behave differently, the unbracketed "complex" is probably unstable, while the liberated Cu^+ ion is likely to undergo rapid oxidation to Cu^{++}.

Summary

1. The autoxidation rate of *l*-ascorbic acid in the presence of copper (2×10^{-5} mole/liter) is investigated from *p*H 2.59 to 9.31.

2. The results agree with the assumption that in the primary reaction, one mole of dehydroascorbic acid and of hydrogen peroxide is formed from one mole of ascorbic acid and oxygen.

3. The rate is independent of the ascorbic acid concentration, and, as shown in the preceding paper of the series, proportional to the oxygen concentration.

4. At very low copper concentrations, the rate increases faster than the concentration of the metal. At higher concentrations of the latter, the rate increase becomes proportional to the increase in the copper concentration.

5. The dependence of the rate on *p*H in the presence of copper has the appearance of being complex. However, analysis shows that neither the neutral acid nor the divalent ion, but *only the monovalent ion of l-ascorbic acid is the substrate of the copper catalysis*, and that the apparent irregularity in the dependence of the rate on *p*H is caused by complex formation of some of the buffers with the copper catalyst. A concentration of 2×10^{-5} mole/liter of copper, about 4 mg. of $Cu(NO_3)_2$ per liter, increases the reaction rate of the monovalent ascorbate ion by a factor of 10^4.

6. No corresponding effect of silver was found at *p*H 5.12.

7. The mechanism of the catalysis is discussed briefly.

ROCHESTER, NEW YORK RECEIVED FEBRUARY 1, 1944

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE UNIVERSITY OF CALIFORNIA AT LOS ANGELES]

The Reaction of Dibromides of Mono-substituted Ethylenes with Potassium Iodide

BY DAVID PRESSMAN AND WILLIAM G. YOUNG

Iodide ion reacts with the dibromide of an olefinic compound to yield the original unsaturated compound, bromide ion and triiodide ion according to equation 1.[1]

$$RCHBrCH_2Br + 3I^- \longrightarrow 2Br^- + I_3^- + RHC = CH_2 \quad (1)$$

The reaction proceeds first order with respect to the dibromide concentration and also with respect to the iodide ion concentration in 99% methanol solution and is practically independent of the triiodide ion concentration.

We have shown that in the reactions of dibromides of *cis* and *trans* olefins with potassium iodide the reaction is slower for the dibromide from the *cis* olefin[2] and that the olefin produced is in its original stereoform in the case of the 2-butene dibromides.[3] The reaction takes place presumably through the attack by an iodide ion to remove a positive bromine atom as iodine bromide while, simultaneously, the electron pair which is left unshared by this removal attacks the carbon face opposite the remaining bromine atom forming a double bond and liberating a negative bromide ion.[3]

Our purpose in this research was to determine the reactivity of various mono-substituted ethylene dibromides with iodide ion. The reactions were studied in 99% methanol at two or more temperatures.

Experimental

Rates of Reaction.—The rate constants were obtained graphically by the use of equation 2

$$d \log_{10} \frac{1 - 3b\varphi/a}{1 - \varphi} \Big/ dt = \frac{a - 3b}{2.303} k_{obs.} \quad (2)$$

where a and b are the initial iodide and dibromide concentrations respectively, φ is the fraction of dibromide reacted and t is the time in hours. The following corrections were made as discussed previously[2]: (a) correction to standard temperature of 45.00, 60.00, 75.00, 80.00 or 85.00°, (b) correction for activity of solvent (methanol preparations I, II, III are the same as those used previously[2]), (c) correction for salt effect, and (d) correction for solvent expansion.[4]

Preparation of Materials

1-Pentene dibromide was prepared by mixing slowly at 0° bromine with the 1-pentene obtained from ethylmagnesium bromide and allyl bromide. The product was fractionated through a Weston column and the fraction boiling at 84.5–85.5° at 15 mm. was used.

Acrylic Acid Dibromide.—The Eastman Kodak Co. product melting at 59–61° was used.

Styrene dibromide was prepared from styrene and bromine in carbon disulfide. The solvent was evaporated and the product recrystallized from alcohol; m. p. 74.0–74.5°.

Allylbenzene dibromide was prepared by brominating, in carbon tetrachloride, allylbenzene obtained from allyl bromide and phenylmagnesium bromide. The product was fractionated and the fraction boiling at 126° at 2 mm. was used.

Allyl Alcohol Dibromide.—The Eastman Kodak Co. product was refractionated and the fraction boiling 113–114° at 13 mm. was used.

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[1] R. T. Dillon, THIS JOURNAL, **54**, 952 (1932).

[2] W. G. Young, D. Pressman and C. D. Coryell, *ibid.*, **61**, 1640 (1939).

[3] S. Winstein, D. Pressman and W. G. Young, *ibid.*, **61**, 1645 (1939).

[4] Corrections of 3.0, 4.5, 6.3, 7.6 and 8.1% were made at temperatures 45, 60, 75, 80 and 85°, respectively; (see ref. 2) Landolt–Börnstein, "Tabellen," Verlag von Julius Springer, Berlin, 1923, Vol. I, p. 278.

TABLE I

REACTION RATE CONSTANTS AND HEATS OF ACTIVATION FOR THE REACTION OF $RCHBrCH_2Br$ WITH POTASSIUM IODIDE

Parent compound	R	*k*, obs.	Temp., °C.	KI, moles/l., *a*	Dibromide moles/l., *b*	*k* cor.[d]	*k* cor., KI = 0.25	Av. *k* l./mole hr.	Deviation, %	$\Delta H^{\ddagger}$, KI = 0.25
Ethylene	H	1.34[a]	74.93	0.22	0.022	1.54		(1.54)[e]		23.2
		0.299[a]	59.72	.22	.027	0.344		(.344)[e]		
Propylene	CH_3	.0582[a]	74.93	.22	.024	.0669		(.0669)[e]		25.2
		.0113[a]	59.72	.22	.022	.0130		(.0130)[e]		
1-Butene	C_2H_5	.0794[a]	74.93	.23	.034	.0913		(.0913)[e]		25.8
		.0148[a]	59.72	.22		.0171		(.0171)[e]		
Acrylic acid	COOH	3.985[b]	85.38	.243	.023	4.26	4.28	4.27	0.2	20.9[f]
		3.985[b]	85.38	.248	.023	4.26	4.26			
		1.665[b]	74.91	.252	.021	1.825	1.822	1.825	.2	
		1.665[b]	74.90	.251	.022	1.830	1.828			
		0.417[b]	79.46	.256	.025	0.468	0.466	0.467	.1	
		.416[b]	79.46	.252	.021	.467	.467			
		.1021[b]	44.75	.238	.023	.1100	.1108	.1109	.1	
		.1028[b]	44.75	.241	.025	.1103	.1109			
Allyl alcohol	CH_2OH	.0978[b]	74.90	.245	.027	.1075	.1090	.1079	1.0	25.2
		.0972[b]	74.91	.254	.027	.1070	.1068			
		.01905[c]	59.64	.248	.028	.02079	.02081	.02107	1.2	
		.01955[c]	59.64	.249	.023	.02131	.02133			
Allylbenzene	CH_2	.0915[c]	74.95	.278	.022	.0983	.0967	.0978	1.0	24.6
		.0915[c]	74.95	.242	.021	.0983	.0988			
		.01834[c]	59.64	.251	.020	.02000	.01998	.01987	0.6	
		.01811[c]	59.64	.252	.022	.01978	.01976			
Bromoethylene	Br	.0557[b]	85.37	.310	.026	.0591	.0571	.0581	1.7	25.0
		.0558[b]	85.37	.251	.028	.0592	.0591			
		.01930[b]	74.90	.255	.029	.02121	.02115	.02134	0.9	
		.01945[b]	74.90	.242	.028	.02141	.02152			
1-Pentene	C_3H_7	.0900	74.55	.242	.023	.1007	.1012	.1006	.6	24.8
		.0896	74.60	.248	.022	.0999	.1000			
		.01837[c]	59.64	.240	.020	.02000	.02012	.02015	.2	
		.01850[c]	59.64	.248	.020	.02016	.02018			
Styrene	C_6H_5	4.05[b]	85.36	.239	.023	4.34	4.37	4.34	.7	21.7[f]
		4.03[b]	85.36	.246	.023	4.30	4.31			
		2.657	80.12	.2[illegible]8	.025	2.880	2.868	2.868		
		1.612	74.55	.244	.021	1.796	1.802	1.811	.5	
		1.648	74.59	.261	.025	1.831	1.820			
		0.389	59.48	.249	.024	0.429	0.429	0.436	1.6	
		.3925[b]	59.48	.211	.028	.433	.442			
		.0860[b]	44.75	.251	.021	.0925	.0924	.0931	0.8	
		.0862[b]	44.75	.231	.021	.0926	.0937			

[a] These data were reported by Dillon using methanol which gave rate constants 7% lower than those obtained with the synthetic product (I) used as a standard in this work. [b] Synthetic methanol II, rates 2% lower than in methanol I. [c] Synthetic methanol III, rates same as in methanol I. [d] The constants have been corrected for reactivity of solvent, solvent expansion and to the temperatures 45.00, 60.00, 75.00, 80.00 or 85.00°. [e] These calculations were made from data reported by Dillon. Correction has not been made for the potassium iodide effect. [f] This value was obtained from slope of the curve in Fig. 2.

Bromoethylene Dibromide.—The Eastman Kodak Co. product was refractionated and the fraction boiling at 86.0–87.0° at 18 mm. was used.

Experimental Results

The pertinent date for the compounds studied are in Table I. There are also listed the values of the heats of activation calculated by the Arrhenius equation

$$\Delta H^{\ddagger} = RT^2 \mathrm{d} \ln k_{\mathrm{obs.}}/\mathrm{d}t \qquad (3)$$

In Table II are given the observed relative values of the heats of activation $\Delta H^{\ddagger} - \Delta H_0^{\ddagger}$ and the entropies of activation $\Delta S^{\ddagger} - \Delta S_0^{\ddagger}$ (equation 4).[5]

$$\Delta S^{\ddagger} - \Delta S_0^{\ddagger} = \frac{\partial}{\partial T}\left(RT \ln \frac{k}{k_0}\right) \qquad (4)$$

$\Delta H_0^{\ddagger}$ and $\Delta S_0^{\ddagger}$ are, respectively, the heat and entropy of activation of the reference compound, ethylene dibromide.

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[5] See L. P. Hammett, "Physical Organic Chemistry," McGraw-Hill Book Co., New York, N. Y., 1940, Chapter IV.

TABLE II

RELATIVE ENTROPIES AND HEATS OF ACTIVATION FOR THE REACTION OF $RCHBrCH_2Br$ WITH POTASSIUM IODIDE

Parent compound	R	$k_{75°}$ l./mole hr.	$\Delta S^{\ddagger} - \Delta S_0^{\ddagger}$ cal./deg	$\Delta H^{\ddagger} - \Delta H_0^{\ddagger}$ kcal.
Ethylene	H	1.54	0	0
Propylene	CH_3	0.0669	−0.5	2.0
1-Butene	C_2H_5	.0913	1.8	2.6
1-Pentene	C_3H_7	.1006	−0.8	1.6
Allylbenzene	$CH_2C_6H_5$	.0978	−1.5	1.4
Styrene	C_6H_5	1.811	−4.0	−1.5
Bromoethylene	Br	0.02134	−3.3	1.8
Allyl alcohol	CH_2OH	.1079	0.4	2.0
Acrylic acid	COOH	1.825	−6.3	−2.3

The observed effect of various substituents, R, on the reaction of $RCHBrCH_2Br$ with iodide ion is to increase the heat of activation in the order $COOH < C_6H_5 < H < C_6H_5CH_2 < C_3H_7 < Br < CH_2OH, CH_3 < C_2H_5$ and to increase the entropy of activation in the order: $COOH < C_6H_5 < Br < C_6H_5CH_2 < C_3H_7 < CH_3 < H < CH_2OH < C_2H_5$.

Discussion

The functions $\Delta H^{\ddagger}$ and $\Delta S^{\ddagger}$ are in general relatively independent of temperature and are more reliable standards of comparison of reactivity than is the rate constant, since a sequence of order of reactivity as determined by the rate constant at any one temperature may be different at some other temperature. Empirically this occurs in the case of acrylic acid dibromide and styrene dibromide.[6] The rate constant of acrylic acid dibromide is larger than that of styrene dibromide at 45° but smaller at 85°.

However, in the reaction of potassium iodide with unsymmetrical dibromide such as styrene dibromide, even the observed heats of activation and the observed entropies of activation are not reliable bases of comparison of reactivity since they would be expected to be functions of temperature, aside from their general variation with temperature due to changes in the vibrational and rotational energy states with temperature. This arises from the fact that either bromine atom in an olefin dibromide molecule may be attacked by iodide ion and in the case of an unsymmetrical dibromide the two bromine atoms would be attacked at different rates. The rate of liberation of iodine is thus given by the equation

$$\begin{aligned} d\,(I_3^-)/dt &= (k_1 + k_2)(\text{Dibromide})(I^-) \\ &= k_{\text{obs.}}\ (\text{Dibromide})(I^-) \end{aligned} \tag{5}$$

where k_1 and k_2 are the rate constants for the attack of the individual bromine atoms by iodide ion and $k_{\text{obs.}}$ is the observed rate constant which is equal to the sum of k_1 and k_2.

The observed heat of activation $\Delta H^{\ddagger}$ is given then by equation 6.

$$d \ln (k_1 + k_2)/dT = \Delta H^{\ddagger}/RT^2 \tag{6}$$

Substituting for k_1 and k_2, their values as given in equations 7 and 8[7]

$$k_1 = \frac{kT}{h} e^{\Delta S_1^{\ddagger}/R} e^{-\Delta H_1^{\ddagger}/RT} \tag{7}$$

$$k_2 = \frac{kT}{h} e^{\Delta S_2^{\ddagger}/R} e^{-\Delta H_2^{\ddagger}/RT} \tag{8}$$

equation 9 is obtained.

$$d \ln \frac{kT}{h} \left(e^{\Delta S_1^{\ddagger}/R} e^{-\Delta H_1^{\ddagger}/RT} + e^{\Delta S_2^{\ddagger}/R} e^{-\Delta H_2^{\ddagger}/RT}\right) dT = \Delta H^{\ddagger}/RT^2 \tag{9}$$

Carrying out the indicated differentiation and neglecting the small term, $1/T$, gives equation 10.

$$\Delta H^{\ddagger} = \frac{\Delta H_1^{\ddagger} e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} e^{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})/RT} + \Delta H_2^{\ddagger}}{e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} e^{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})/RT} + 1} \tag{10}$$

As the temperature increases the apparent value $\Delta H^{\ddagger}$ approaches the value $(\Delta H_1^{\ddagger} e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} + \Delta H_2^{\ddagger})/(e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} + 1)$. This reduces to the average $(\Delta H_1^{\ddagger} + \Delta H_2^{\ddagger})/2$ if the entropy of activation is the same for the two reactions, *i. e.*, $\Delta S_1^{\ddagger} = \Delta S_2^{\ddagger}$. As the temperature decreases, the apparent value of $\Delta H^{\ddagger}$ approaches the smaller of the two values $\Delta H_1^{\ddagger}$ or $\Delta H_2^{\ddagger}$. We have adopted the convention that $\Delta H_1^{\ddagger}$ is the smaller.[8]

A more useful equation for discussion is obtained by subtracting $\Delta H_1^{\ddagger}$ from both sides of equation 10 to give equation 11.

$$\Delta H^{\ddagger} - \Delta H_1^{\ddagger} = \frac{\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}}{e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} e^{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})/RT} + 1} \tag{11}$$

This equation shows that the difference between the observed value $\Delta H^{\ddagger}$ and the heat of activation for the reaction of either of the bromine atoms is a function of the relative values $(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})$ and $(\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger})$ and of T and is independent of the absolute magnitude of $\Delta H^{\ddagger}$.

In Table III are given for several different assumed values of $\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}$ and of $\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger}$, the values at 75° of (a) $\Delta H^{\ddagger} - \Delta H_1^{\ddagger}$; (b) the rate of change of $\Delta H^{\ddagger}$ with temperature[9]; and (c) the fraction of the total reaction which is due to the bromine atom with the lower heat of activation,[10] $k_1/(k_1 + k_2)$. The assumed values of

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[6] Similar cases have been pointed out by T. L. Davis and R. Heggie, *J. Org. Chem.*, **2**, 470 (1937).

[7] H. Eyring, *J. Chem. Phys.*, **3**, 107 (1935).

[8] Similar to $\Delta H^{\ddagger}$, $\Delta S^{\ddagger}$ is a function of T, $\Delta S_1^{\ddagger}$, $\Delta S_2^{\ddagger}$, $\Delta H_1^{\ddagger}$ and $\Delta H_2^{\ddagger}$.

[9] The rate of change of the observed heat of activation with temperature is given by equation 12.

$$\frac{d\ \Delta H^{\ddagger}}{dT} = \frac{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})^2 e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} e^{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})/RT}}{RT^2(e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} e^{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})/RT} + 1)^2} \tag{12}$$

[10] This fraction is given by equation 13.

$$\frac{k_1}{k_1 + k_2} = \frac{e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} e^{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})/RT}}{1 = e^{(\Delta S_1^{\ddagger} - \Delta S_2^{\ddagger})/R} e^{(\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger})/RT}} \tag{13}$$

TABLE III

APPARENT HEAT OF ACTIVATION, ITS RATE OF INCREASE WITH TEMPERATURE AND THE FRACTION OF THE REACTION WHICH IS DUE TO THE BROMINE ATOM WITH THE LOWER HEAT OF ACTIVATION AS FUNCTIONS OF $\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}$ AND $\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger}$ AT 75°

	Value of $\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger}$ (cal./deg.) (assumed)		
$\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}$, cal. (assumed)	−4.6	0	4.6
	$\Delta H^{\ddagger} - \Delta H_1^{\ddagger}$ (cal.)		
700	25	260	550
1400	19	189	800
2100	10.4	100	700
2800	5.4	53	450
3500	2.3	23	220
	$d\Delta H^{\ddagger}/dT$ (cal./deg.)		
700	0.069	0.75	0.33
1400	.104	1.08	1.93
2100	.094	0.81	4.00
2800	.062	.60	4.30
3500	.032	.32	2.95
	$k_1/(k_1 + k_2)$		
700	0.96	0.73	0.21
1400	.99	.87	.43
2100	1.00	.95	.67
2800	1.00	.98	.84
3500	1.00	.99	.94

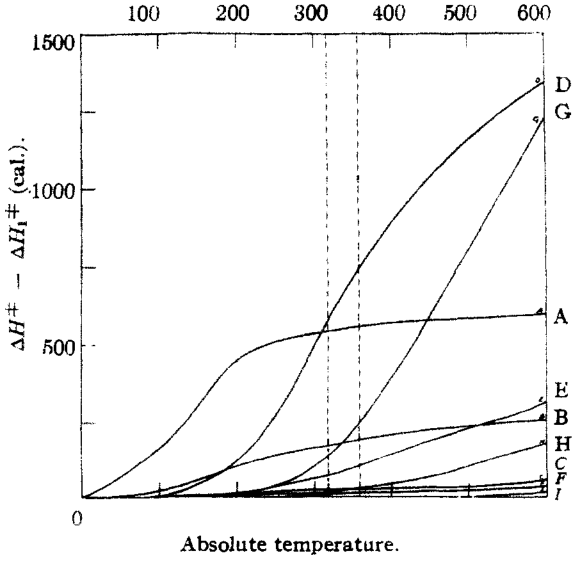


Fig. 1.—Plot of values of $\Delta H^{\ddagger} - H_1^{\ddagger}$ against absolute temperature for various assumed values of $\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}$ and $\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger}$:

Curve	$\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}$, cal.	$\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger}$, cal.
A	700	4.6
B	700	0
C	700	−4.6
D	2100	4.6
E	2100	0
F	2100	−4.6
G	3500	4.6
H	3500	0
I	3500	4.6

$\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}$ and $\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger}$ used are of the same magnitude as was observed in changing substituents in symmetrically substituted ethylene dibromides.

Figure 1 shows plots of the apparent heat of activation relative to the lower of the individual values, $\Delta H_1^{\ddagger}$, as a function of temperature as calculated from equation 11 for several different assumed values of $\Delta H_2^{\ddagger} - \Delta H_1^{\ddagger}$ and of $\Delta S_2^{\ddagger} - \Delta S_1^{\ddagger}$. The temperature region in which our experiments were carried out is marked by dotted lines.

The calculated values in Table III and the plots of Fig. 1 show that the observed value, $\Delta H^{\ddagger}$, may be quite different from either of the values $\Delta H_1^{\ddagger}$ and $\Delta H_2^{\ddagger}$ which must be known before any quantitative theoretical consideration can be given to a discussion of the effect of structure on the reaction.

The calculated values of the rate of change of $\Delta H^{\ddagger}$ with temperature indicate that such a change would probably not be observed within the accuracy of our experiments. Experimentally, the heats of activation of the reactions of styrene dibromide and of acrylic acid dibromide appear to be constant over a range of 40° as is shown by the straight line character of the plots of the logarithms of the reaction rate constants against the reciprocals of the absolute temperatures (Fig. 2).

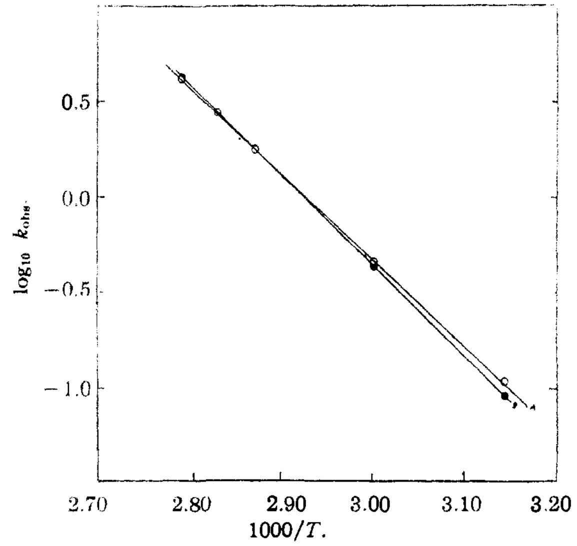


Fig. 2.—Plot of $\log_{10} k$ against reciprocal absolute temperature for acrylic acid dibromide ●, and for styrene dibromide ○.

The authors are indebted to Dr. C. D. Coryell for assistance in the calculation of the reaction rate constants.

Summary

The reaction rate constants and the heats and entropies of activation were determined for the

reaction in 99% methanol of potassium iodide with monosubstituted ethylene dibromides, $RCHBrCH_2Br$ where R was C_3H_7, C_6H_5, $C_6H_5CH_2$, CH_2OH, COOH and Br.

It is shown that, since each of the bromine atoms can react and at a different rate and with a different heat of activation, no theoretical significance can be given to the effect of various substituents, R, on the observed values of the heats of activation and entropies of activation of the reaction of $RCHBrCH_2Br$ with potassium iodide.

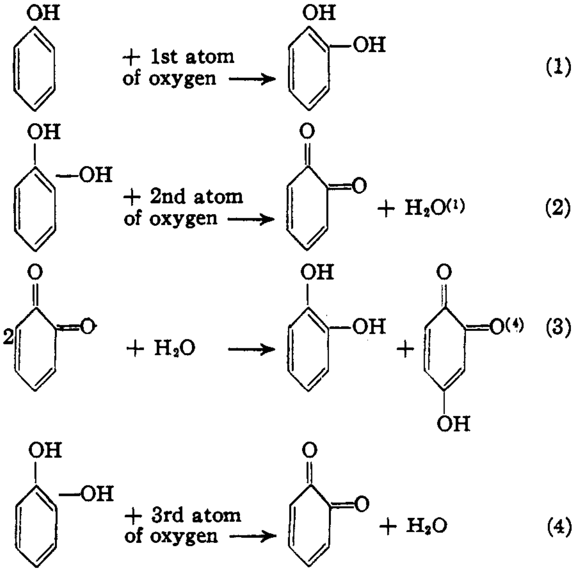
LOS ANGELES, CALIFORNIA RECEIVED JANUARY 18, 1944

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, COLUMBIA UNIVERSITY]

The Activity of Tyrosinase toward Phenol

BY RALPH C. BEHM AND J. M. NELSON

As shown by several investigators[1,2,3] the phenol oxidase, tyrosinase, possesses two essentially different enzymatic activities. It not only catalyzes the aerobic oxidation of many *o*-dihydric phenols to their corresponding *o*-quinones, but also catalyzes the introduction of a second hydroxyl group into several monohydric phenols, *ortho* to the hydroxyl group already present. These oxidation reactions are not simple, but involve several consecutive and concurrent reactions, especially when monohydric phenols constitute the substrate. Taking phenol and catechol as representing the two types of phenols, the final oxidation product in both instances is *p*-hydroxyquinone.[4] In the enzymatic oxidation of phenol at *p*H 6, 3 atoms of oxygen[5] are consumed per molecule of phenol oxidized.



The method usually used for estimating the activity of tyrosinase toward a monohydric phenol is based on determining the rate of oxygen uptake by means of a Warburg or Barcroft respirometer.[6] It is apparent, however, from what has been stated above, that only the first atom of oxygen is concerned with the enzyme's action on the monohydric phenol. The remaining two atoms of oxygen are consumed in the enzyme's action on the *o*-dihydric phenol. The question therefore arises: Can the rate of oxygen uptake serve as a true measure of the enzyme's activity toward a monohydric phenol?

To answer the latter question, it was decided to determine, instead of the rate of oxygen uptake, the rate of disappearance of the monohydric phenol from the reaction mixture. Such a method would be independent of all the accompanying reactions encountered in the oxygen uptake method. For the purpose a reaction mixture, consisting of the enzyme, phenol, buffer and water, was placed in a reaction vessel and air bubbled through the mixture. Samples were removed at regular time intervals and their phenol contents determined. The results obtained are shown as Curve I. Curve II represents the rate of oxygen uptake for a reaction mixture, identical in composition with the one used in following the rate of phenol disappearance. It will be observed that the two curves coincide within experimental error, showing that, at least when phenol is the monohydric phenol being oxidized by means of the enzyme, the concurrent reactions, equations (2), (3) and (4) must proceed at rates at least as great as the rate of the initial oxidation of the phenol (equation 1) and that, therefore, the rate of oxygen uptake can serve as a true measure of the enzyme's activity toward phenol.

Tyrosine preparations, depending on the method of preparation, often vary widely with respect to their relative activities toward monohydric and *o*-dihydric phenols. Therefore, it has become customary, in these Laboratories, to designate a preparation low in activity toward *p*-cresol compared to its activity toward catechol as a high catecholase preparation and a preparation high in *p*-cresolase activity as a high cresolase preparation.[7] Since in the present study phenol was used instead of *p*-cresol, it should be men-

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(1) Pugh and Roper, *Biochem. J.*, **21**, 1370 (1927).

(2) Dalton and Nelson, THIS JOURNAL, **61**, 2946 (1939).

(3) Bordner and Nelson, *ibid.*, **61**, 1507 (1939).

(4) Wagreich and Nelson, *ibid.*, **60**, 1545 (1938).

(5) Unpublished results obtained in these Laboratories.

(6) Adams and Nelson, THIS JOURNAL, **60**, 2472 (1938).

(7) Parkinson and Nelson, *ibid.*, **62**, 1693 (1940).