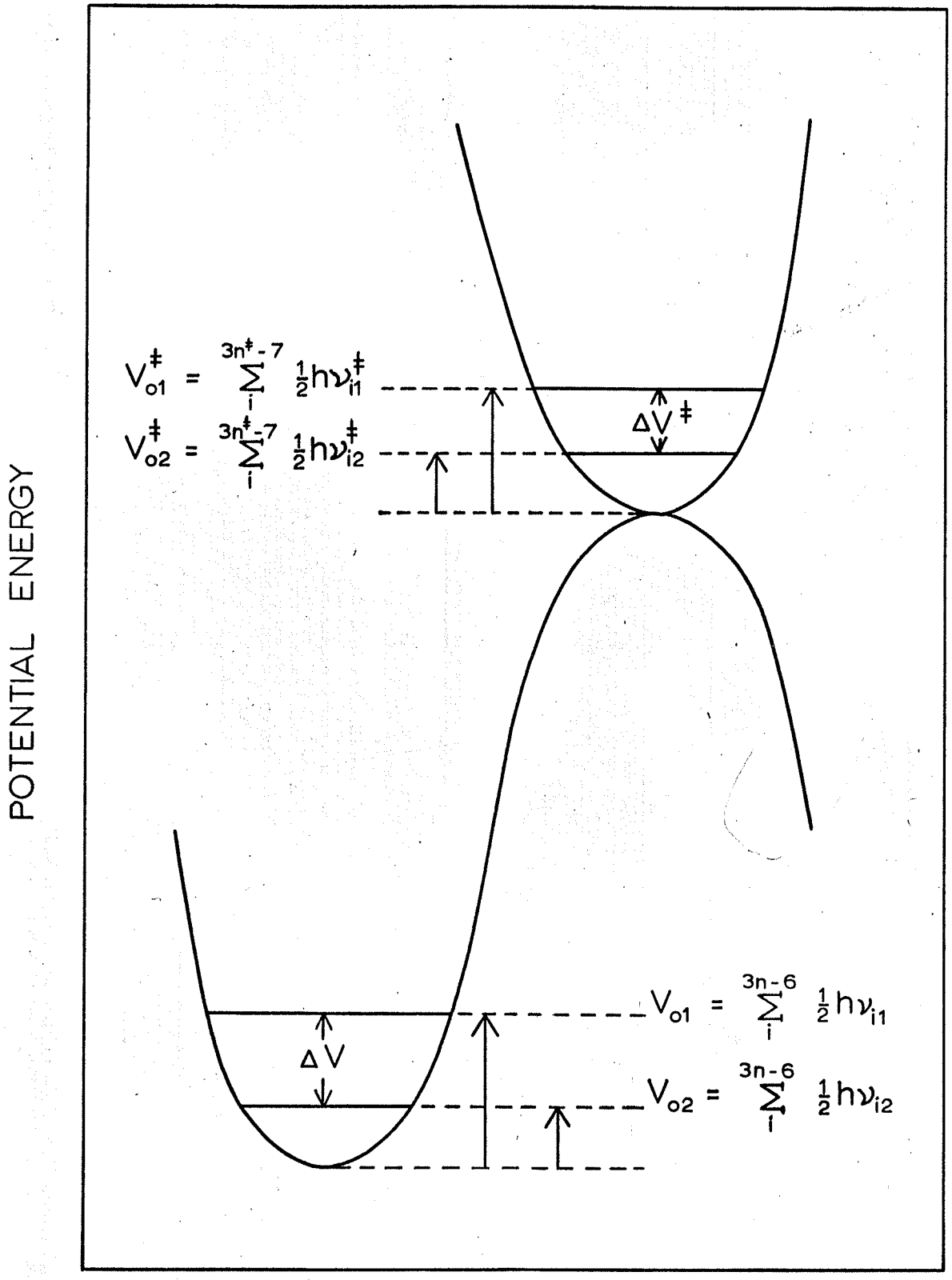
$$\sum_{i}^{3n-6} G(u_{i2})\Delta u_i - \sum_{i}^{3n^{\ddagger}-7} G(u^{\ddagger}_{i2})\Delta u^{\ddagger}_i$$

Thus the kinetic isotope effect will be proportional to the quantity $(\Delta V - \Delta V^{\ddagger})$. It is interesting to note that the kinetic isotope effect is not dependent upon the activation energy of the reaction (i.e. the difference in energy level between the reactants and the activated complexes).

The problem of predicting a kinetic isotope effect has now been reduced to one of evaluating the vibrational frequencies of the normal modes of oscillation of the reactants and the activated complexes. While these frequencies in the reactants can be evaluated from infra red spectra, generally they have been found only for very simple molecules. In the last few years the use of computers has enabled some theoreticians to obtain the vibrational frequencies of some rather complex molecules, and some exact calculations of $k_1/k_2$ for various models of the activated complex have been carried out (17). In the case of activated complexes, the vibrational frequencies would have to be estimated on the basis of known frequencies for the stable compounds and would, of course, depend

FIGURE 1: The potential energy profile showing the relation between the zero-point energies of the isotopic reactants and of the isotopic activated complexes.

FIGURE 1


POTENTIAL ENERGY
$V^{\ddagger}_{o1} = \sum_{i}^{3n^{\ddagger}-7} \frac{1}{2} h\nu^{\ddagger}_{i1}$
$V^{\ddagger}_{o2} = \sum_{i}^{3n^{\ddagger}-7} \frac{1}{2} h\nu^{\ddagger}_{i2}$
$\Delta V^{\ddagger}$
$\Delta V$
$V_{o1} = \sum_{i}^{3n-6} \frac{1}{2} h\nu_{i1}$
$V_{o2} = \sum_{i}^{3n-6} \frac{1}{2} h\nu_{i2}$
REACTION COORDINATE

upon the model chosen for the complex. However, in more complex molecules, the reaction center comprises only a small portion of the molecule, and, if all the vibrational frequencies cannot be evaluated, it is usually assumed that the vibrational frequencies of the bonds removed from the reaction site are unchanged in going from the initial state to the activated state, i.e. $\nu_j = \nu_j^\ddagger$ for those bonds removed from the reaction site. This results in the cancellation of such pairs of terms as $G(u_{j2})\Delta u_j$ and $G(u_{j2}^\ddagger)\Delta u_j^\ddagger$. If, however, within the reaction center there is one bond to the labelled atom that is considerably weakened, or completely broken in the activated complex, the pair of terms corresponding to this vibrational mode will not vanish. For the limiting case in which the bond is completely broken in the activated complex, $\nu_i^\ddagger = \nu_L^\ddagger$ , and the term for the activated state vanishes. Thus, assuming all other frequencies unchanged, the maximum zero-point energy contribution to the kinetic isotope effect would be $1 + G(u_{i2})\Delta u_i$ .

If the bond to the labelled atom is only weakened, then the contribution to the kinetic isotope effect is less by the amount $G(u_{i2}^\ddagger)\Delta u_i^\ddagger$ . The zero-point energy contribution would be, therefore,

$$1 + G(u_{i2})\Delta u_i - G(u_{i2}^\ddagger)\Delta u_i^\ddagger$$

As $\Delta u$ is always positive, and $G(u)$ is also always positive and increases with u (2), if the bond to the labelled atom is weakened or broken in the activated state, $k_1/k_2 > 1$, and the rate of reaction will be greater for the lighter isotope.

If a bond to the labelled atom had actually strengthened in the activated complex, then $G(u_{i2})\Delta u_i < G(u_{i2}^\ddagger)\Delta u_i^\ddagger$ and $k_1/k_2 < 1$. This is referred to as a reverse isotope effect. It is much less common than the previous case.

In general, then, if a bond to a labelled atom is appreciably changed (weakened or strengthened) in the activated complex, an isotope effect will occur.

At this point it should be noted that the zero-point energy contribution is the most important term for those cases involving the isotopes of hydrogen. For the isotopes of carbon and heavier elements, the reduced mass factor must also be taken into consideration.

Temperature Dependence of the Isotope Effect

The magnitude of $k_1/k_2$ will be determined by two factors, i.e. the reduced mass factor and the zero-point energy factor. As the former is temperature independent its contribution will be the same at all temperatures. Therefore any effect of temperature on the value of $k_1/k_2$ can be attributed to the zero-point energy term, unless the transmission coefficients and/or the tunnel correction come into play.

The zero-point energy term involves the energy difference $\Delta V - \Delta V^{\ddagger}$. As the temperature is decreased, this energy difference will become more and more important, and in the limit as the temperature approaches the absolute zero, the ratio $k_1/k_2$ will approach infinity for the usual case in which $k_1/k_2 > 1$. For the less common case of the reverse isotope effect, $k_1/k_2$ will approach zero.

We have defined $\Delta u_i$ as follows;

$$\Delta u_i = u_{i1} - u_{i2} = h(\nu_{i1} - \nu_{i2})/kT$$

In the high temperature limit, as the temperature approaches infinity, the $\Delta u$ terms in the zero-point energy term approach zero. Therefore, both $G(u)\Delta u$ terms vanish and the zero-point energy term reduces to unity. Thus the high temperature limit is

$$k_1/k_2 = \nu^{\ddagger}_{L1}/\nu^{\ddagger}_{L2}$$

for both the common and reverse isotope effects.

The temperature variation of $k_1/k_2$ is usually quite noticeable over temperature ranges of about 60°C. Yankwich and Belford (46) have studied the temperature dependence of the intermolecular kinetic isotope effect in the decarboxylation of malonic acid in quinoline solutions. They used acid labelled with carbon-13 in the -COOH group, and compared the rates of decarboxylation of malonic-1-$C^{12}$ and malonic-1-$C^{13}$ acids. The value of $k_{12}/k_{13}$ varied from 1.0567 at 34°C to 1.0379 at 118°C.

## EXPERIMENTAL EVALUATION OF THE KINETIC ISOTOPE EFFECT (+)

The magnitude of the ratio $k/k^*$ may be experimentally evaluated by two methods.

I - The Method of Absolute Rates

In this method, the rate of reaction for each of the isotopic species is measured directly, and the ratio $k/k^*$ is calculated. Except for a few special cases, the accuracy of the measurements is not better than a few per cent. However, for isotopes of the elements heavier than hydrogen, the ratio of rate constants differs from unity by only a few per cent. Thus an error of a few per cent in each of k and k* would mean that a small isotope effect might not be detected. For this reason, this method is limited to the isotopes of hydrogen, where k is usually greater than k* by several orders of magnitude.

II - The Method of Competitive Rates

There are two types of competitive methods which may be used; the chemical competitive method and the isotopic competitive method. For several reasons (6) the former method is limited to the isotopes of hydrogen, and will not be discussed here.

In the isotopic competitive method, the isotopic reactants, A and A*, are present in a mixture and either undergo unimolecular decomposition, or react with other chemical species, B, C, etc., where these latter species may be other A molecules. This method is especially simple when applied to systems in which A*is present to the extent of 1 % or less. As the natural abundances of $C^{13}$, $N^{15}$ and $O^{18}$ are of the order of 1 % or less, this method can conveniently

(+) In the previous discussion, $k_1$ and $k_2$ represented the specific rate constants for the reactions of the species containing the light and heavy isotopes, respectively. In the following discussion, as a matter of convenience, $k_1$ and $k_2$ are replaced by k and k*, respectively.

be used in those cases requiring carbon, nitrogen or oxygen labelling in A*, as in these cases natural samples of a reactant can be used to achieve the mixture of isotopic reactants. The necessity for isotopic enrichment of the reactant mixture is thus avoided. Also, systems employing A* at tracer concentrations (much less than 1 %) can be treated by this method. This method is not, however, limited to those cases for which $A^*/A \leq .01$ .

Consider the case in which the rate of reaction is of first order with respect to the concentration of A or A*, and of some arbitrary order b with respect to the concentration of B, c with respect to the concentration of C, etc. . The reactions can be written as follows, where X and X* are the isotopic products.

$$A + B + C + .... \xrightarrow{k} X + Y + .... \quad ....(6)$$

$$A^* + B + C + .... \xrightarrow{k^*} X^* + Y + .... \quad ....(7)$$

If $k \neq k^*$, the difference in the reaction rates will result in a continual change in the isotopic content of both the reactant and the product. The value of $k/k^*$ can be evaluated by measuring the fractional amounts of conversion of A and A* and the isotopic content of either the remaining reactant or the product.

(1) Analysis of the isotopic content of the remaining reactant after a known amount of reaction

If $A_o$, $A_o^*$, $B_o$, $C_o$, etc. represent the initial amounts of the reactants, and A, A*, B, C, etc. represent the amounts of the reactants remaining after a time t, and extent of reaction f, then the rates of reactions (6) and (7) at time t can be expressed as follows.

$$- dA/dt = k(A)(B)^b(C)^c\text{-----}$$

$$- dA^*/dt = k^*(A^*)(B)^b(C)^c\text{-----}$$

It follows from these two equations that.....

$$\frac{dA}{dA^*} = \frac{k(A)}{k^*(A^*)}$$

and therefore $\quad d\ \ln(A) = (k/k^*)\ d\ \ln(A^*)$

Integrating this equation between the limits $A_o$ and A for the left hand side, and $A_o^*$ and $A^*$ for the right hand side, we get, after converting to logarithms to base 10,

$$\frac{k}{k^*} = \frac{\log(A/A_o)}{\log(A^*/A_o^*)} \qquad \ldots.(8)$$

The measured fractional conversion, f, is given by

$$(1 - f) = \frac{(A + A^*)}{(A_o + A_o^*)}$$

By setting $R_o = A_o^*/A_o$ (i.e. the ratio of labelled to unlabelled molecules in the initial reactant) and $R_{af} = A^*/A$ (i.e. the ratio of labelled to unlabelled molecules in the reactant remaining after an amount of reaction, f) expressions (9) and (10) can be derived for $A/A_o$ and $A^*/A_o^*$, respectively.

$$(1 - f) = \frac{A(1 + A^*/A)}{A_o(1 + A_o^*/A_o)} = \frac{A(1 + R_{af})}{A_o(1 + R_o)}$$

$$\frac{A}{A_o} = \frac{(1 - f)(1 + R_o)}{(1 + R_{af})} \qquad \ldots.(9)$$

$$(1 - f) = \frac{A^*(1 + A/A^*)}{A_o^*(1 + A_o/A_o^*)} = \frac{A^*(1 + 1/R_{af})}{A_o^*(1 + 1/R_o)}$$

$$\frac{A^*}{A_o^*} = \frac{(1 - f)(1 + R_o)R_{af}}{(1 + R_{af})R_o} \qquad \ldots.(10)$$

Substituting equations (9) and (10) into equation (8) gives

$$\frac{k}{k^*} = \frac{\log\left[(1 - f)(1 + R_o)/(1 + R_{af})\right]}{\log\left[(1 - f)(1 + R_o)R_{af}/(1 + R_{af})R_o\right]} \qquad \ldots.(11)$$

If we let $\Theta = (k/k^* - 1)$, then the deviation of $\Theta$ from zero will be a measure of the kinetic isotope effect. A positive value

of $\Theta$ will correspond to the normal isotope effect ($k > k^*$); a negative value of $\Theta$, to the reverse isotope effect. For elements heavier than hydrogen, and showing a normal isotope effect, the value of $\Theta$ is usually between zero and + 0.1 .

Thus, in this method, the problem of evaluating the magnitude of the kinetic isotope effect has been reduced to one of measuring the extent of reaction, f, and the two ratios, $R_o$ and $R_{af}$ . The values of the isotopic ratios can be determined with a precision of 0.1 % or better by mass spectrometric analysis. If radioactive isotope is used, a precision of about 0.2 % can be obtained by measurements of radioactivity. The error in the value of f is usually about 0.5 % . If a precision of a few per cent in $\Theta$ is required, then it can be shown (6) that the extent of reaction must be greater than 0.5 . For $f > 0.5$ , the precision in $\Theta$ increases with increasing extent of reaction, until $f > 0.9$ , at which point the error in the determination of f increases, and the precision in $\Theta$ again decreases.

(2) Analysis of the isotopic content of the product after a known amount of reaction

If X and X* are the amounts of isotopic products produced up to time t and extent of reaction f, then, in the previous notation, $A = A_o - X$ and $A^* = A_o^* - X^*$ . At any time t, the rates of reactions (6) and (7) can now be written as follows.

$$dX/dt = k(A_o - X)(B)^b(C)^c\text{-----}$$

$$dX^*/dt = k^*(A_o^* - X^*)(B)^b(C)^c\text{-----}$$

It follows from these two equations that

$$\frac{dX}{dX^*} = \frac{k(A_o - X)}{k^*(A_o^* - X^*)}$$

and therefore $d \ln(A_o - X) = (k/k^*)\, d \ln(A_o^* - X^*)$

Integrating this equation between the limits zero and X for

the left hand side, and zero and X* for the right hand side, we get, after converting to logarithms to base 10,

$$\frac{k}{k^*} = \frac{\log(1 - X/A_o)}{\log(1 - X^*/A_o^*)} \qquad \ldots.(12)$$

The measured fractional conversion, f, is given by

$$f = \frac{(X + X^*)}{(A_o + A_o^*)}$$

By setting $R_o = A_o^*/A_o$ (as before) and $R_{xf} = X^*/X$ (i.e. the ratio of labelled to unlabelled molecules in the product collected after a measured extent of reaction, f) expressions (13) and (14) can be derived for $X/A_o$ and $X^*/A_o^*$, respectively.

$$f = \frac{X(1 + X^*/X)}{A_o(1 + A_o^*/A_o)} = \frac{X(1 + R_{xf})}{A_o(1 + R_o)}$$

$$\frac{X}{A_o} = \frac{f(1 + R_o)}{(1 + R_{xf})} \qquad \ldots.(13)$$

$$f = \frac{X^*(1 + X/X^*)}{A_o^*(1 + A_o/A_o^*)} = \frac{X^*(1 + 1/R_{xf})}{A_o^*(1 + 1/R_o)}$$

$$\frac{X^*}{A_o^*} = \frac{f(1 + R_o)R_{xf}}{(1 + R_{xf})R_o} \qquad \ldots.(14)$$

Substituting equations (13) and (14) into equation (12) gives

$$\frac{k}{k^*} = \frac{\log\left[1 - f(1 + R_o)/(1 + R_{xf})\right]}{\log\left[1 - f(1 + R_o)R_{xf}/(1 + R_{xf})R_o\right]} \qquad \ldots.(15)$$

Thus in this method, the problem of evaluating the magnitude of the kinetic isotope effect has been reduced to one of measuring the extent of reaction, f, and the two ratios, $R_o$ and $R_{xf}$ .

Bigeleisen and Allen (5) have reported a study of the errors invelved in evaluating the quantity $(k^*/k - 1)$ by this method. They calculated the per cent error in this quantity as a function of the extent of reaction, assuming

(i) $(k^*/k - 1) = -.1$ ,

(ii) a tracer concentration of $A_o^*$,

(iii) an error of $10^{-3}$ in the value of $R_{xf}/R_o$ , and

(iv) an accuracy of 0.5 % in the measured extent of reaction.

The results are shown graphically in Figure 2. Inspection of this graph shows that for this method, it is advantageous to attempt to evaluate $\Theta$ at small conversions. The difference between the values of $R_{xf}$ and $R_o$ is a maximum at the start of the reaction and decreases with the extent of reaction until $R_{xf} = R_o$ at $f = 1$. Therefore, the present method is preferred over that in which $\Theta$ is determined by isotopic analysis of the remaining reactant, because the present method permits an accurate determination of $\Theta$ in the region of the maximum value of $R_{xf}/R_o$. The previous method requires f to be at least 0.5 . If the difference between $R_{xf}$ and $R_o$ is small to begin with, $R_{xf}/R_o$ may not be sufficiently different from one at this conversion to permit a detection of the isotope effect.

The Isotopic Competitive Method at a Tracer Concentration of $A_o^*$

If A* is present in very small concentrations ($A_o^*/A_o \leq .01$), or if the fractionation factor is sufficiently small that

$$1 + R_o \approx 1 + R_{af}$$

and

$$1 + R_o \approx 1 + R_{xf}$$

then equations (11) and (15) reduce to equations (16) and (17), respectively.

$$\frac{k}{k^*} = \frac{\log(1 - f)}{\log[(1 - f)R_{af}/R_o]} \qquad \ldots.(16)$$

$$\frac{k}{k^*} = \frac{\log(1 - f)}{\log(1 - fR_{xf}/R_o)} \qquad \ldots.(17)$$

It has also been shown (8, 10) that if A* is present in tracer concentrations, then equations (16) and (17) are valid for reactions in which the rate is proportional to any power of the concentration of A.

FIGURE 2: The per cent error in (k*/k - 1) when determined by isotopic analysis of the product, as a function of the extent of reaction.

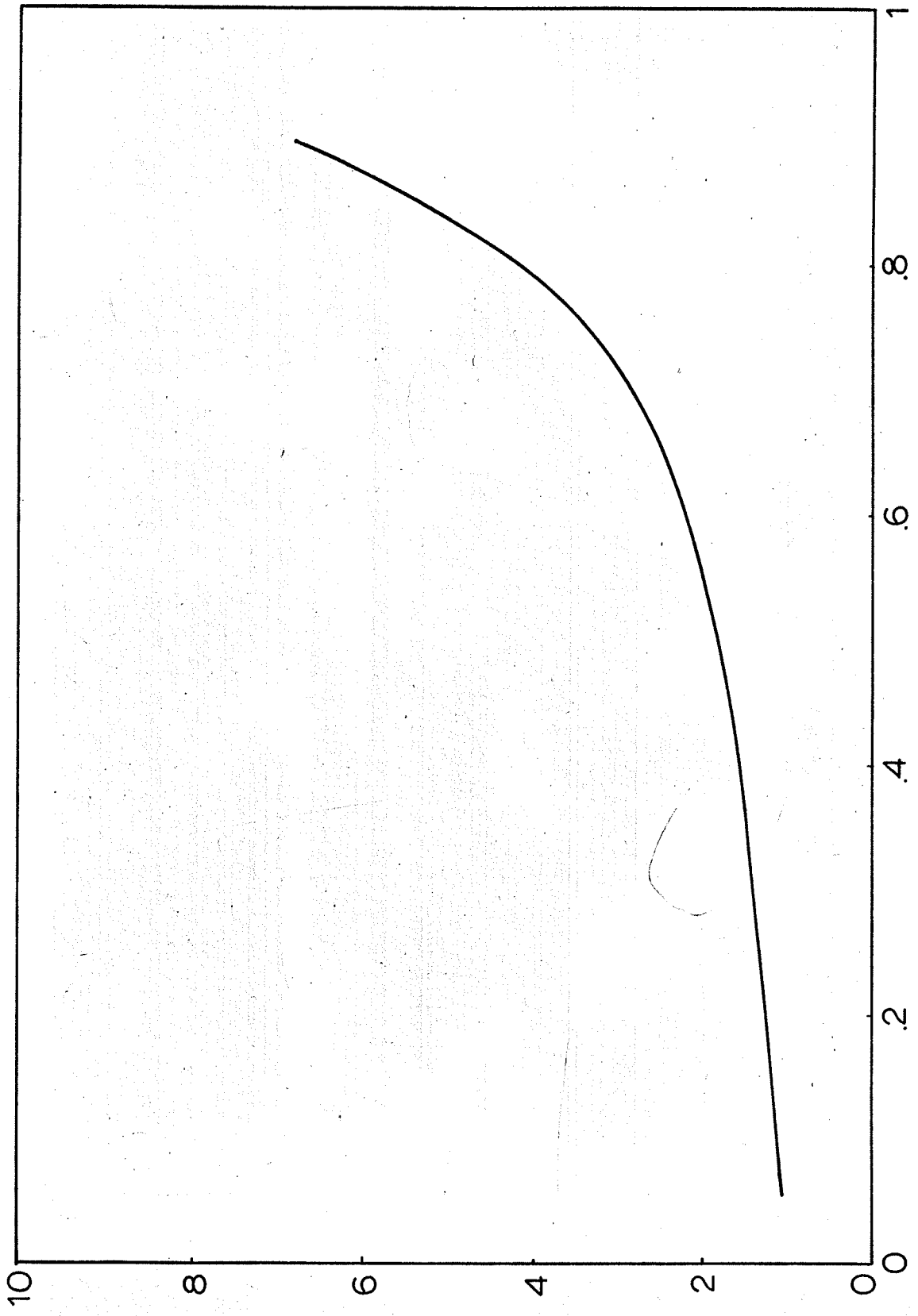



FIGURE 2

Intermolecular and Intramolecular Isotope Effects

So far, this discussion has been limited to the comparison of the rates of bond breaking of the same bond in two molecules which are identical except for the substitution of a labelled atom for its isotope in one molecule. This is referred to as the intermolecular isotope effect. In some cases, however, one could also consider the comparison of the rates of bond breaking of two bonds in the same molecule which are identical except for the substitution of a labelled atom at the end of one bond. This is referred to as the intramolecular isotope effect. It is a special case of the isotopic competitive method and will not be discussed further.

## HISTORICAL REVIEW

In 1932, Washburn and Urey (37) reported the first experimental demonstration of a kinetic isotope effect. They observed that in the electrolysis of water, the remaining liquid becomes enriched in $D_2O$. At the time, it was thought that only those elements lighter than carbon could show an effect large enough to be observed experimentally. It was not until 1948 that a kinetic isotope effect was observed for the isotopes of carbon (or any heavier element). In that year, Beeck et al (1) reported a study on the effect of electron impact on propane-1-$C^{13}$ in a mass spectrometer. These workers observed that the dissociation probability of the C-C bonds in the labelled molecule is decreased by $12 \pm 1$ % for the $C^{13}$-$C^{12}$ bond and increased by $7 \pm .2$ % for the $C^{12}$-$C^{12}$ bond, below and above the $C^{12}$-$C^{12}$ bonds in unlabelled propane.

Since this discovery, many reactions have been investigated to determine whether or not a kinetic isotope effect could be observed for the isotopes of carbon and the heavier elements. Among those reactions investigated for carbon isotope effects are several decarboxylation reactions, which may be represented as

$$R\text{-}COOH \longrightarrow R\text{-}H + CO_2 \ .$$

These reactions are particularly easy to study for two reasons.

(1) Carbon labelling in the -COOH group can be readily achieved. Both $C^{13}$ and $C^{14}$ labelling have been used: the former is achieved by using natural samples of the acid (which contain about 1 % $C^{13}$ in the carboxyl group) and the latter, by an appropriate synthesis of the acid from a $C^{14}$ labelled reactant.

(2) The ratio $R_o = (R\text{-}C^*OOH)_o/(R\text{-}COOH)_o$ can be readily obtained by mass spectrometric analysis or radioassay of the $CO_2$ produced by the complete decarboxylation of the acid. Then the ratio $k/k^*$ can be accurately evaluated by the isotopic analysis of the product ($CO_2$)

produced after a known extent of reaction.

The first, and by far the most extensively studied of the decarboxylation reactions, is that of malonic acid. In the notation of Bigeleisen and Friedman (4), the reactions are written as follows.

$$\begin{aligned}
&\text{HOOC-CH}_2\text{-COOH} \xrightarrow{k_1} \text{HOOC-CH}_3 + \text{CO}_2 \\
&\text{HOOC-C*H}_2\text{-COOH} \xrightarrow{k_2} \text{HOOC-C*H}_3 + \text{CO}_2 \\
&\text{HOOC-CH}_2\text{-C*OOH} \xrightarrow{k_3} \text{HOOC-CH}_3 + \text{C*O}_2 \\
&\qquad\qquad\qquad\quad \xrightarrow{k_4} \text{HOOC*-CH}_3 + \text{CO}_2
\end{aligned}$$

The ratio $k_1/2k_3$ is a measure of the intermolecular isotope effect: the ratio $k_4/k_3$ , the intramolecular isotope effect. Since the first investigation by Yankwich and Calvin (42) on molten malonic-1-$C^{14}$ acid, there has followed a series of experiments on the measurement of the intermolecular and/or intramolecular isotope effects, using $C^{13}$ and/or $C^{14}$ as the label, for malonic acid in the melt, in concentrated sulfuric acid solution, in quinoline solution and in dioxane solution. The decarboxylations of α-napthyl-, phenyl- and bromomalonic acids, as well as of the anion of malonic acid have also been reported. The observed effects are summarized in Tables I, II, and III.

These reactions are always first order in malonic acid, and the observance of isotope effects of from 2-6 % for $C^{13}$, and 5.5-13.2 % for $C^{14}$, indicate that the bond between the carboxyl carbon and the methylene carbon is considerably weakened, or broken in the activated complex. Thus carbon-carbon bond-breaking is involved in the slow step of this reaction.

The kinetics of the decarboxylation of mesitoic acid (2,4,6-trimethylbenzoic acid) in solutions of 82-100 % sulfuric acid have been investigated by Schubert (32) over the temperature range 50-90°C. On the basis of his investigation, Schubert proposed the following

TABLE I

A Summary of the Carbon Kinetic Isotope Effects Observed in the Decarboxylations of Malonic-1-C* Acids in the Melt

| RATIO | ISOTOPE | TEMP.(°C) | 100(k/k* - 1) | REF. |
|---|---|---|---|---|
| $k_1/2k_3$ | 13 | 137.5 | 3.7 ± .2 | ( 4) |
| | 13 | 137 | 3.4 | (23) |
| | 14 | 154 | 6.4 | (31) |
| $k_4/k_3$ | 13 | 137.5 | 2.0 ± .1 | ( 4) |
| | 13 | 138 | 2.6 ± .4 | (22) |
| | 13 | 138 | 2.1 ± .2 | (22) |
| | 13 | 138 | 2.6 ± .1 | (43) |
| | 13 | 137 | 2.8 ± .2 | (44) |
| | 13 | 140 | 2.92 ± .07 | (48) |
| | 13 | 140 | 2.85 ± .09 | (49) |
| | 14 | 153 | 6 ± 2 | (30) |
| | 14 | 140 | 5.45 ± .46 | (49) |
| | 14 | 140 | 5.77 ± .24 | (18) |

TABLE II

A Summary of the Intramolecular Carbon Kinetic Isotope Effects

Observed in the Decarboxylations of Some Substituted Malonic-1-C* Acids

| ACID | CONDITIONS | ISOTOPE | TEMP.(°C) | 100(k/k* - 1) | REF. |
|---|---|---|---|---|---|
| ∝-Naphthylmalonic | melt | 14 | 163 | 7.6 ± .5 | (14) |
| | dioxane-1 N HCl | 14 | 87.5 | 9.5 ± .8 | (14) |
| | dioxane-1 N HCl | 14 | 72.8 | 9.8 ± 1.2 | (14) |
| Phenylmalonic | melt | 14 | 163 | 8.8 ± 1.8 | (14) |
| | dioxane-1 N HCl | 14 | 72.8 | 13.2 ± 1.7 | (14) |
| Bromomalonic | melt | 13 | 117 | 2.4 ± .3 | (44) |
| | melt | 14 | 117 | 11.6 ± .4 | (44) |
| | melt | 14 | 118 | 6.46 ± .24 | (18) |

TABLE III

A Summary of the Inter- and Intramolecular Carbon Kinetic Isotope Effects Observed in the Decarboxylations of Malonic Acid and Its Monoanion in Various Solutions

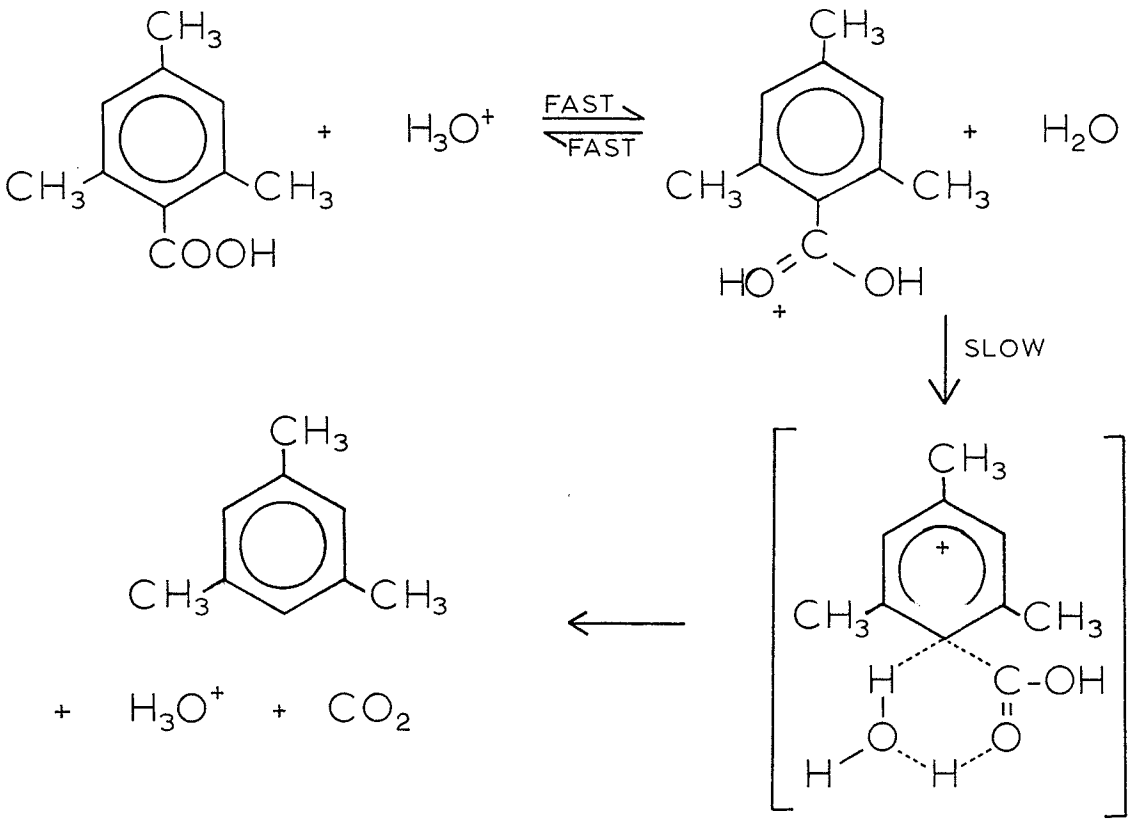
| SOLUTION | RATIO | ISOTOPE | TEMP.(°C) | 100(k/k* - 1) | REF. |
|---|---|---|---|---|---|
| Malonic acid in 80% $H_2SO_4$ | $k_1/2k_3$ | 13 | 56 | 4.53 | (45) |
| | | | 79 | 3.76 | |
| | | | 100 | 3.48 | |
| | | | 129 | 3.36 | |
| Malonic acid in quinoline | $k_1/2k_3$ | 13 | 34 | 5.67 | (46) |
| | | | 59 | 4.94 | |
| | | | 79 | 4.38 | |
| | | | 99 | 4.09 | |
| | | | 118 | 3.79 | |
| | $k_4/k_3$ | 13 | 86 | 4.45 | (47) |
| | | | 100 | 4.10 | |
| | | | 110 | 3.73 | |
| | | | 123 | 3.56 | |
| | | | 138 | 3.17 | |
| | $k_4/k_3$ | 14 | 85 | 8.5 | (18) |
| | | | 95 | 8.1 | |
| | | | 105 | 7.5 | |
| | | | 115 | 7.0 | |
| | | | 125 | 6.5 | |
| | | | 135 | 6.0 | |

TABLE III (cont'd)

| SOLUTION | RATIO | ISOTOPE | TEMP.(°C) | 100(k/k* - 1) | REF. |
|---|---|---|---|---|---|
| Malonic acid in dioxane | $k_1/2k_3$ | 13 | 40.3 | 4.33 | (52) |
| | | | 50.3 | 4.15 | |
| | | | 60.2 | 3.98 | |
| | | | 71.3 | 3.82 | |
| | | | 80.4 | 3.58 | |
| | | | 88.8 | 3.28 | |
| | | | 99.1 | 3.20 | |
| | $k_4/k_3$ | 13 | 89.5 | 3.59 | (53) |
| | | | 99.5 | 3.23 | |
| | | | 109.6 | 3.16 | |
| | | | 119.9 | 2.80 | |
| | | | 128.6 | 2.62 | |
| | | | 139.5 | 2.21 | |
| Monoanion in quinoline | $k_1/2k_3$ | 13 | 67.5 | 4.17 | (50) |
| | | | 79 | 4.15 | |
| | | | 98 | 3.91 | |
| | | | 119 | 3.77 | |
| | $k_4/k_3$ | 13 | 79 | 3.54 | (51) |
| | | | 89.5 | 3.68 | |
| | | | 102.5 | 3.52 | |
| | | | 115.5 | 3.32 | |
| | | | 138 | 3.28 | |

Note: The errors in the values of 100(k/k* - 1) for $C^{13}$ and $C^{14}$ were $\pm$ 0.1 and $\pm$ 0.2 , respectively.

mechanism for the reaction.




Bothner-By and Bigeleisen (8) measured the carboxyl-$C^{13}$ kinetic isotope effect for the decarboxylation of natural mesitoic acid in 86 % sulfuric acid solution, at 61.2°C and at 92.0°C. Stevens et al (33) have simultaneously measured the $C^{13}$ and $C^{14}$ isotope effects under the same conditions, using a sample of mesitoic acid with 0.8 % $C^{14}$ in the carboxyl group. Below is a summary of the results of both sets of workers.

| TEMP.(°C) | ISOTOPE | 100(k/k* - 1) | REF. |
|---|---|---|---|
| 60 ± .5 | $C^{13}$ | 3.8 ± .1 | (33) |
| 61.2 ± .1 | $C^{13}$ | 3.7 ± .3 | ( 8) |
| 92.0 ± .1 | $C^{13}$ | 3.2 ± .1 | ( 8) |
| 60 ± .5 | $C^{14}$ | 10.1 ± .5 | (33) |

These results indicate that carboxyl-carbon bond-breaking occurs in the slow step of the decarboxylation, in agreement with Schubert's proposed mechanism.

Kinetic investigations by Verhoek have indicated that the decarboxylations of sodium trichloroacetate in water (35) and sodium 2,4,6-trinitrobenzoate in ethanol (36) both proceed by the same mechanism - the unimolecular decomposition of the acid anion with carbon-carbon bond-breaking occurring in the slow step.

$$R\text{-}COO^- \longrightarrow R^- + CO_2 \qquad \text{(slow)}$$

This mechanism predicts that a carboxyl-carbon kinetic isotope effect should be observed in both reactions. Using natural samples of the reactants, carboxyl-$C^{13}$ isotope effects of 3.38 $\pm$ .07 % and 3.26 $\pm$ .08 % have been observed in the decarboxylations of sodium trichloroacetate in water at 70.4°C (5) and sodium trinitrobenzoate in 90 % ethanol at 50°C (28). Thus, the mechanism of Verhoek seems acceptable.

Stevens et al (34) have investigated the kinetics of the thermal, aqueous, and acid-catalysed decarboxylations of anthranilic acid. These workers suggested that the reaction occurs by a mechanism involving proton attack on the $\alpha$-carbon (i.e. the ring carbon to which the carboxyl group is attached). The absence of a carboxyl-$C^{13}$ kinetic isotope effect (34) led to the conclusion that the rate determining step was the formation of the bond between the proton and the $\alpha$-carbon. This step would then be followed by rapid carbon-carbon bond-breaking and no isotope effect would be observed.

Zlotowski and Zielinski (55) have measured the carboxyl-$C^{14}$ kinetic isotope effect in the decarboxylation of picolinic acid in the melt, and in solution in quinoline, phenol, hydroquinone, o-cresol, and o-nitrophenol at various temperatures (155-200°C). The results are summarized on page 30.

Assuming that the reaction in the melt proceeds by way of a species containing the $-COO^-$ group (such as the zwitterion form of the acid) theoretical calculations of the isotope effect were made for several

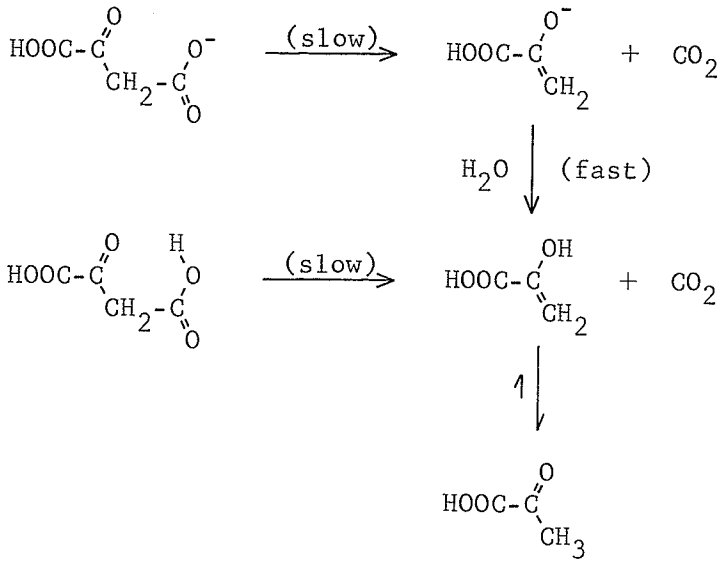
| MEDIUM | 100(k/k* - 1) at 156°C | at 185°C |
|---|---|---|
| Melt | 4.7 | 4.6 |
| Quinoline | 4.9 | 4.7 |
| Phenol | 5.8 | 5.4 |
| o-Cresol | 5.8 | 5.4 |
| Hydroquinone | - | 5.6 |
| o-Nitrophenol | - | 5.0 |

possible models of the activated complex. The best agreement between theory and experiment was found for a model of the activated complex in which the carbon-carbon bond is considerably weakened (but not broken) and the carbon-oxygen bonds of the carboxylate group are essentially unchanged.

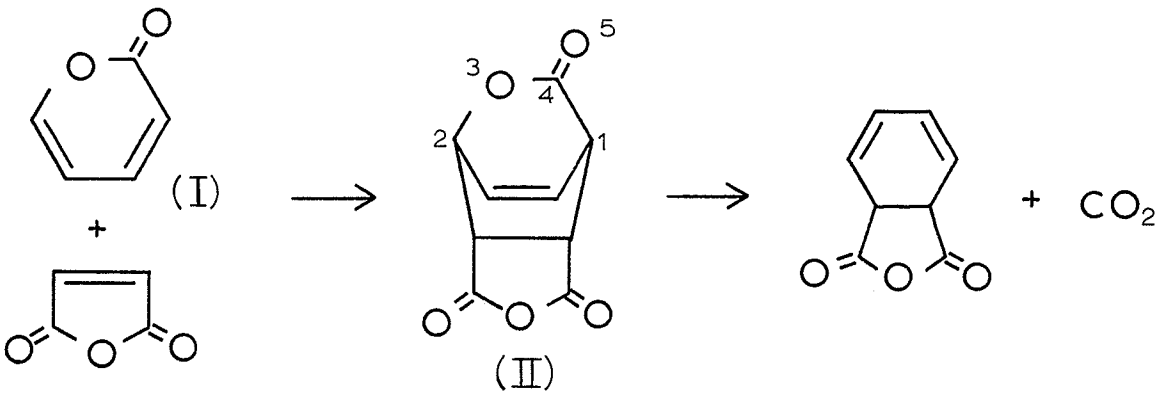
As quinoline shows no measurable influence on the isotope effect, it seems that the intramolecular interaction between the carboxyl hydrogen atom and the pyridine nitrogen is much stronger than the intermolecular interaction between the carboxyl hydrogen and the quinoline nitrogen. The measurable effects of the phenolic solvents (except o-nitrophenol, where the hydroxyl hydrogen forms a bridge to one of the oxygen atoms of the nitro group) indicates a relatively strong interaction between the phenolic -OH group and the picolinic acid molecule, leading probably to the formation of a hydrogen bridge linking the two molecules together.

Zielinski (54) has measured the carboxyl-$C^{13}$ and -$O^{18}$ isotope effects in the decarboxylation of quinaldic acid in the melt over a range of temperatures (141-181°C). The observed carbon isotope effects were in the range 1.04-1.13 ($\pm$ .06) %, and the oxygen isotope effects were in the range .03-.23 ($\pm$ .04) %. The interpretation of these results has been postponed pending vibrational analysis of the quinaldic acid molecule.

Wood (41) has observed a $C^{13}$ kinetic isotope effect of 4.48 $\pm$ .4 % in the decarboxylation of oxaloacetic acid in aqueous solutions at 25°C. He points out that this effect is in agreement with the mechanism which had been previously proposed for this reaction by Pedersen (27) - i.e. in the decomposition of β-keto carboxylic acids, the keto-form of the acid (or its anion) decomposes spontaneously into $CO_2$ and an enol (or enolate anion), which by a consecutive reaction is transformed into the stable ketone. The mechanism for the decomposition of oxaloacetic acid is then




Goldstein and Thayer have investigated the mechanism of the decarboxylation of the maleic anhydride adduct (II) of α-pyrone (I). First order kinetics were observed for the reaction in dimethyl phthalate solution (16).



These workers have also measured (17) the $C^{13}$ isotope effect for the carbon atom in position 4, and the $O^{18}$ isotope effect for the oxygen atom in position 3. Using natural samples of the reactants in dimethyl phthalate solution at 130.2°C, effects of 3.03 $\pm$ .25 % for carbon and 1.39 $\pm$ .19 % for oxygen were observed. An attempt was then made to determine whether the 1-4 and 2-3 bonds were broken simultaneously, or in a stepwise manner. Both approximate and exact theoretical calculations were carried out for various models of the activated complex. It was concluded that the observed isotope effects require that the carbon-carbon (1-4) bond be effectively broken in the activated state, whereas the carbon-oxygen (2-3) bond remains essentially intact, and the carbonyl carbon-oxygen (4-5) bond is somewhat strengthened.

Lynn and Bourns (24) have reported a study on the mechanism of the decarboxylation of 2,4-dihydroxybenzoic acid in aqueous solutions. The kinetic evidence indicates a bimolecular mechanism (24) involving electrophilic attack of a proton on the $\alpha$-carbon. Willi has established (38, 39) that proton transfer occurs in a rate-determining step and he has proposed (39) the following mechanism for the reaction.

$$\text{Ar-COO}^- + \text{HA} \xrightarrow{\text{(slow)}} \text{H-Ar}^+\text{-COO}^- + \text{A}^-$$

$$\text{H-Ar}^+\text{-COO}^- \xrightarrow{\text{(fast)}} \text{H-Ar} + \text{CO}_2$$

However, a concerted mechanism (in which carbon-carbon bond-breaking and carbon-hydrogen bond-making occur simultaneously) cannot be completely excluded.

Using natural samples of the acid, Lynn and Bourns (24) observed a carboxyl-$C^{13}$ isotope effect of 0.60 $\pm$ .05 % at 85°C for the reaction in aqueous perchloric acid solutions of varying concentrations (.002-.01 M). The magnitude of this isotope effect is about five to seven times smaller than those previously observed in reactions in which the carbon-carbon bond-breaking occurs in the rate-determining step. While this is in

agreement with Willi's mechanism, it could also be argued to be in favor of the concerted mechanism for the case in which the C-C bond has been only slightly altered after the reactant has passed into the activated state.

Lynn and Bourns (24) solved the problem by measuring the carboxyl-$C^{13}$ isotope effect for the decarboxylation in acetic acid - sodium acetate buffer solutions of different concentrations. In the pH range used (4.50-5.32) the acid is essentially ionized and the two possible reaction mechanisms can be written as follows.

(a) The concerted mechanism

$$Ar\text{-}COO^- + HA \xrightarrow{k} \left[H\text{-}Ar^+\text{-}COO^-\right] \longrightarrow H\text{-}Ar + CO_2 + A^-$$

(b) The stepwise mechanism

$$Ar\text{-}COO^- + HA \underset{k_2}{\overset{k_1}{\rightleftharpoons}} H\text{-}Ar^+\text{-}COO^- + A^-$$

$$H\text{-}Ar^+\text{-}COO^- \xrightarrow{k_3} H\text{-}Ar + CO_2$$

If the stepwise mechanism were the correct one, then an increase in the concentration of $A^-$ would increase the rate of decomposition of the intermediate to reactants. In the limiting case, a large amount of $A^-$ would cause the decomposition of intermediate to products to become at least partially rate-determining. As carbon-carbon bond-breaking is involved in this step, we would expect to find a carboxyl-$C^{13}$ isotope effect which increases with an increase in the concentration of $A^-$.

If the concerted mechanism were the correct one, then the concentration of $A^-$ should have no effect on the magnitude of the isotope effect.

The results of Lynn and Bourns (24) are summarized on page 34.

| pH | $CH_3COOH$ | $CH_3COONa$ | $100(k/k^* - 1)$ |
|---|---|---|---|
| 4.50 | .10 | .067 | $0.5 \pm .1$ |
| 5.15 | .05 | .15 | $1.1 \pm .1$ |
| 5.32 | .33 | 1.00 | $1.8 \pm .1$ |

Thus, it is experimentally observed that the isotope effect increases with the concentration of $A^-$, and the stepwise mechanism is indicated.

This "variable isotope effect" test for the existence of a reaction intermediate has also been used to establish the existence of an intermediate in the bromodecarboxylation of 3,5-dibromo-4-hydroxybenzoic acid (19) and in other organic reactions (56).

## PURPOSES OF THE PRESENT INVESTIGATION

### (A) The Decarboxylation of 4-Methylsalicylic Acid in Quinoline

Rodewald (29) has measured the rates of decarboxylation of a series of 4- and 5- substituted salicylic acids in quinoline solution over a temperature range of 60°C. His observations and conclusions can be summarized as follows.

(1) The reaction in quinoline is first order with respect to the concentration of the acid.

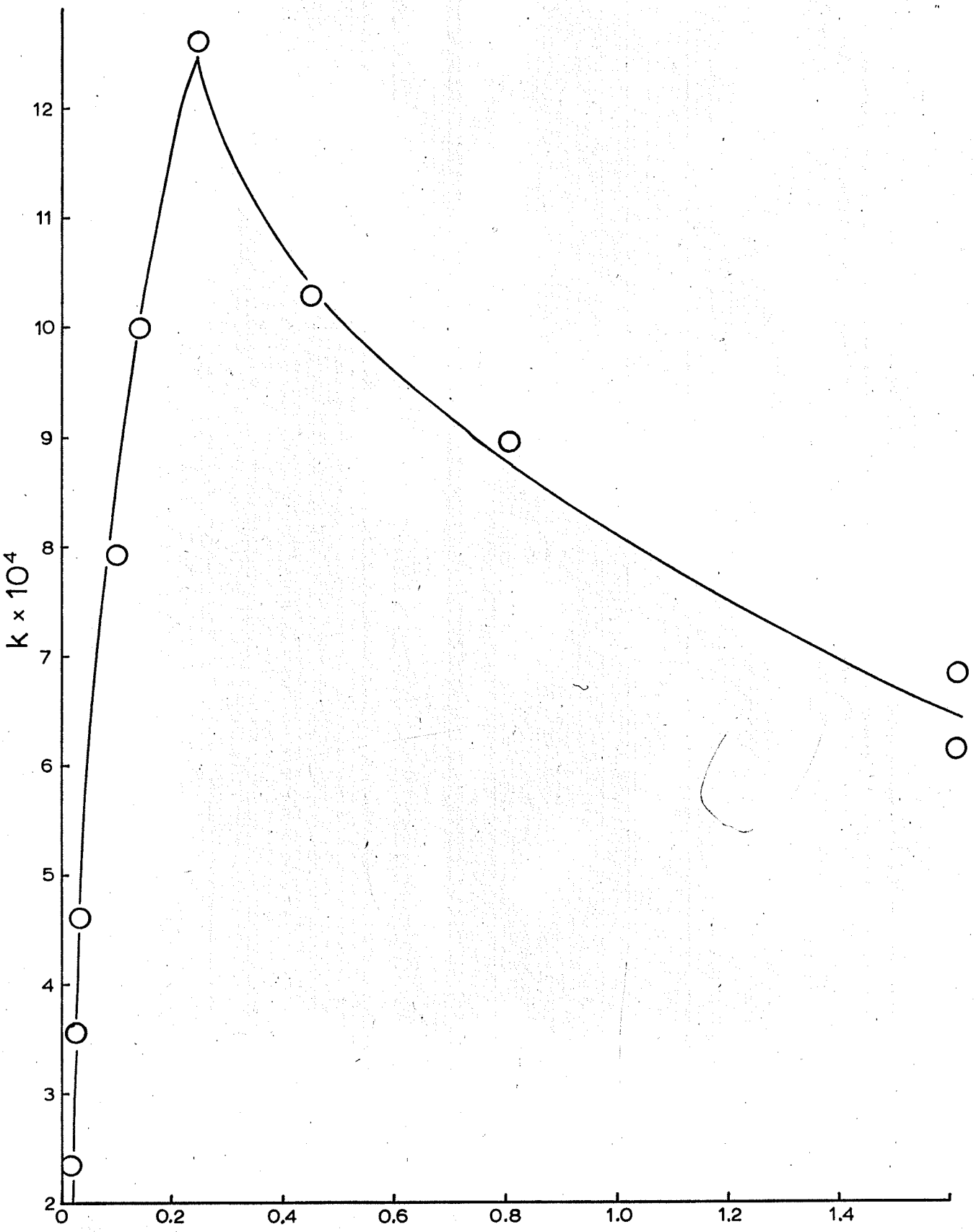
(2) While 4-methylsalicylic acid does not decarboxylate in nitrobenzene solution, the reaction does proceed in mixtures of quinoline and nitrobenzene. The effect of quinoline concentration on the rate of decarboxylation is shown in Figure 3. For quinoline concentrations up to .03 molar, it was observed that the reaction is first order with respect to the concentration of quinoline. Thus quinoline is involved in the reaction prior to, or in the rate-determining step.

Figure 3 shows that as the quinoline concentration increases, the rate of the reaction increases until a maximum is reached at a quinoline concentration of about .25 molar, after which the rate decreases. The shape of this curve suggests that the catalytic effect of increasing the quinoline concentration is opposed by some other retarding factor. Rodewald suggests that this effect is probably due to the decrease in the dielectric constant of the medium as the quinoline concentration increases.

(3) As quinoline is involved prior to or in the rate-determining step of the reaction, Rodewald determined what effect a change in the electron density on the N-atom of quinoline would have on the rate of the reaction. It was found that substituents on quinoline which increased the electron density on the nitrogen atom enhanced the rate of decarboxylation. It was shown that this was not a dielectric effect.

FIGURE 3: The effect of quinoline concentration upon the rate of decarboxylation of 4-methylsalicylic acid in quinoline-nitrobenzene solutions.

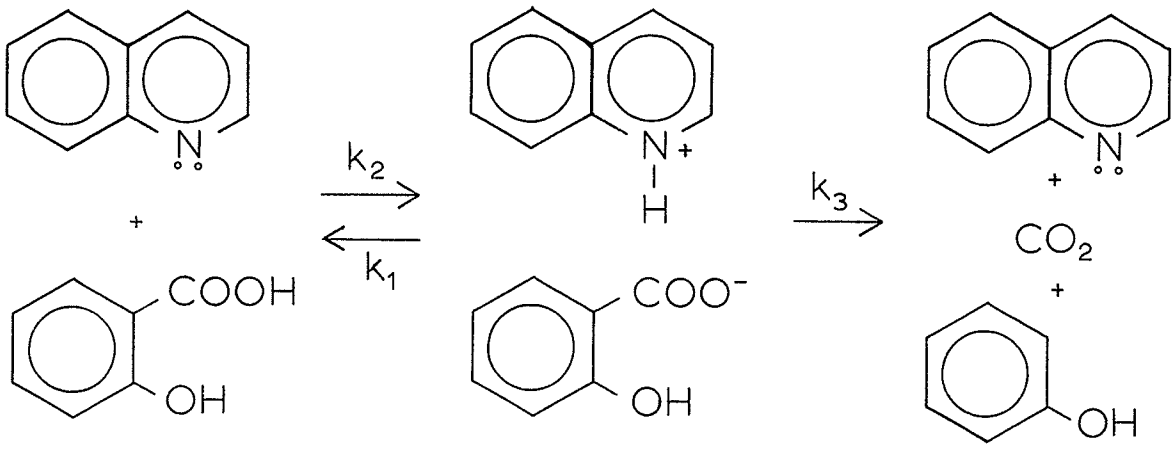
FIGURE 3


k x 10⁴
12
11
10
9
8
7
6
5
4
3
2
0
0.2
0.4
0.6
0.8
1.0
1.2
1.4
CONCENTRATION OF QUINOLINE IN NITROBENZENE (MOLES / LITER)

(4) Application of the Hammett equation and its extensions leads to the conclusion that O-H bond-breaking in the COOH group and ( $\alpha$-C)-H bond-making are involved prior to or in the rate-determining step.

(5) The possibility that the reaction occurred by a free radical mechanism was eliminated.

Any mechanism proposed for the decarboxylation of salicylic acids in quinoline would have to account for all of the above evidence. Rodewald has proposed the following mechanism for the reaction.




This mechanism is consistent with all of the experimental facts.

The mechanism predicts the observed pseudo first order kinetics for the reaction in quinoline solution, and also predicts that O-H bond-breaking and C-H bond-making are involved prior to, or in the rate-determining step.

The mechanism can also explain the variation in the rate of decarboxylation of 4-methylsalicylic acid in quinoline-nitrobenzene mixtures. The rate of the reaction is determined by the concentration of ion pairs and their rate of decomposition. As nitrobenzene is more polar than quinoline, we would expect that ion pair formation would be favored in solutions of low quinoline concentration and the rate of reaction should be greater than in pure quinoline. As the concentration of

quinoline is increased, however, two opposing effects come into play. The added quinoline will tend to shift the ion pair formation to the right, but will also tend to decrease the dielectric constant of the medium, thus retarding ion pair formation. These two opposing effects would explain the shape of the curve in Figure 3. The addition of quinoline up to a concentration of about .25 M would favor the formation of ion pairs; further addition of quinoline results in the dielectric effect predominating.

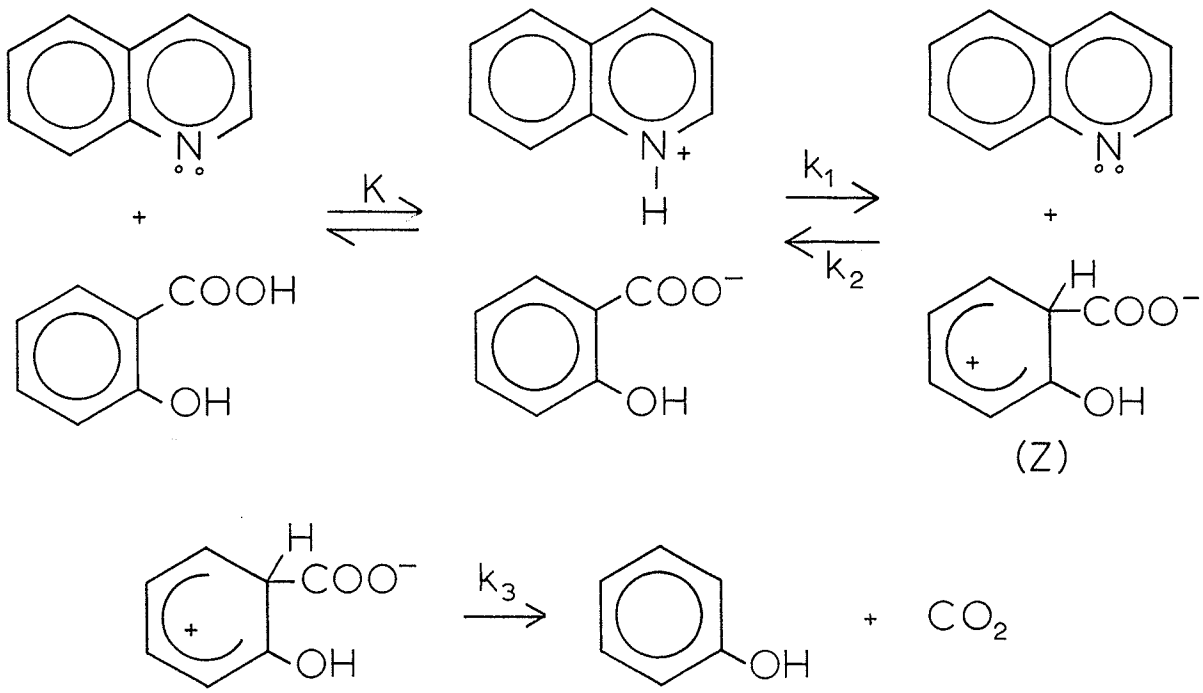
As the electron density on the quinoline nitrogen atom increases, the equilibrium formation of ion pairs shifts to the right while the rate of C-H bond-making is decreased. If the former effect is more important, then the proposed mechanism can account for the observed increase in rate with the increase in the electron density of the quinoline nitrogen atom.

Thus, a possible mechanism for this reaction involves the equilibrium formation of ion pairs, followed by a rate-determining step in which the quinolinium ion protonates the α-carbon of the acid anion and C-H bond-making and C-C bond-breaking occur in a concerted manner.

Another possible mechanism for this reaction, proposed by Bourns (9), involves the equilibrium formation of ion pairs, followed by protonation of the α-carbon of the acid anion by quinolinium ion, to form a reaction intermediate, which can decompose either to products or to an ion pair. This "stepwise" mechanism is given on page 39.

The two mechanisms differ in the status of the species Z. In the concerted mechanism, Z is an activated complex while in the stepwise mechanism, it is a reaction intermediate.

The present problem is, therefore, to decide which of these two mechanisms is the correct one. The problem may be solved by measuring the carboxyl-$C^{13}$ kinetic isotope effect for this reaction. This problem is very similar to the one reported by Lynn and Bourns (24), which has




already been discussed. In both cases, either a concerted or a stepwise mechanism is possible. As in the previous case, the measurement of the isotope effect in this reaction under any given conditions will not lead to any definite conclusion as to which mechanism is the correct one. Should an isotope effect be observed in this reaction, then the concerted mechanism merely requires that there be appreciable C-C bond-breaking in the activated complex, while the stepwise mechanism would require that the decomposition of Z to products (with C-C bond-breaking) must be the rate-determining step. If no isotope effect is observed, the concerted mechanism merely requires that C-C bond-breaking is not appreciable in the activated complex, whereas the stepwise mechanism would require that the formation of Z is rate-determining. However, the two mechanisms do give rise to different predictions as to the variation of the magnitude of the kinetic isotope effect with the variation in the concentration of quinoline.

If the stepwise mechanism were the correct one, then we would predict that in solutions of very low quinoline concentration, the formation of ion pairs, and therefore of Z, should be at least partially

rate-determining. Therefore, we would expect to observe a small (if any) isotope effect under these conditions. As the quinoline concentration is increased, the rate of decomposition of Z to ion pairs would also be increased. In the limiting case of pure quinoline solvent, we would expect that the rate of decomposition of Z to products should become at least partially rate-determining and we would expect an isotope effect larger than that observed in solutions of very low quinoline concentration. Thus, the stepwise mechanism would predict that the magnitude of the kinetic isotope effect should increase with an increase in quinoline concentration.

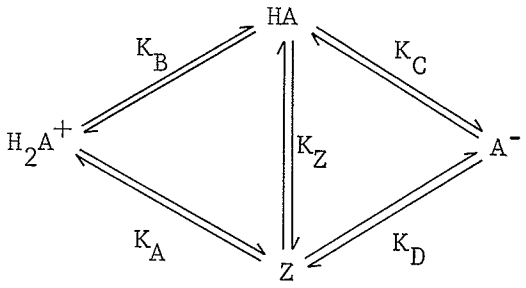
If the concerted mechanism were the correct one, then any observed isotope effect would be due to the extent of C-C bond-breaking achieved in the activated complex formed in the decomposition of an ion pair. As the quinoline concentration cannot affect the extent of C-C bond-breaking, we would expect that the quinoline concentration would have no effect on the magnitude of the observed isotope effect.

Thus a measurement of the carboxyl-$C^{13}$ kinetic isotope effect in the decarboxylation of 4-methylsalicylic acid in both pure quinoline and a quinoline-nitrobenzene solution of very low quinoline content, should settle the problem.

(B) The Decarboxylation of 4-Methoxyanthranilic Acid in Aqueous Solutions

The rate of decarboxylation of 4-methoxyanthranilic acid in aqueous solutions is of first order with respect ot the concentration of the acid. Dunn, Legatte and Scheffler (11) have measured the rates of decarboxylation of this acid in buffer solutions of different pH (from -.3 to 4) and constant ionic strength (0.50) at 60°C. The observed dependence of the rate constant (k) upon pH is shown graphically in Figure 4. Examination of this curve shows that k is a maximum at a pH of about 1.1-1.4, and decreases at both higher and lower pH values. Any mechanism proposed for this reaction must explain the shape of the curve in Figure 4 and the value of the pH at the maximum value of k.

Bjerrum (7) has proposed that in aqueous solutions of amino acids, four organic species are in equilibrium with hydronium ion. These species, referred to as the Bjerrum species, can be represented by $HOOC\text{-}Ar\text{-}NH_3^+$ (abbreviated $H_2A^+$), $HOOC\text{-}Ar\text{-}NH_2$ (HA), $^-OOC\text{-}Ar\text{-}NH_3^+$ (Z), and $^-OOC\text{-}Ar\text{-}NH_2$ ($A^-$). The equilibria are shown in equation (18), with the hydronium ions omitted.



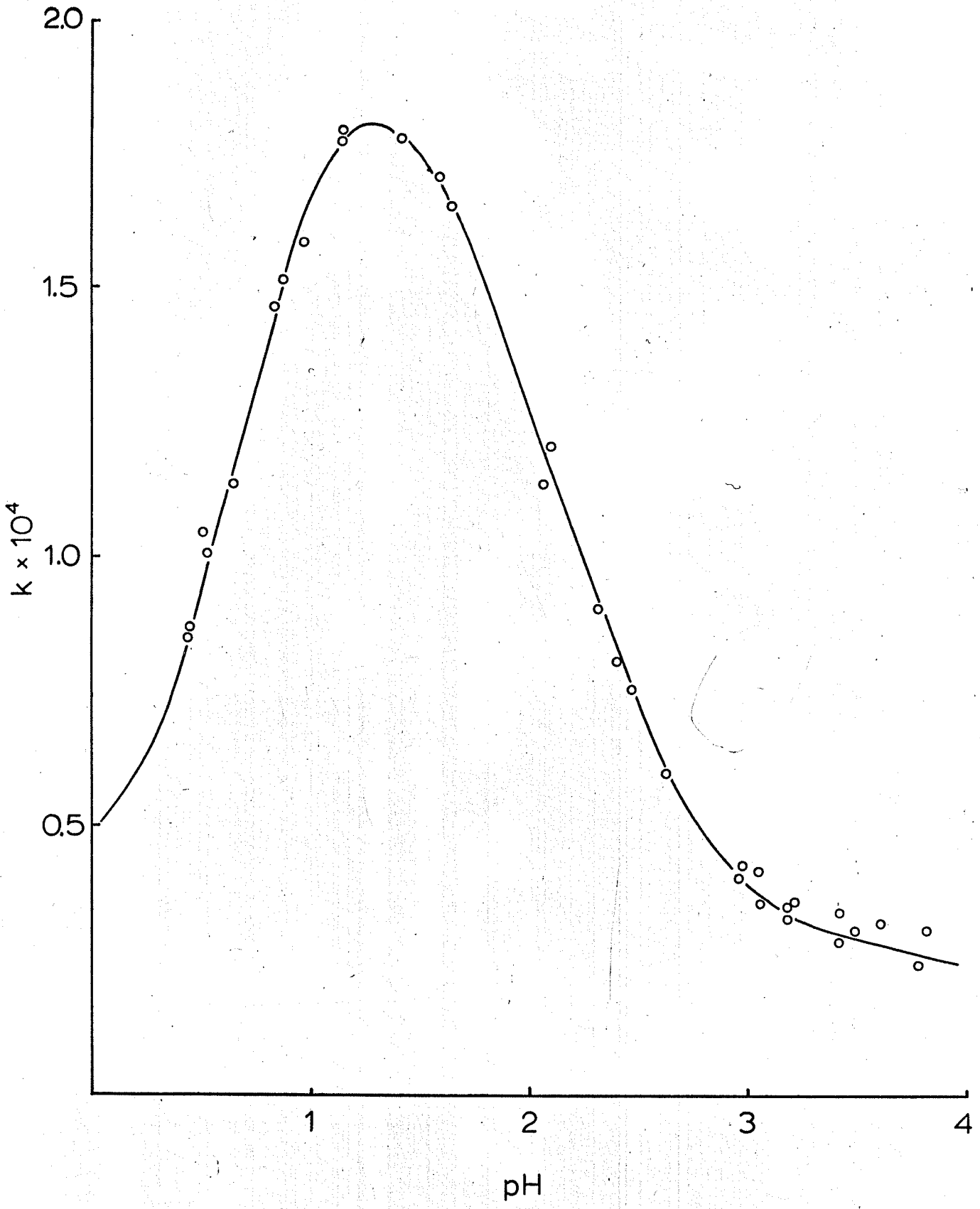

....(18)

It is possible that the reaction takes place by way of one (or more) of the Bjerrum species. However, Dunn et al (11) have shown that the pH dependence of the rate constant cannot be accounted for by reaction of any combination of Bjerrum species, and it was concluded that decarboxylation must take place via some intermediate which is not part of the Bjerrum system.

Dunn et al have proposed a mechanism for the reaction in which

FIGURE 4: The observed pH dependence of the rate constant (k) for the decarboxylation of 4-methoxyanthranilic acid in solutions of ionic strength 0.5 at 60°C.

FIGURE 4



the non-Bjerrum intermediates $H_2A^*$, HA* and HZ* are formed by protonation of the α-carbon of HA, $A^-$ and Z, respectively. The protonation of $H_2A^+$ to form $H_3A^*$ can be ignored as this would lead to an increase in the rate at low pH. This mechanism is shown in equation (19) on page 44.

Assuming that all three non-Bjerrum intermediates decarboxylate, the following expression has been derived (11) for the rate of decarboxylation, v.

$$v = \frac{\left[N\right]\left\{k_A K_2 + (k_{HA}K_B/K_1 + k_Z K_A/K_1)\left[H^+\right]\right\}\left\{k^* + (k^+/K_C^* + k^{\neq}/K_D^*)\left[H^+\right]\right\}}{k^* + k_{-A} + \left\{(k^+ + k_{-HA})/K_C^* + (k^{\neq} + k_{-Z})/K_D^*\right\}\left[H^+\right]} \quad \ldots.(20)$$

Where $\left[N\right] = \left[HA\right] + \left[Z\right]$

and $K_1$ and $K_2$ are defined in equations (21) and (22).

$$H_2A^+ \underset{}{\overset{K_1}{\rightleftharpoons}} H^+ + N \quad \ldots.(21)$$

$$N \overset{K_2}{\rightleftharpoons} H^+ + A^- \quad \ldots.(22)$$

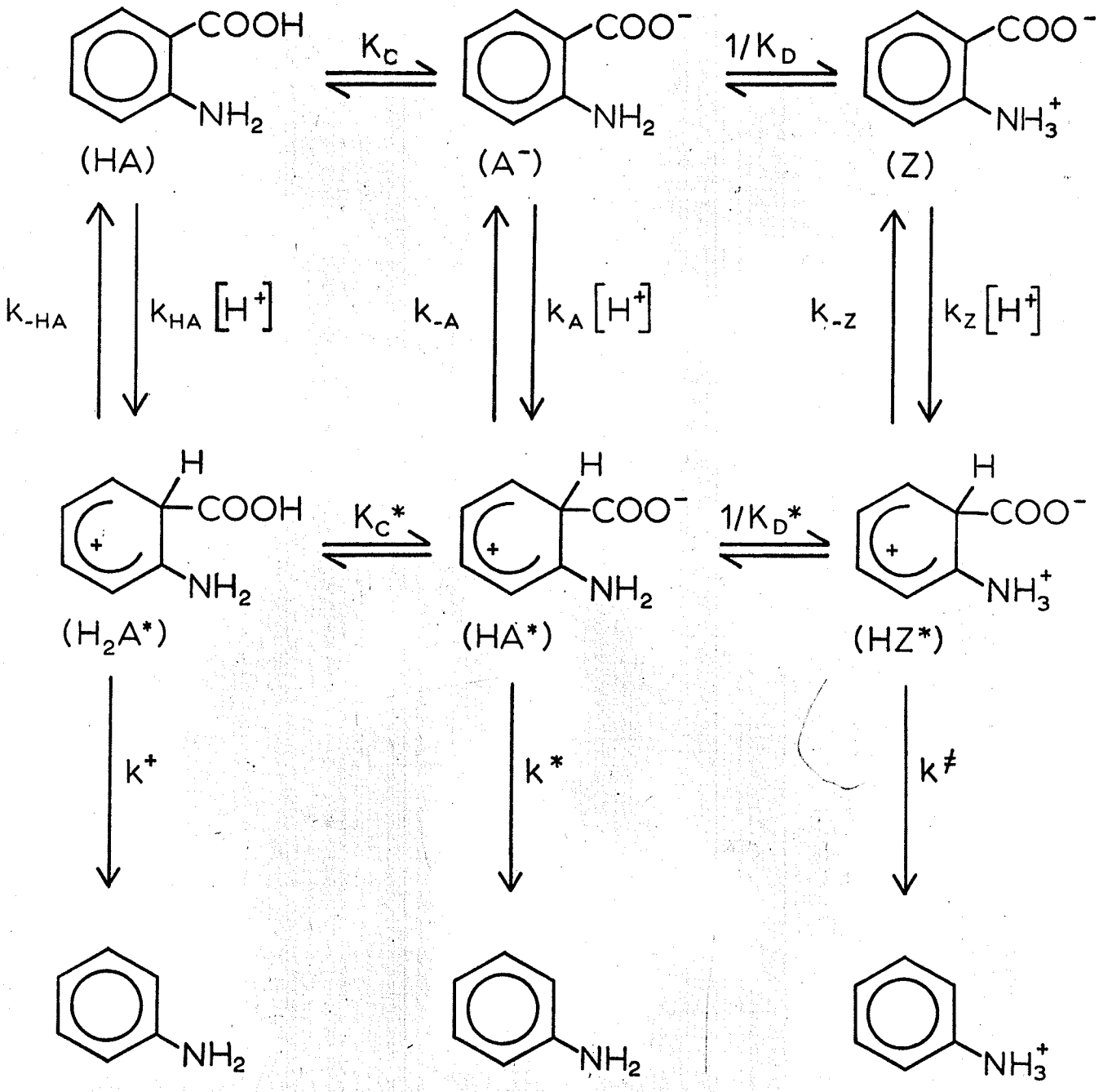
If the rate of decarboxylation is much greater than the rate of deprotonation for all three intermediates, or if the rate of deprotonation is much greater than the rate of decarboxylation for all three, equation (20) reduces to a form which does not fit the experimental data. Thus neither of these conditions actually exists.

In its present form, equation (20) does not fit the experimental data as the $\left[H^+\right]^2$ terms in the numerator prevent the rate from becoming small at low pH. For the rate to be small at low pH we require that either

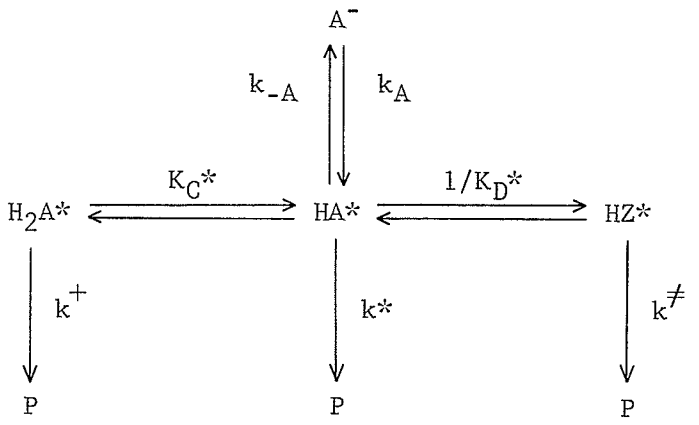
(a) $k_{HA} = k_Z = 0$, in which case equations (19) and (20) reduce to equations (23) and (24), respectively.

or (b) $k^+ = k^{\neq} = 0$, in which case equations (19) and (20) reduce to equations (25) and (26), respectively.

Equations (23) to (26) are given on page 45.

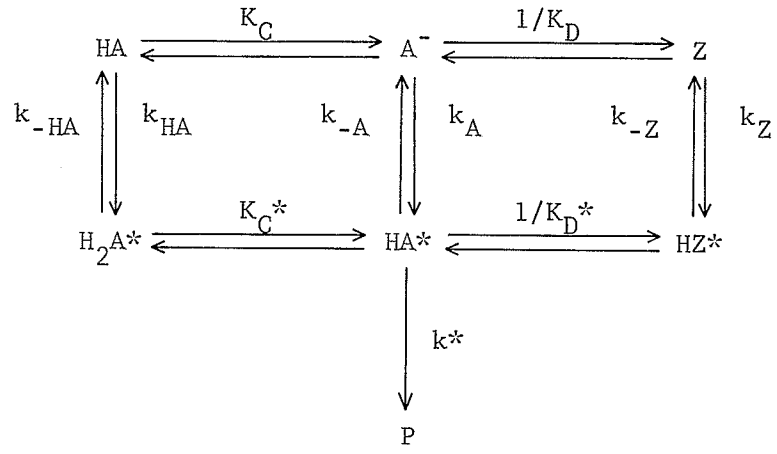

COOH
NH2
Kc
COO-
1/KD
NH3+
(HA)
(A-)
(Z)
k-HA
kHA [H+]
k-A
kA [H+]
k-Z
kZ [H+]
H
Kc*
1/KD*
(H2A*)
(HA*)
(HZ*)
k+
k*
k≠

....(19)




....(23)

$$v = \frac{[N]k_A K_2\left\{k^* + (k^+/K_C^* + k^{\neq}/K_D^*)[H^+]\right\}}{k^* + k_{-A} + \left\{(k^+ + k_{-HA})/K_C^* + (k^{\neq} + k_{-Z})/K_D^*\right\}[H^+]} \qquad \ldots.(24)$$




....(25)

$$v = \frac{[N]k^*\left\{k_A K_Z + (k_{HA}K_B/K_1 + k_Z K_A/K_1)[H^+]\right\}}{k^* + k_{-A} + (k_{-HA}/K_C^* + k_{-Z}/K_D^*)[H^+]} \qquad \ldots.(26)$$

Therefore, if equation (19) represents the general mechanism of this reaction, one or both of $H_2A^*$ and HZ* must participate, but either they are not formed by direct protonation of HA and Z or they do not decarboxylate. Thus the present problem is to determine whether the reaction proceeds by mechanism (23) or by mechanism (25). The problem may be solved by measuring the carboxyl-$C^{13}$ kinetic isotope effect in this reaction.

Equations (24) and (26) are both of the general form

$$v = \frac{a + b[H^+]}{c + d[H^+]}[N]$$

where a, b, c and d are constants.

$$\therefore v = \frac{b}{d}\frac{a/b + [H^+]}{c/d + [H^+]}[N] \qquad \ldots.(27)$$

The ratios a/b, b/d and c/d have been evaluated (11): substitution of the values of these ratios in the above equation gives

$$v = 0.00506\frac{0.001 + [H^+]}{0.205 + [H^+]}[N] \qquad \ldots.(28)$$

If the pH of the solution is greater than 4 (i.e. $[H^+] < .0001$ N), the $[H^+]$ terms in equation (28) become negligable and equation (27) reduces to

$$v = (a/c)[N]$$

For equations (24) and (26),

$$v = \frac{k^*K_AK_2}{k^* + k_{-A}}[N] \qquad \ldots.(29)$$

Therefore, at any pH greater than 4, both mechanisms obey equation (29). As this equation contains k* (a rate of carbon-carbon bond rupture), we would predict that there will be an isotope effect unless $k^* >> k_{-A}$, as this would result in the disappearance of k* from equation (29).

As we cannot predict the relative magnitudes of k* and $k_{-A}$ in mechanism (25), the existence or absence of a kinetic isotope effect could be accommodated by this mechanism.

However, for mechanism (23),

$$\frac{a/b}{c/d} = \frac{k^*}{k^* + k_{-A}}\left(1 + \frac{k_{-HA}K_D^* + k_{-Z}K_C^*}{k^+K_D^* + k^{\neq}K_C^*}\right) = 0.0018$$

As the value of the term in parentheses must be greater than one, $k^*/(k^* + k_{-A})$ must be much less than one and, therefore, we must have $k^* << k_{-A}$.

Thus if the reaction actually proceeds via mechanism (23), we would predict that an isotope effect would be observed when pH > 4.

If the pH of the reaction solution is -.3 or less, then we can see from equation (28) that the a/b and c/d terms are negligible compared to the $[H^+]$ terms. Thus, equation (27) reduces to

$$v = (b/d)\,[N]\ .$$

Substituting for b and d from equations (24) and (26), we arrive at equations (30) and (31), respectively.

$$v = \frac{[N]k_A K_2(k^+/K_C^* + k^{\neq}/K_D^*)}{(k^+ + k_{-HA})/K_C^* + (k^{\neq} + k_{-Z})/K_D^*} \qquad \ldots.(30)$$

$$v = \frac{[N]k^*(k_{HA}K_B/K_1 + k_Z K_A/K_1)}{k_{-HA}/K_C^* + k_{-Z}/K_D^*} \qquad \ldots.(31)$$

As equation (30) contains $k^+$ and $k^{\neq}$ (both of which refer to a step involving carbon-carbon bond-breaking), we would predict that a kinetic isotope effect should be observed if mechanism (23) is operative, unless $k^+ >> k_{-HA}$ and $k^{\neq} >> k_{-Z}$. However, as previously stated, this condition cannot exist as equation (20) - and therefore also equation (24) - would reduce to a form which would not fit the data.

Thus, mechanism (23) predicts that a kinetic isotope effect would be observed at a pH of -.3 or less.

As k* is found in equation (31), mechanism (25) would also predict that a kinetic isotope effect would be observed at a pH of -.3 or less.

In summary then, mechanism (23) requires a kinetic isotope effect at both low and high pH values: mechanism (25) requires an isotope effect at a low pH value but can accommodate an effect or none at a high pH.

# EXPERIMENTAL

## MATERIALS

Quinoline (synthetic, Matheson Coleman and Bell) was dried over Drierite (anhydrous $CaSO_4$) and then distilled under vacuum. The fraction boiling from 105-106°C at 25 mm. was collected and stored in the dark over BaO (Barium and Chemicals, Inc.) .

Nitrobenzene (Fisher) was dried over Drierite and then fractionated on a 28 cm. Vigreux column. The fraction collected over the range 208-209°C (at 739 mm.) was stored in the dark over Drierite.

4-Methylsalicylic acid was synthesized according to the directions of Rodewald (29). The product had a melting point of 176.5-178.1°C [literature values: 176°C (26), 177°C (39)] .

4-Methoxyanthranilic acid was synthesized according to the method of Legatte and Dunn (21). The product had a melting point of 180.9-181.7°C [literature value, 180-181°C (21)] .

Buffer Solutions

2 N HCl: Prepared by dilution of 12 N HCl, this buffer had a pH of -.3 .

HCl-KCl: This buffer was prepared by mixing 139.4 ml. of 0.557 N HCl with 782.5 ml. of 0.540 N KCl and diluting the resulting solution to one liter. This buffer was of ionic strength 0.5 and had a pH of 1.3 .

Formate: This buffer was prepared by mixing 500 ml. of a solution which was 0.45 N KCl and 0.05 N sodium formate, with 32.75 ml. of another solution which was 0.45 N KCl and 0.20 N HCl. The resulting solution had a pH of 4.0 and ionic strength of 0.5 .

## APPARATUS

### Apparatus Used in the Decarboxylations of 4-Methylsalicylic Acid

The reaction train is shown in Figure 5. The nitrogen sweep gas was purified by passing it through two anhydrone U-tubes (A) and one ascarite U-tube (B) to remove water and $CO_2$, respectively. The three-way stopcock I would permit the nitrogen gas to sweep through either arm "a" (through the reaction vessel (D), the condenser (E) and stopcock II), or arm "b" (bypassing D and E). The thermostat (C) consisted of a two liter round-bottom flask with an attached condenser. About 350 ml. of liguid in C were brought to a gentle reflux by heating with an electric heating mantle. The reaction vessel (D) was thus bathed in the hot vapors of the liquid and maintained at a constant temperature ($\pm$ .5°C). Nitrobenzene and ethylene glycol were used to attain reaction temperatures of 208°C and 195°C, respectively. Temperatures were measured using an iron-constantan thermocouple with the hot junction in the glass well of the reaction vessel and the cold junction in an ice-water bath (J). The difference of potential between the two junctions was measured with a potentiometer (K). The reaction vessel (D) is shown enlarged in Figure 6. The condenser (E), containing a coil rather than a straight tube, trapped the vapors of quinoline, nitrobenzene and meta-cresol which escaped from the reaction vessel during the run. The n-butylphthalate bubbler (F) served as a visual check of the nitrogen flow rate and also removed traces of organic vapors. The trap (G) was immersed in a dry ice-acetone bath (H) and removed any remaining impurities. The exit arm of G was equipped with a 10/30 ground glass outer joint.

### Apparatus Used in the Decarboxylations of 4-Methoxyanthranilic Acid

The reaction vessel (L) is shown in Figure 7. It consisted of either a 250 or a 500 ml. round-bottom flask, with a 24/40 ground glass

FIGURE 5: The apparatus used in the decarboxylations of 4-methylsalicylic acid.

FIGURE 5

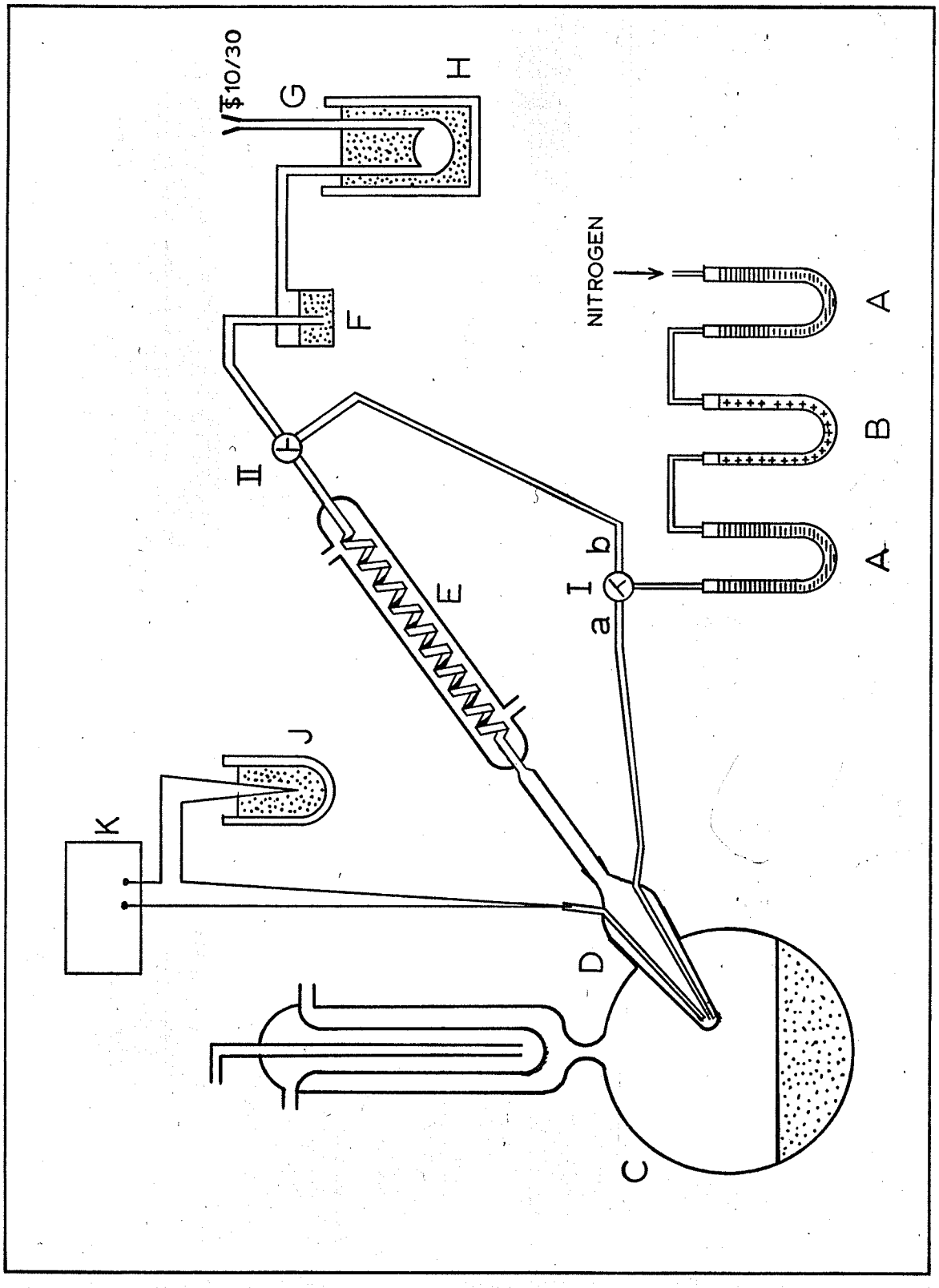


FIGURE 6

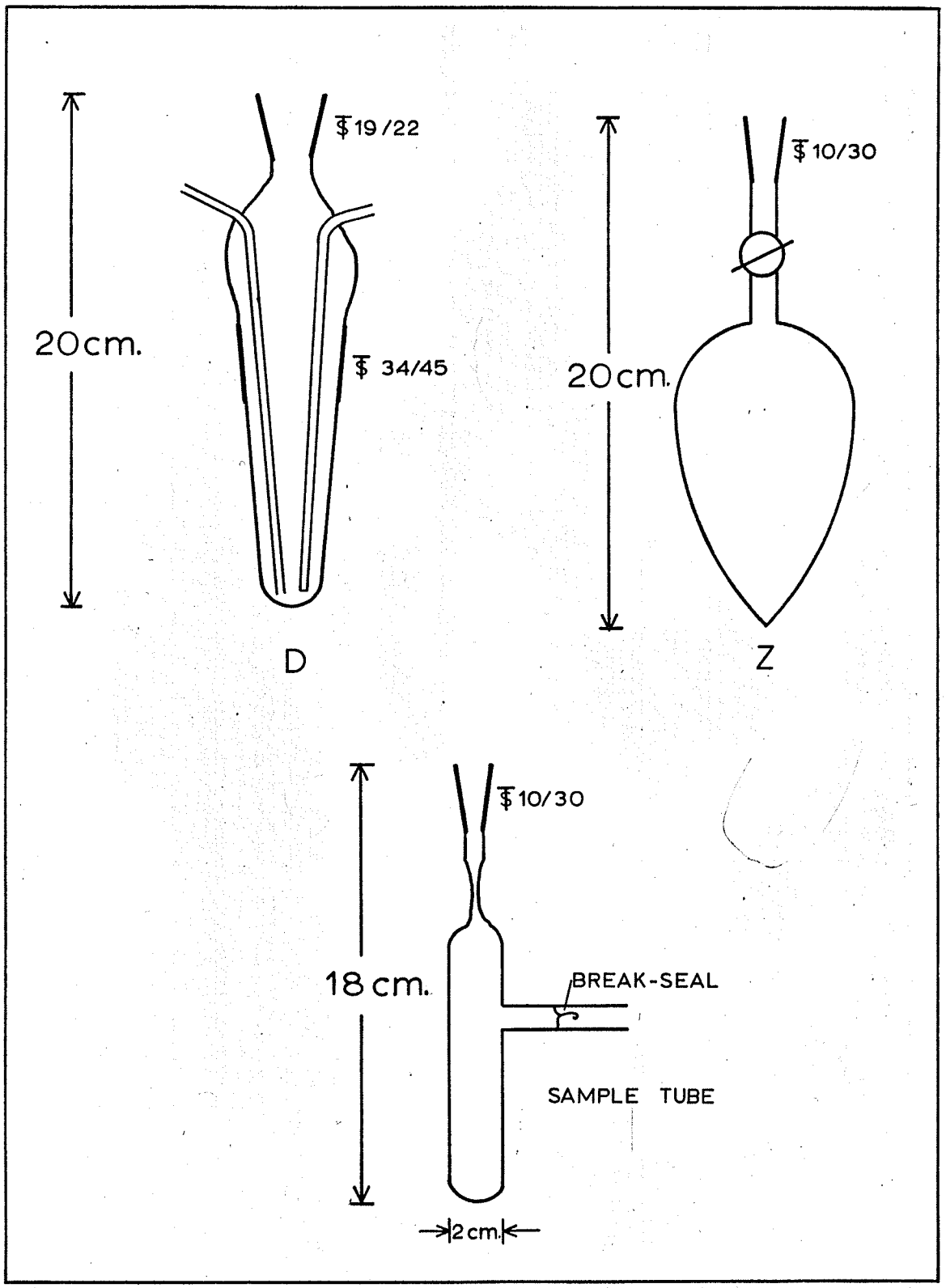
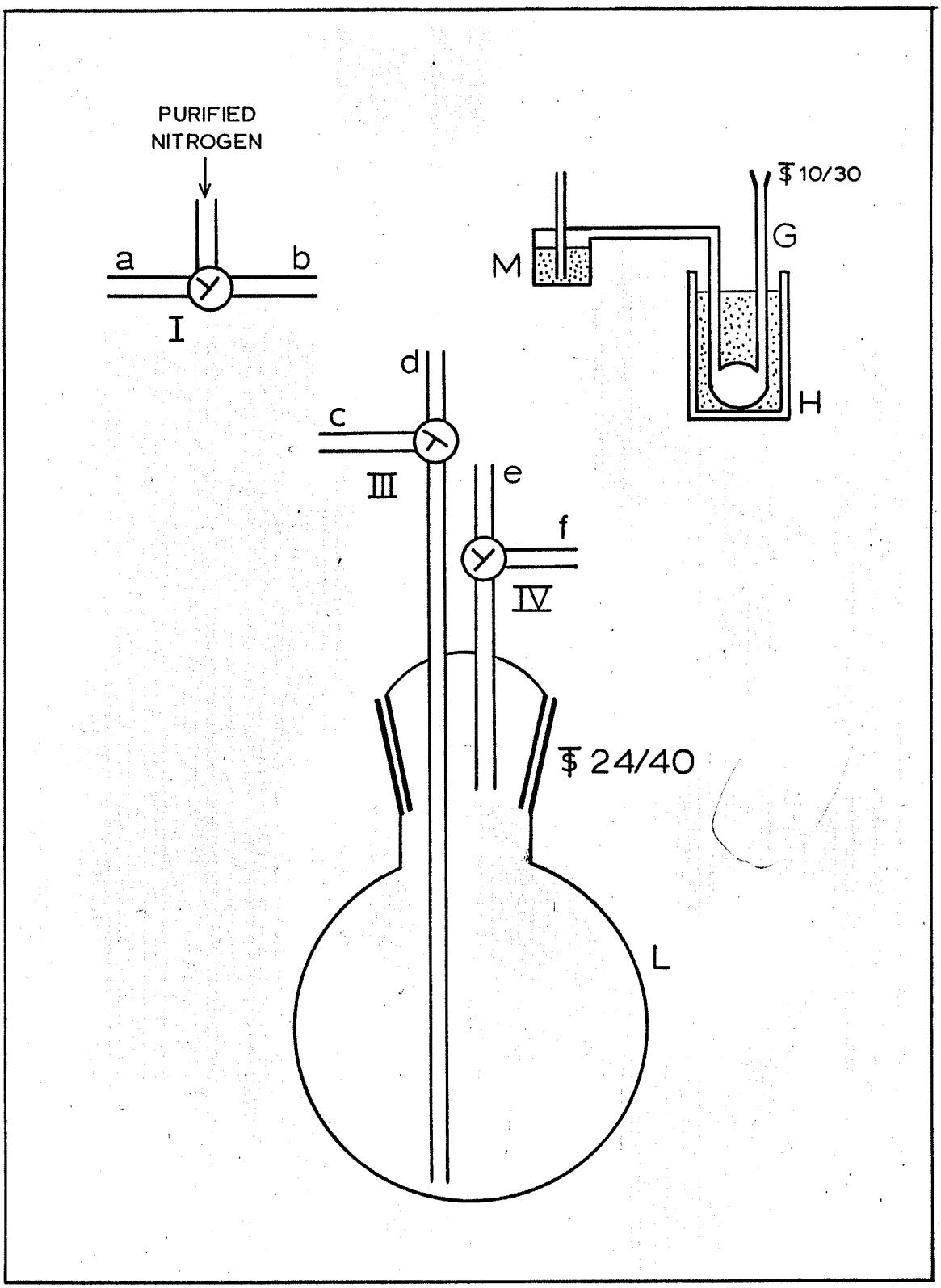

20cm.
$ 19/22
$ 34/45
D
20cm.
$ 10/30
Z
18 cm.
$ 10/30
BREAK-SEAL
SAMPLE TUBE
2 cm.

FIGURE 7: The apparatus used in the decarboxylations of 4-methoxyanthranilic acid.

FIGURE 7



joint and the accompanying fitting with the same glass joint and the two three-way stopcocks III and IV. A nitrogen sweep was used to collect the $CO_2$ produced in the reaction. The nitrogen gas was purified by passing it through the U-tubes A and B (shown in Figure 5) and the flow of the purified gas was controlled with the three-way stopcock I. The bubbler (M), containing concentrated $H_2SO_4$, served as a visual check of the nitrogen flow rate and also removed water vapor from the $N_2$-$CO_2$ mixture. The trap (G), immersed in the dry ice-acetone bath (H), was the same as in Figure 5 and served to remove any remaining impurities. The reaction temperature ($60 \pm .5^oC$) was obtained by immersing the vessel (L) up to the neck of the flask in a vigorously stirred oil bath maintained at $60^oC$.

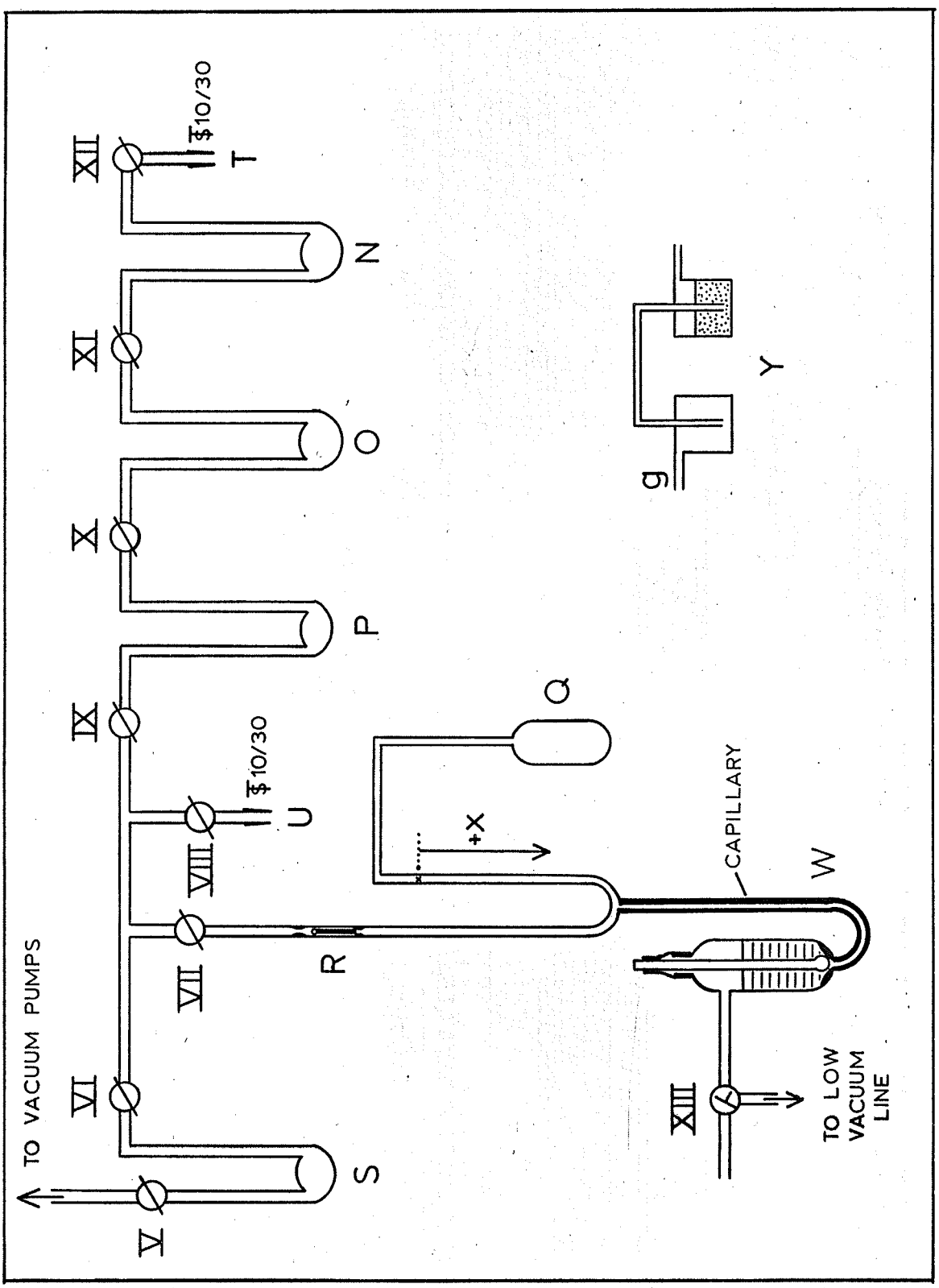
The High Vacuum System

The high vacuum system shown schematically in Figure 8 was used to collect and purify the $CO_2$ produced in the decarboxylation reactions, and also to measure the number of moles of $CO_2$. The high vacuum, two-way stopcocks (V - XII) were greased with either Apiezon "M" or Dow Corning Silicone stopcock grease. A rotary oil pump and a mercury diffusion pump were used to attain the high vacuum. The system was of a conventional design and the only parts requiring any special description are those labelled R, Q, and W.

W permitted mercury to be admitted to, and removed from the U-bend in the glass tubing between R and Q. It consisted of a mercury reservoir connected by capillary tubing to the bottom of the U-bend between R and Q. In the mercury reservoir was a glass rod, the lower end of which was ground glass. This rod was seated in a ground glass base at the mouth of the capillary tubing and it was held in place by a piece of rubber tubing stretched over the upper end of the rod and the neck of the reservoir. Above the mercury level in the reservoir was attached

FIGURE 8: The high vacuum system.

FIGURE 8


TO VACUUM PUMPS
XII
$10/30
T
N
XI
O
X
P
IX
$10/30
U
VIII
VII
R
Q
+X
CAPILLARY
W
VI
S
V
g
Y
XIII
TO LOW VACUUM LINE

a three-way stopcock (XIII); one arm was connected to a (low) vacuum line and the other was open to the atmosphere. With Qunder high vacuum, VII was closed and XIII turned so that the vacuum line was blocked and the reservoir open to the atmosphere. By raising the glass rod in the reservoir, mercury was allowed to flow through the capillary and into the U-bend at a slow and controllable rate. Releasing the glass rod sealed off the reservoir, and the flow of mercury ceased. By turning XIII so that the reservoir was connected to the vacuum line, raising the glass rod allowed the mercury to flow back into the reservoir. To remove all the mercury from the U-bend, the length of the capillary should be at least 20 cm., as the vacuum line provides a vacuum of only about 40 mm. and the mercury level in the capillary will remain at least this height above the level of mercury in the reservoir.

R is a mercury float valve. It consisted of a metal bar encased in a glass tube, the upper end of which was ground glass. The lower end of the glass tube rested on a constriction in the tubing. If the mercury in the U-bend rose past the constriction where R was resting, the glass-encased rod floated up to a ground glass constriction above. Thus, the float valve sealed off the tubing and prevented mercury from getting through VII and into the rest of the vacuum system.

Q was a glass bulb with a capacity of about 25 ml. .

## PROCEDURE

### Collection and Purification of $CO_2$

(A) 4-Methylsalicylic Acid

The reaction train was assembled as shown in Figure 5, except that trap G was not yet immersed in the bath H. The exit arm of G was attached to the vacuum system at T. Stopcocks VI and VII of the vacuum system were closed and all of VIII to XII were open. The bubbler (Y) was

attached at g to the vacuum system at U. The right hand bulb of Y contained concentrated $H_2SO_4$, and served as a visual check of the flow rate of the effluent nitrogen. After this system was assembled, the flow of nitrogen was begun, and I and II were turned so that the nitrogen flowed through arms a and b simultaneously. The entire system was swept with nitrogen for 1-2 hours. During this time, the liquid in the thermostat was brought to a gentle reflux, and a sample of the acid was weighed out. In runs Q-1 and Q-2, .04 gram samples were weighed into small glass "cups". In the remaining runs, .37 gram samples were shaped into pellets with a hydraulic pellet press. After the nitrogen flushing, D and E were separated and 10 ml. of solvent (quinoline, or quinoline and nitrobenzene) were quickly added by syringe to D. Then D and E were quickly joined again, and I was turned so that the nitrogen flowed only through arm a. The nitrogen flushing was continued for 15-30 minutes, allowing the solvent to equilibrate with the thermostat. Then G was immersed in the bath H, and traps N, O and P were immersed in liquid nitrogen baths. I was turned so that the nitrogen flowed through arms a and b. Then D and E were separated, the acid quickly dropped into D, and D and E were immediately rejoined. I and II were turned so that the nitrogen flowed only through arm a. Thus as the $CO_2$ was produced, it was swept out of D, purified by passage through E, F and G, and frozen out in N, O and P. After the desired time, I and II were turned so that the nitrogen flowed only through arm b and the system was flushed for 10-20 minutes. Then VIII and XII were closed, VI and VII were opened, and the system was evacuated. After a high vacuum had been obtained, IX was closed and the contents of O and P were transferred to N by removing the liquid nitrogen baths from O and P. Then O and P were immersed in dry ice-acetone baths, VI was closed, and IX opened. By moving the liquid nitrogen bath from N to Q, the $CO_2$ in N was transferred to Q, and any impurities were trapped

in O and P. Repeating this process, the $CO_2$ was passed through O and P a total of eleven times(+), and the purified $CO_2$ ended up in Q. After closing VII, mercury was admitted to the U-bend between R and Q, and the liquid nitrogen bath was removed from Q. The purified $CO_2$ thus warmed to room temperature and expanded against the mercury in the U-bend.

(B) 4-Methoxyanthranilic Acid

A sample of acid was weighed into a small weighing bottle (about 8 ml. capacity) and then dissolved in a minimum amount of 0.25 N NaOH. The weighing bottle was then dropped into the round-bottom flask (L) containing buffer solution. The acid at first precipitated, and then redissolved upon swirling. After all the acid was dissolved, the bubbler attachment was greased and connected to L. Arm a of I was connected to c of III, and then III and IV were turned from the positions shown in Figure 7 to the positions ⊖ and ⊣, respectively, to permit nitrogen flushing of the reaction vessel and its contents. After flushing for 30-40 minutes, III and IV were turned to ⊗ and ⊗, respectively, to seal the flask, and a and c were disconnected. A low vacuum line was then connected to e. The reaction vessel was partially evacuated by turning IV to ⊣, and then resealed as before. The flask (L) was then immersed in the oil bath up to the neck, and shaken continuously for ten minutes. After standing for the desired time, the vessel was removed from the oil bath and cooled in an ice bath. Connections were made between a and d, b and f, and e and M. Bath H was not yet in place. G was attached to the vacuum system at T. Stopcocks VI and VII of the vacuum system were closed and all of VIII to XII were open. The bubbler Y was attached at g to the vacuum system at U. III and IV were both turned to ⊥ and I turned so that nitrogen flowed through both arms a and b. The lines were flushed for 1-1.5 hours and then L was resealed as before. G was immersed

(+) This procedure was used in the purification of all $CO_2$ samples.

in H, and N, O and P were immersed in liquid nitrogen baths. I was turned so that the nitrogen flowed only through a, and III and IV were turned to ⊕ and ⊖, respectively. The $CO_2$ in the vessel was thus swept out, purified by passage through M and G, and frozen out in N, O and P. After flushing for 1.5-2 hours, VIII and XII were closed and VI and VII opened. The $CO_2$ was treated in exactly the same manner as that produced from 4-methylsalicylic acid and was also stored in Q against a mercury column.

Measurement of the Amount of $CO_2$

(1) Calibration of Bulb Q

The volume of the bulb Z (shown in Figure 6) was accurately determined by weighing the bulb when evacuated and then when filled with distilled water at a known temperature. Knowing the density of the water, the volume of Z was calculated and found to be $V_Z = 58.14 \pm .03$ ml. . This bulb was attached to the vacuum system at U. A lecture bottle of $CO_2$ was attached at T with a short piece of reinforced, rubber vacuum tubing. All stopcocks were opened and the system evacuated. After a high vacuum had been obtained, XI was closed, N was immersed in liquid nitrogen, and a small amount of $CO_2$ was frozen into N by opening the valve of the $CO_2$ cylinder. Then XII and VI were closed, XI was opened, and after purification the $CO_2$ was frozen into Z. After closing IX, mercury was admitted to the U-bend between R and Q, and the $CO_2$ in Z was warmed to room temperature, thus creating a difference in the mercury levels in the two arms of the U-bend. The pressure of the $CO_2$ in Z ($P_Z$) was then determined by measuring the difference in the mercury levels with a cathetometer which could be read to 0.1 mm. . The stopcock on Z was closed, VI was opened, and the $CO_2$ in the tubing was frozen into S. Then VI was closed, and the $CO_2$ in Z transferred to Q after first removing the mercury in the U-bend. The mercury was again admitted to the U-bend and the $CO_2$ was warmed to room temperature. The cathetometer was then used to measure

the pressure ($P_Q$) of the $CO_2$ and also the position of the mercury meniscus in the right arm of the U-bend relative to a permanent mark on the glass tubing. The position of the meniscus relative to the mark (in cm.) was referred to as the "x" value of the meniscus. As all pressure measurements were taken at the same temperature, we have

$$V_Q = P_Z V_Z / P_Q$$

where $V_Q$ is the volume of bulb Q and the glass tubing up to the point x (i.e. up to the mercury level).

From each value of $P_Q$, one value of x and of $V_Q$ were obtained. As $P_Q$ could be continuously varied by altering the amount of mercury in the U-bend, a series of corresponding x and $V_Q$ values were obtained for one sample of $CO_2$. This procedure was carried out three times with different amounts of $CO_2$ ($P_Q \simeq$ 50, 85 and 120 mm.) and a total of forty corresponding x and $V_Q$ values were obtained. These points are plotted in Figure 9, where the vertical line through each point indicates the maximum probable error in $V_Q$. Using the method of least squares, the points were fitted to the following equation.

$$V_Q = 30.67 + .3165x \qquad (\pm 0.3\ \%)$$

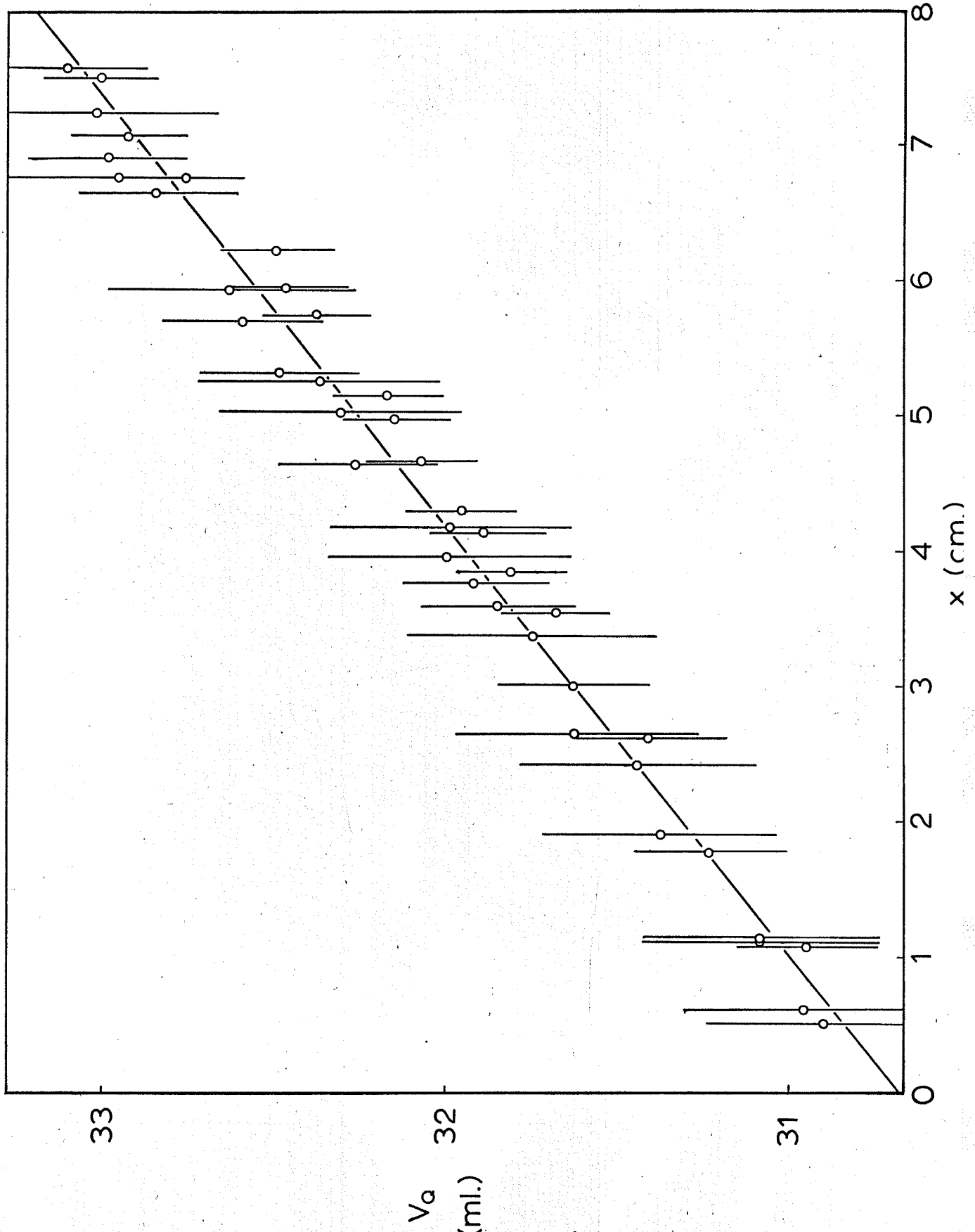
This equation gives $V_Q$ in milliliters. The error indicated is the root mean square deviation.

(2) The purified $CO_2$, obtained from the decarboxylation reactions, was left in Q until at room temperature. The amount of $CO_2$ collected was then determined by measuring several sets of $P_Q$ and x values at temperature T. The number of moles of $CO_2$ (n) in Q is then given by $n = P_Q V_Q / RT$, where $V_Q$ is obtained from the above equation.

About 3.6 ml. of distilled water were placed in Z and then frozen by cooling Z in a dry ice-acetone bath. With the vacuum system under high vacuum, Z was attached at U while still immersed in the bath. After closing VII and IX, VIII and the stopcock on Z were opened. The

FIGURE 9: The calibration curve for bulb Q.

FIGURE 9



ice was degassed in this manner for 30-60 minutes, and then Z was sealed and removed from U. The ice was thawed, refrozen and degassed again. After degassing the ice for the third time, the known amount of $CO_2$ in Q was frozen into Z. Z was then sealed, removed from U and warmed to room temperature. The $CO_2$-water mixture was shaken for twelve hours and then refrozen with dry ice and acetone. Z was attached to the vacuum system at U (while still in the dry ice-acetone bath). VII and IX were closed and VIII opened. After 30-60 minutes, VI was closed, N placed in liquid nitrogen and IX opened. The $CO_2$ in Z was then frozen into N by opening the stopcock on Z. After resealing Z, IX and VIII were closed and Z was removed and warmed to room temperature. The contents of Z were then refrozen and the process repeated twice more. After the third extraction, the $CO_2$ was purified. A sample tube (shown in Figure 6) was attached at U and evacuated for 12-18 hours. The purified $CO_2$ was frozen into the bulb with liquid nitrogen, and after closing VIII, the sample tube was sealed at the constriction. The $CO_2$ samples thus prepared were saved for mass spectrometric analysis.

The $CO_2$ samples were analyzed on a mass spectrometer by Mr. Jan Monster of McMaster University. He measured the relative heights of the mass 44 and 45 peaks and reported the values of $C^{13}/C^{12}$ for each sample of $CO_2$ after making a correction for the contribution of $C^{12}O^{16}O^{17}$ to the mass 45 peak.

## RESULTS

### (A) Decarboxylation of 4-Methylsalicylic Acid

The experimental details of the runs are given in Table IV. The extent of reaction (f) was calculated by dividing the number of millimoles of $CO_2$ obtained by the number of millimoles of acid initially present.

The kinetic isotope effect was evaluated by measuring the isotopic content of the $CO_2$ produced after a measured extent of reaction, and was calculated from equation (17).

$$\frac{k}{k^*} = \frac{\log(1 - f)}{\log(1 - fR_{xf}/R_o)} \qquad \ldots.(17)$$

The values of f were taken from Table IV. $R_o$ and $R_{xf}$ were taken from the values of $C^{13}O_2/C^{12}O_2$ given in Table V. These isotopic ratios were obtained from the mass spectrometric analyses of the $CO_2$ samples. $R_o$ was taken as the average value of $C^{13}O_2/C^{12}O_2$ for runs Q-1 and Q-2. $R_{xf}$ was taken as the value of $C^{13}O_2/C^{12}O_2$ given in Table V for each of runs Q-3 to Q-7. The kinetic isotope effect is given in the last column of Table V.

### (B) Decarboxylation of 4-Methoxyanthranilic Acid

The experimental details of the runs are given in Table VI. From runs W-2 and W-3 it is seen that only 98 % of the theoretical amount of $CO_2$ was obtained after complete decarboxylation of the acid. A correction was made to account for this, and the number of millimoles of acid indicated in Table VI for the partial decarboxylations was calculated from the following expression.

$$\text{mmoles of acid} = (.9799)\frac{\text{weight of sample}}{\text{molecular weight of acid}}$$

The kinetic isotope effect was calculated using equation (17) and the values of f and $C^{13}O_2/C^{12}O_2$ in Tables VI and VII. The isotope effect is given in the last column of Table VII.

TABLE IV

A Summary of the Experimental Details of the Decarboxylations of 4-Methylsalicylic Acid

| RUN | SOLVENT | TEMP. (°C) | REACTION TIME (MIN) | mg. OF ACID | MMOLES OF ACID | MMOLES OF $CO_2$ | f |
|---|---|---|---|---|---|---|---|
| Q-1 | Quinoline | 207.7 | 540 | 33.47 | - | .2142 | - |
| Q-2 | Quinoline | 207.5 | 540 | 34.30 | 0.2256 | .2236 | .9911 |
| Q-3 | Quinoline | 207.7 | 9 | 367.33 | 2.4157 | .4937 | .2044 |
| Q-4 | Quinoline | 195.7 | 8 | 374.80 | 2.4648 | .2300 | .09331 |
| Q-5 | Quinoline | 195.7 | 8 | 359.32 | 2.3630 | .1973 | .08350 |
| Q-6 | .02 M quinoline in nitrobenzene | 194.3 | 8 | 397.08 | 2.6113 | .2184 | .08364 |
| Q-7 | .02 M quinoline in nitrobenzene | 193.3 | 8 | 382.24 | 2.5137 | .1844 | .07336 |

TABLE V

A Summary of the Data Used in Calculating the Kinetic Isotope Effect in the Decarboxylations of 4-Methylsalicylic Acid

| RUN | log(1 - f) | $C^{13}O_2/C^{12}O_2$ | $R_{xf}/R_o$ | $\log(1 - fR_{xf}/R_o)$ | $100(k/k^* - 1)$ |
|---|---|---|---|---|---|
| Q-1 | - | .010685 | - | - | - |
| Q-2 | - | .010692 | - | - | - |
| Q-3 | -.09931 | .010487 | .98110 | -.09718 | 2.19 |
| Q-4 | -.04254 | .010467 | .97923 | -.04161 | 2.24 |
| Q-5 | -.03787 | .010471 | .97961 | -.03706 | 2.19 |
| Q-6 | -.03793 | .010618 | .99336 | -.03767 | 0.69 |
| Q-7 | -.03309 | .010612 | .99280 | -.03284 | 0.76 |

Note: $R_o$ taken as .010689

TABLE VI

A Summary of the Experimental Details of the Decarboxylations of 4-Methoxyanthranilic Acid

| RUN | BUFFER | VOL. (ml) | pH | REACTION TIME (MIN) | mg. OF ACID | MMOLES OF ACID | MMOLES OF $CO_2$ | f |
|---|---|---|---|---|---|---|---|---|
| W-2 | HCl-KCl | 100 | 1.3 | 2250 | 37.78 | 0.2261 | .2214 | .9792 |
| W-3 | HCl-KCl | 100 | 1.3 | 2355 | 40.51 | 0.2425 | .2378 | .9806 |
| W-9 | 2 N HCl | 200 | -.3 | 80 | 349.32 | 2.0487 | .1641 | .08010 |
| W-10 | 2 N HCl | 200 | -.3 | 87 | 350.18 | 2.0538 | .1870 | .09105 |
| W-5 | HCl-KCl | 200 | 1.3 | 20 | 350.07 | 2.0531 | .2458 | .1197 |
| W-6 | HCl-KCl | 200 | 1.3 | 18 | 350.24 | 2.0541 | .2071 | .1008 |
| W-14 | HCl-KCl | 225 | 1.3 | 17 | 354.24 | 2.0776 | .1898 | .09136 |
| W-12 | Formate | 400 | 4.0 | 85 | 322.41 | 1.8909 | .1544 | .08165 |
| W-13 | Formate | 425 | 4.0 | 90 | 318.48 | 1.8679 | .1579 | .08453 |

TABLE VII

A Summary of the Data Used in Calculating the Kinetic Isotope Effect in the Decarboxylations of 4-Methoxyanthranilic Acid

| RUN | log(1 - f) | $C^{13}O_2/C^{12}O_2$ | $R_{xf}/R_o$ | $\log(1 - fR_{xf}/R_o)$ | $100(k/k^* - 1)$ |
|---|---|---|---|---|---|
| W-2 | - | .010825 | - | - | - |
| W-3 | - | .010825 | - | - | - |
| W-9 | -.03626 | .010412 | .96185 | -.03482 | 4.14 |
| W-10 | -.04146 | .010409 | .96157 | -.03979 | 4.20 |
| W-5 | -.05537 | .010680 | .98661 | -.05458 | 1.45 |
| W-6 | -.04614 | .010694 | .98790 | -.04556 | 1.27 |
| W-14 | -.04161 | .010672 | .98587 | -.04100 | 1.49 |
| W-12 | -.03699 | .010804 | .99806 | -.03692 | 0.19 |
| W-13 | -.03835 | .010798 | .99751 | -.03826 | 0.24 |

Note: $R_o$ taken as .010825

## DISCUSSION

### (A) Decarboxylation of 4-Methylsalicylic Acid

The kinetic isotope effects in Table V are summarized below.

| SOLVENT | TEMP.(°C) | 100(k/k* - 1) |
|---|---|---|
| Quinoline | 196 | 2.2 ± .1 |
| .02 M quinoline in nitrobenzene | 194 | 0.7 ± .1 |

It is clear that the magnitude of the isotope effect increases with increasing quinoline concentration. As this agrees with the predictions made by Bourns' mechanism, it would appear that the reaction proceeds via the equilibrium formation of ion pairs, followed by the formation of a reaction intermediate, which can decompose to either products or to an ion pair.

### (B) Decarboxylation of 4-Methoxyanthranilic Acid

The kinetic isotope effects in Table VII are summarized below.

| pH | TEMP.(°C) | 100(k/k* - 1) |
|---|---|---|
| -.3 | 60 | 4.2 ± .1 |
| 1.3 | 60 | 1.4 ± .1 |
| 4.0 | 60 | 0.2 ± .1 |

The value of 0.2 ± .1 is too small to be considered as an isotope effect. Thus, a large effect was found at low pH, and no isotope effect was found at high pH. As mechanism (23) required an isotope effect at both low and high pH, it cannot be the operative mechanism in this reaction. Therefore, the reaction proceeds via mechanism (25), in which both of HA and Z are protonated to form $H_2A^*$ and HZ*, but neither $H_2A^*$ nor HZ* decarboxylates directly.

# BIBLIOGRAPHY

( 1) O. Beeck, J. W. Otvos, D. P. Stevenson and C. D. Wagner, J. Chem. Phys., 16, 255 (1948).

( 2) J. Bigeleisen and M. Goeppert Mayer, J. Chem. Phys., 15, 261 (1947).

( 3) J. Bigeleisen, J. Chem. Phys., 17, 675 (1949).

( 4) J. Bigeleisen and L. Friedman, J. Chem. Phys., 17, 998 (1949).

( 5) J. Bigeleisen and T. L. Allen, J. Chem. Phys., 19, 760 (1951).

( 6) J. Bigeleisen and M. Wolfsberg, Advan. Chem. Phy., I, 15 (1958).

( 7) N. Bjerrum, Z. Physik. Chem., 104, 147 (1923).

( 8) A. A. Bothner-By and J. Bigeleisen, J. Chem. Phys., 19, 755 (1951).

( 9) A. N. Bourns, Private Communication.

(10) A. M. Downes and G. M. Harris, J. Chem. Phys., 20, 196 (1952).

(11) G. E. Dunn, P. Legatte and I. E. Scheffler, Can. J. Chem., 43, 3080 (1965).

(12) M. G. Evans and M. Polanyi, Trans. Faraday Soc., 31, 875 (1935), 33, 448 (1937): M. Polanyi, J. Chem. Soc., 629 (1937).

(13) H. Eyring, J. Chem. Phys., 3, 107 (1935).

(14) A. Fry and M. Calvin, J. Phys. Chem., 56, 901 (1952).

(15) S. Glasstone, K. J. Laidler and H. Eyring, "The Theory of Rate Processes", McGraw-Hill, New York (1941).

(16) M. J. Goldstein and G. L. Thayer Jr., J. Am. Chem. Soc., 87, 1925 (1965).

(17) M. J. Goldstein and G. L. Thayer Jr., J. Am. Chem. Soc., 87, 1933 (1965).

(18) E. C. M. Grigg, Australian J. Chem., 9, 252 (1956).

(19) E. Grovenstein Jr. and G. A. Ropp, J. Am. Chem. Soc., 78, 2560 (1956).

(20) J. O. Hirschfelder and E. P. Wigner, J. Chem. Phys., 7, 616 (1939).

(21) P. Legatte and G. E. Dunn, Can. J. Chem., 43, 1158 (1965).

(22) J. G. Lindsay, A. N. Bourns and H. G. Thode, Can. J. Chem., 29, 192 (1951).

(23) J. G. Lindsay, A. N. Bourns and H. G. Thode, Can. J. Chem., 30, 163 (1952).

(24) K. R. Lynn and A. N. Bourns, Chem. Ind., 782 (1963).

(25) L. Melander, "Isotope Effects on Reaction Rates", Ronald Press, New York (1960).

(26) O. Metzler and O. Jeger, Helv. Chim. Acta, 30, 675 (1947).

(27) K. J. Pedersen, J. Am. Chem. Soc., 58, 240 (1936).

(28) P. Riesz and J. Bigeleisen, J. Am. Chem. Soc., 81, 6187 (1959).

(29) W. Rodewald, Ph.D. Thesis, University of Manitoba (1964).

(30) A. Roe and M. Hellmann, J. Chem. Phys., 19, 660 (1951).

(31) G. A. Ropp and V. F. Raaen, J. Am. Chem. Soc., 74, 4992 (1952).

(32) W. M. Schubert, J. Am. Chem. Soc., 71, 2639 (1949).

(33) W. H. Stevens, J. M. Pepper and M. Lounsbury, J. Chem. Phys., 20, 192 (1952).

(34) W. H. Stevens, J. M. Pepper and M. Lounsbury, Can. J. Chem., 30, 529 (1952).

(35) F. H. Verhoek, J. Am. Chem. Soc., 56, 571 (1934).

(36) F. H. Verhoek, J. Am. Chem. Soc., 61, 186 (1939).

(37) E. R. Washburn and H. C. Urey, Proc. Nat. Acad. Sci. U. S., 18, 496 (1932).

(38) A. V. Willi, Z. Naturforsch., 13A, 997 (1958).

(39) A. V. Willi, Trans. Faraday Soc., 55, 433 (1959).

(40) M. Wolfsberg and M. J. Stern, Pure Appl. Chem., 8, 225 (1964).

(41) A. Wood, Trans. Faraday Soc., 60, 1263 (1964).

(42) P. E. Yankwich and M. Calvin, J. Chem. Phys., 17, 109 (1949).

(43) P. E. Yankwich, E. C. Stivers and R. F. Nystrom, J. Chem. Phys., 20, 344 (1952).

(44) P. E. Yankwich and E. C. Stivers, J. Chem. Phys., 21, 61 (1953).

(45) P. E. Yankwich, R. L. Belford and G. Fraenkel, J. Am. Chem. Soc., 75, 832 (1953).

(46) P. E. Yankwich and R. L. Belford, J. Am. Chem. Soc., 75, 4178 (1953).

(47) P. E. Yankwich and R. L. Belford, J. Am. Chem. Soc., 76, 3067 (1954).

(48) P. E. Yankwich and A. L. Promislow, J. Am. Chem. Soc., 76, 4648 (1954).

(49) P. E. Yankwich, A. L. Promislow and R. F. Nystrom, J. Am. Chem. Soc., 76, 5893 (1954).

(50) P. E. Yankwich and H. S. Weber, J. Am. Chem. Soc., 77, 4513 (1955).

(51) P. E. Yankwich and H. S. Weber, J. Am. Chem. Soc., 78, 564 (1956).

(52) P. E. Yankwich and R. M. Ikeda, J. Am. Chem. Soc., 81, 5054 (1959).

(53) P. E. Yankwich and R. M. Ikeda, J. Am. Chem. Soc., 82, 1891 (1960).

(54) M. Zielinski, J. Chem. Phys., 41, 3646 (1964).

(55) I. Zlotowski and M. Zielinski, Nukleonika, 6, 511 (1961).

(56) H. Zollinger, Helv. Chim. Acta, 38, 1597, 1617 (1955): E. Grovenstein Jr. and N. S. Aprahamian, J. Am. Chem. Soc., 84, 212 (1962).