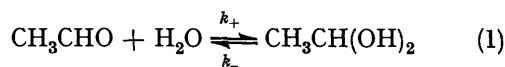


Metal-ion Assisted Catalysis of Nucleophilic Attack. Part 3.† Co-operation between Metal Ion and Hydroxide

By Paul Woolley, Magdalene College, Cambridge

A rapid path for the hydration of acetaldehyde is demonstrated, in which a zinc complex and the hydroxide ion co-operate as catalysts. In the mechanism proposed a zinc-bound hydroxide ion attacks directly the carbonyl group of the aldehyde. Comparison of the nucleophilic strengths of hydroxide ions in various complexes demonstrates a correlation similar to that obtained by other workers for attack on propionic anhydride and on carbon dioxide. The enzyme carbonic anhydrase behaves consistently with this correlation for the substrate acetaldehyde, but not for the substrate carbon dioxide.

ZINC ions catalyse weakly the hydration of acetaldehyde¹ [equation (1)]. This catalysis is enhanced by

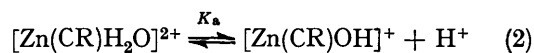


other species in solution, notably acetate,² pyridine,³ and hydroxide.² Kinetic analysis has shown³ that acetate or pyridine can modify the rate of the zinc-catalysed reaction in two ways: (i) when it is bound to the metal ion the base blocks some of the metal's catalytic sites, suppressing the catalysis; (ii) when it is not bound to the metal ion the base produces, together with the metal, an enhanced ('co-operative') catalysis, which is greater than that due to base or metal ion alone.

These effects are clearly illustrated by [pyridine-2-carboxylatozinc]⁺ (Znpc⁺) (1) and [(pyridine-2-carb-aldehyde oximato)zinc]⁺ (Znpyox⁺) (2).⁴ Complex (1) contains two basic groups, both co-ordinated to the zinc

ion, while (2) contains a free basic group, -O⁻. The order of catalytic activity is (2) ≫ [Zn(OH₂)₆]²⁺ > (1), confirming the general picture but yielding no further mechanistic information.

Detailed study of the co-operative catalysis involving Zn²⁺ and OH⁻ is prevented by polynucleation and precipitation of zinc hydroxide above neutral pH values.⁵ A more amenable system was therefore sought. Zinc complexes containing the macrocyclic ligands (3)–(5) are stable in aqueous solution over the required pH range and show a single hydrolytic deprotonation [equation (2)], with a pK_a in the region 8–9. Such complexes were investigated as potential catalysts for acetaldehyde hydration. A summary of their properties is included in ref. 6.



³ P. R. Woolley, *J.C.S. Dalton*, 1975, 1570.

⁴ P. Woolley, *J.C.S. Chem. Comm.*, 1975, 579.

⁵ B. Pokric and Z. Pucar, *J. Inorg. Nuclear Chem.*, 1971, **33**, 445.

⁶ P. Woolley, *Nature*, 1975, **253**, 677.

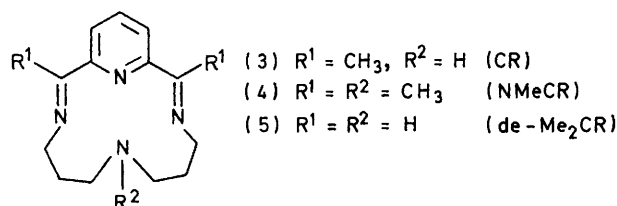
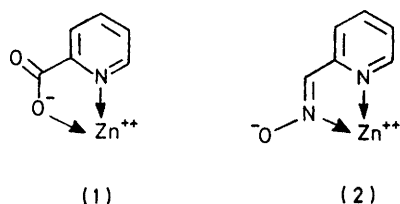
† Part 3, ref. 3.

¹ Y. Pocker and J. E. Meany, *J. Phys. Chem.*, 1967, **71**, 3113.

² R. H. Prince and P. R. Woolley, *J.C.S. Dalton*, 1972, 1548.

EXPERIMENTAL

The rate of hydration of acetaldehyde was measured by the spectrophotometric method already described.^{3,3} The high u.v. absorbance of the CR complexes made an optical



path of 1 mm necessary. The observation wavelength in the presence of ZnCR-type complexes was 262 nm, where the complex has an extinction minimum. Buffers were prepared in the usual way,³ but to avoid precipitation no 'inert' salt was added. Thus the ionic strength was uncontrolled: it lay normally between 5 and 25mM, and never exceeded 50mM.

Materials purchased were of analytical grade and deionised distilled water was used. Macrocyclic complexes were prepared according to ref. 7. In experiments involving cobalt(II) complexes precautions were taken to exclude oxygen.

The electronic spectra of the metal-complex catalysts were checked after the hydration reaction, to ascertain that the catalyst had not changed during the reaction.

The rate parameter obtained from each kinetic run is k_{obs} [equation (3)*]. k_+ and k_- were obtained when

$$k_{\text{obs}} = k_+ + k_- = \frac{-1}{([\text{MeCHO}] - [\text{MeCHO}]_{\infty})} \frac{d[\text{MeCHO}]}{dt} \quad (3)$$

necessary using the equilibrium constant⁸ for the hydration: $K_{\text{eq}} = k_+/k_-$. No significant deviation from first-order kinetics with respect to acetaldehyde was observed.

RESULTS

The general rate law for the catalysed reaction is given by equation (4), if water is in great excess and if the various

$$k_{\text{obs}} = k_0 + \sum_i k_i [i] \quad (4)$$

catalysts i in solution act independently. k_0 , the rate constant of the uncatalysed reaction, is small (ca. 0.001 s⁻¹ at 0 °C). k_i is termed the catalytic activity of catalyst i . If co-operative catalysis is present, further terms such as $k_{jk}[j][k]$ must be added.

(a) *Aquated Zn²⁺*.— k_{obs} Plotted against the total concentration of zinc ions $[\text{Zn}^{2+}]_0$ yields in the absence of complexing agents a straight line² whose gradient is pH-

* This equation was given incorrectly in refs. 3 and 4. It is derived by applying first-order kinetics to equation (1).

dependent. This pH dependence implies co-operation between zinc and hydroxide ions, and should be expressed by the derivative of equation (4). The approximation

$$\frac{\partial k_{\text{obs}}}{\partial [\text{Zn}^{2+}]_0} \approx \frac{\partial k_{\text{obs}}}{\partial [\text{Zn}^{2+}]} = k_{\text{Zn}} + k_{\text{Zn,OH}}[\text{OH}^-] \quad (5)$$

$[\text{Zn}^{2+}]_0 \approx [\text{Zn}^{2+}]$ is possible as the equilibrium $\text{Zn}_{\text{aq}}^{2+} \rightleftharpoons \text{ZnOH}^+ + \text{H}^+$ lies over to the left at the pH values where measurement is made.

Figure 1 shows that equation (5) is fairly well verified. k_{Zn} is 0.011 l mol⁻¹ s⁻¹ and $k_{\text{Zn,OH}}$ is 6.3×10^5 l² mol⁻² s⁻¹. The accessible pH range reveals only the onset of a large effect.

(b) *ZnCR²⁺ and Related Complexes*.—The catalytic effect of ZnCR²⁺ is summarised in Figures 2 and 3. A small but significant activity is observed at low pH and a high activity at high pH. The sigmoidal rate curve in Figure 3 (point of inflection at pH 9.07) follows within experimental error the titration curve of ZnCR²⁺ at 0 °C (pK_a 9.17⁶). This curve can be represented empirically by an equation analogous to (5), with ZnCR replacing Zn, or as the sum of separate catalytic contributions from $[\text{ZnCR}]^{2+}$ and $[\text{Zn}(\text{CR})\text{OH}]^+$; the two representations are kinetically equivalent. Using the latter representation, the values $\partial k_{\text{obs}}/\partial [\text{ZnCR}]^{2+}$ and $\partial k_{\text{obs}}/\partial [\text{Zn}(\text{CR})\text{OH}]^+$ (the activities of ZnCR²⁺ at extremes of low and high pH respectively) were obtained from the best fit to the sigmoid curve.

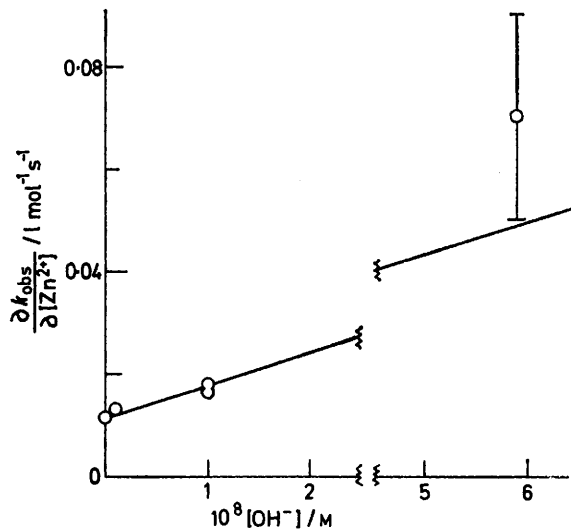


FIGURE 1 Onset of co-operative catalysis by Zn²⁺ and OH⁻ in acetaldehyde hydration: error bars on the left are too small to appear in the Figure. The line represents best fit by a weighted least-squares procedure. Measurement was at 0 °C and 1.4M (sodium perchlorate). The four points on the left were obtained by extrapolation to zero buffer concentration, the point on the right by using a non-interfering buffer (collidine) at low concentration (10mM). The OH⁻ concentration was inferred from the pH using the expression $[\text{OH}^-] = 10^{(\text{pH} - 14)}$

For comparison, the activities of the complexes $[\text{Zn}(\text{NMeCR})]^{2+}$ and $[\text{Zn}(\text{de-Me}_2\text{CR})]^{2+}$ were measured at high pH; using their known pK_a values and neglecting their low pH activity, it was possible to calculate the high pH

⁷ R. H. Prince, D. A. Stotter, and P. R. Woolley, *Inorg. Chim. Acta*, 1974, 9, 51.

⁸ R. P. Bell and J. C. Clunie, *Trans. Faraday Soc.*, 1952, 48, 439.

limits of these activities, $\partial k_{\text{obs}}/\partial[\text{Zn}(\text{NMeCR})\text{OH}]^+$ and $\partial k_{\text{obs}}/\partial[\text{Zn}(\text{de-Me}_2\text{CR})\text{OH}]^+$.

The activity of $[\text{CuCR}]^{2+}$ was insignificantly different from zero at pH 5.0 and 7.5; the onset of activity was observed at pH 9.5. $[\text{NiCR}]^{2+}$ and $[\text{CoCR}]^{2+}$ underwent a side-reaction with acetaldehyde, so that their activities could not be measured.

The activation energy of the ZnCR^{2+} -catalysed hydration at high pH was measured. This was achieved by taking measurements at pH 8.5 and 10 and converting them to the high-pH values using the known $\text{p}K_a$ of ZnCR^{2+} . The Arrhenius plot (Figure 4) yields an activation energy of 6.3 kcal mol⁻¹.^{*} Figure 4 refers to the forward reaction [k_+ in equation (1)].

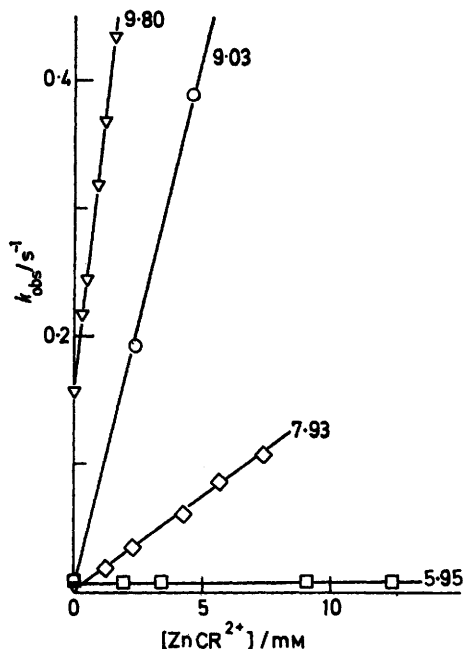


FIGURE 2 Typical plots of k_{obs} against ZnCR^{2+} concentration at different pH values. Buffer concentrations were 10mM (acetate, triethanolamine, or borate, according to pH range). Temperature, 0 °C; $I \lesssim 0.025\text{M}$

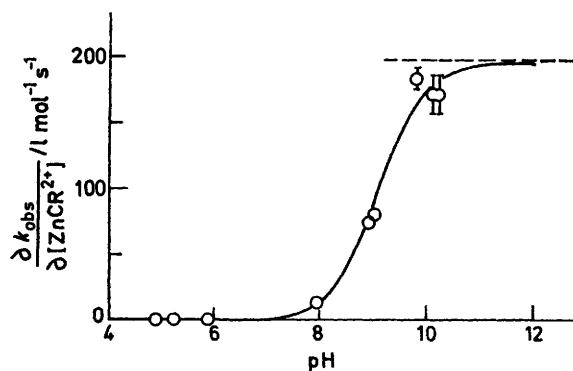


FIGURE 3 Gradients from Figure 2 and similar experiments plotted against pH. The error increases at high pH owing to the increasingly rapid parallel catalysis by OH^- . The broken line is the high-pH value inferred from the known $\text{p}K_a$ of ZnCR^{2+} (9.17 at 0 °C and low I)

The kinetic results are summarized in Table 1 and the catalytic activities obtained from them, together with the

* 1 cal \equiv 4.18 J.

activities of some other catalysts for comparison, are displayed in Table 2.

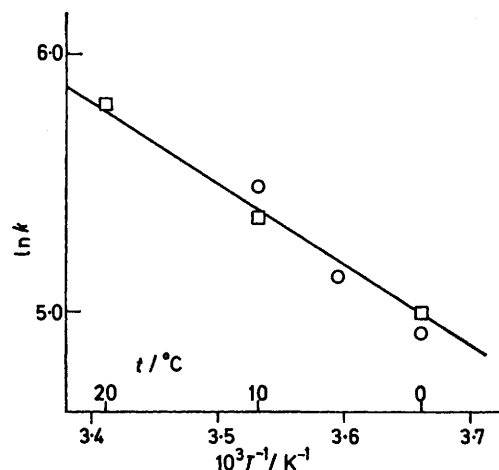


FIGURE 4 Arrhenius plot to find the activation energy corresponding to the $[\text{Zn}(\text{CR})\text{OH}]^+$ term in acetaldehyde hydration. k is rate of the catalysed forward reaction in $\text{l mol}^{-1}\text{s}^{-1}$, given by $\partial k_+/\partial[\text{Zn}(\text{CR})\text{OH}]^+$, obtained using the equilibrium constant of reaction (1) (evaluation of k_+ , ref 2; thermodynamic data, ref. 7). Squares, triethanolamine buffer; circles, borate buffer. Correction for the changing $\text{p}K_a$ values of the buffers and of $[\text{Zn}(\text{CR})\text{OH}_2]^{2+}$ with temperature was made, along with allowance for the temperature dependence of the acetaldehyd hydration equilibrium⁷

TABLE 1

Summary of experimental data

Catalytic system	Temperature (°C)	pH	Rate ($\text{l mol}^{-1}\text{s}^{-1}$) ^a
ZnCR^{2+}	0	4.90 ^b	0.069
	0	5.26 ^b	0.11
	0	5.95 ^b	0.15
	0	7.93 ^c	14.5
	0	8.93 ^c	73.9
	0	9.03 ^c	79.6
	0	9.80 ^d	185
	0	10.20 ^d	172
	5	10.15 ^d	226
	10	8.72 ^c	116
	10	10.10 ^d	343
	20	8.50 ^c	194
ZnNMeCR^{2+}	0	9.70 ^d	141
$\text{Zn}(\text{NMeCR})\text{OH}^+$ ($\text{p}K_a$ 8.68 at 0 °C)	0	9.70 ^d	189
$\text{Znde-Me}_2\text{CR}^{2+}$	0	10.20 ^d	146
$\text{Zn}(\text{de-Me}_2\text{CR})\text{OH}^+$ ($\text{p}K_a$ 8.53 at 0 °C)	0	5.00 ^b	0
CuCR^{2+}	0	7.93 ^c	0
$\text{Cu}(\text{CR})\text{OH}^+$ ($\text{p}K_a$ ca. 12.8)	0	9.70 ^d	3 ± 1

^a This is the gradient in plots such as Figure 2. ^b Acetate buffer, 10mM. ^c Triethanolamine buffer, 10mM. ^d Borate buffer, 10 mM.

DISCUSSION

(a) *Mechanism of the Co-operative Catalysis by ZnCR^{2+} and Hydroxide.*—The rate law for this co-operative catalysis shows (Figure 3) that the transition complex

has a formula $[\text{ZnCR}^{2+}, \text{CH}_3\text{CHO}, \text{OH}^- (\text{H}_2\text{O})_n]$. The four reaction paths (α)—(δ) are kinetically equivalent and involve this transition complex. They will now be considered in turn, with the assumption that only one of them is actually operating.

(iii) Path (γ) involves the formation of a $[\text{Zn}(\text{CR})\text{-acetaldehyde}]^{2+}$ complex. This will have a very low stability constant, as acetone (and therefore presumably also acetaldehyde) is a weaker ligand than water,^{9,10} and in competition with 55.5M- H_2O will

TABLE 2
Catalysis of acetaldehyde hydration

Acids	Catalyst <i>i</i>	Rate constant k_i	Rate constant for forward reaction ^a
	CH_3COOH	0.100	0.073
	Pyridinium ion	0.024	0.017 4
	Pyridine-2-carboxylic acid zwitterion	0.012	0.008 7
	H_{aq}^+	225	163
	$\text{Zn}_{\text{aq}}^{2+}$	0.011	0.08
	ZnCR^{2+}	0.036	0.026 1
Base with an OH^- group	$\text{Zn}(\text{CR})\text{OH}^+$	196	142
	$\text{Zn}(\text{NMeCR})\text{OH}^+$	154	112
	$\text{Zn}(\text{de-Me}_2\text{CR})\text{OH}^+$	179	130
	$\text{Cu}(\text{CR})\text{OH}^+$	3 600 ^b	2 600
	OH^-	58 000 ^c	42 000
Other bases	H_2O	0.000 013 ^e	0.000 009 4
	CH_3COO^-	0.031	0.023
	Pyridine	0.032	0.023
	Pyridine-2-carboxylate	0.015	0.011
Bifunctional groupings	Borate	7	5
	$\text{Zn}_{\text{aq}}^{2+} + \text{CH}_3\text{COO}^-$	0.33 ^d	
	$\text{Zn}_{\text{aq}}^{2+} + \text{pyridine}$	1.3 ^d	
	Complex (1)	0.008	
	Complex (2)	2	

Units are $1 \text{ mol}^{-1} \text{ s}^{-1}$ and data from refs. 2—4 and this work unless otherwise stated. All data are for 0 °C.

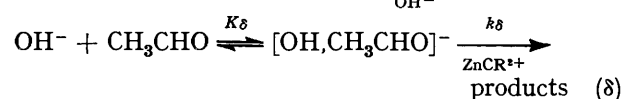
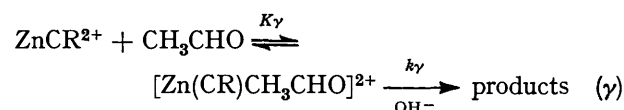
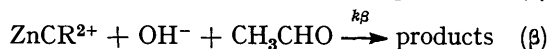
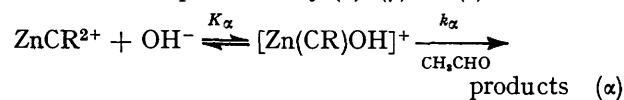
^a The corrected figure for the forward reaction only (see text) is used to construct Figures 4, 6, and 8a. ^b Based on the observed figure of ca. $2 \text{ l mol}^{-1} \text{ s}^{-1}$ for the complex at pH 9.5 and a spectrophotometrically estimated pK_a value of 12.8. ^c From ref. 15. ^d Units, $1^2 \text{ mol}^{-2} \text{ s}^{-1}$.

(i) Comparing equation (2) with path (α) and using the values $K_w = [\text{H}^+][\text{OH}^-] = 10^{-14} \text{ mol}^2 \text{ l}^{-2}$ and $K_a = 10^{-9.17}$ for ZnCR^{2+} (ref. 6), K_α is calculated to be $6.76 \times 10^4 \text{ l mol}^{-1}$ at 0 °C and low ionic strength. If the observed reaction proceeds by path (α), then from Table 2 $k_\alpha = 142 \text{ l mol}^{-1} \text{ s}^{-1}$, with an activation energy $E_{A(\alpha)}$ equal to $E_{A(\text{obs})}$, $6.3 \pm 0.3 \text{ kcal mol}^{-1} \text{ K}^{-1}$.

(ii) Comparing mechanisms (α)—(δ) gives equation (6).

$$K_\alpha k_\alpha = k_\beta = K_\gamma k_\gamma = K_\delta k_\delta \quad (6)$$

Path (β) is merely hypothetical, since genuine termolecular collisions in solution do not occur; the cage effect always reduces them to a pre-equilibrium between two species followed by a further reaction step involving collision with the third. The three-body reaction must therefore be represented by (α), (γ), or (δ).



⁹ R. F. Pasternack and R. A. Plane, *Inorg. Chem.*, 1965, 4, 1171; N. J. Friedman and R. A. Plane, *ibid.*, 1963, 2, 11.

therefore show a binding constant of $<0.018 \text{ l mol}^{-1}$. ZnCR^{2+} is a strongly polarising metal ion, so this effect will tend to be increased. We shall take $K_\gamma \lesssim 0.01 \text{ mol}^{-1} \text{ l}$. Unpublished ^1H and ^{13}C n.m.r. measurements by P. Wyeth and H. Földnes have confirmed that K_γ is very small ($<0.1 \text{ l mol}^{-1}$). Thus, using equation (6), $k_\gamma \gtrsim 9.6 \times 10^8 \text{ mol}^{-1} \text{ l s}^{-1}$. This is less by a factor of ca. 10 than the diffusion-controlled limit for reaction in water at 0 °C between OH^- and a positively-charged centre.¹¹

The entropy change of the K_γ step, ΔS_γ , may be estimated by comparison with other reactions in which a neutral, unidentate ligand displaces one water molecule, e.g. the substitutions of ammonia, pyridine, or imidazole for water in the equatorial plane of $\text{Cu}_{\text{aq}}^{2+}$. The average value is $\Delta S -5 \text{ cal mol}^{-1} \text{ K}^{-1}$.¹² Another compilation gives similar values.¹³ This provides sufficient information to calculate the activation energy of the k_γ step, given by equation (7) if path (γ) operates.

$$E_{A(\text{obs})} = -\Delta H_\alpha + \Delta H_\gamma + E_{A(\gamma)} \quad (7)$$

equal to the difference between the heats of ionisation of water ($14.62 \text{ kcal mol}^{-1}$ at 0 °C and low ionic strength¹⁴)

¹⁰ Compare the very low protonation constant of acetaldehyde, $10^{-10.2} \text{ l mol}^{-1}$, G. C. Levy, J. D. Cargioli, and W. Racela, *J. Amer. Chem. Soc.*, 1970, 92, 6238.

¹¹ D. N. Hague, 'Fast Reactions,' Wiley-Interscience, New York, 1971, especially pp. 80—94.

¹² L. G. Sillén and A. E. Martell, 'Stability Constants,' Chem. Soc. Special Publ., No. 17, 1964 and Supplement, No. 25, 1969.

¹³ R. J. P. Williams, *J. Phys. Chem.*, 1954, 58, 121.

¹⁴ V. P. Vasil'ev and G. A. Lobanov, *Zhur. fiz. Khim.*, 1967, 41, 838.

and of $[\text{Zn}(\text{CR})\text{OH}_2^{2+}]$,⁶ and has a value (-7.6 ± 1) kcal mol⁻¹. Thus if ΔS_\ddagger is -5 cal mol⁻¹ K⁻¹, $E_{A(\gamma)}$ is calculated to be -2.5 kcal mol⁻¹. A negative activation energy is a general criterion for the impossibility of a proposed elementary reaction. Path (γ), therefore, does not represent the true mechanism.

(iv) Path (δ) cannot be rigorously ruled out. However it is chemically implausible, since no complex intermediate of the type $[\text{CH}_3\text{CHO}, \text{OH}^-]$ is known other than that going directly to products without the assistance of ZnCR^{2+} . The k_δ step would presumably involve ligand substitution at zinc and its rate would be limited thereby to *ca.* 10^7 l mol⁻¹ s⁻¹, requiring a stability constant for $[\text{CH}_3\text{CHO}, \text{OH}^-]$ of the order of magnitude of 1 or greater.

We are therefore left with path (α) as the one which most plausibly represents the course of co-operative acetaldehyde hydration by ZnCR^{2+} and OH^- . Since $[\text{Zn}(\text{CR})\text{OH}^+]$ is a better catalyst than the stronger base borate (see Table 2), its effect must be attributed to direct attack of the bound hydroxide ion rather than to general-base catalysis.

(b) *Catalysis by $\text{Zn}_{\text{aq}}^{2+}$ and ZnCR^{2+} Alone.*—As previously shown,² $\text{Zn}_{\text{aq}}^{2+}$ most probably catalyses the hydration by activation of the carbonyl group² (Figure 5). A similar mechanism may be assigned to ZnCR^{2+} .

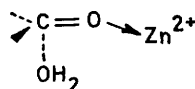


FIGURE 5 Catalysis of acetaldehyde hydration by $\text{Zn}_{\text{aq}}^{2+}$ or ZnCR^{2+}

The polarising effect of the three Lewis acids H^+ , ZnCR^{2+} , and $\text{Zn}_{\text{aq}}^{2+}$ upon the carbonyl group (*i.e.* $\log k_i$) may be compared with the polarising effect of these acids upon the water molecule (*i.e.* their $\text{p}K_a$). This is shown in Figure 6 (circles). The linear relationship suggested, with a slope of -0.34 , is also approximately obeyed when neither carbonyl group nor H_2O is polarised by a Lewis acid (square), but is not obeyed by the proton acids acetic acid, pyridinium ion, and α -picolinic acid zwitterion (diamonds); these have been shown to obey the Bronsted relation with an α value of 0.54 ,¹⁵ and our data are consistent with this (Figure 6). Since H_3O^+ obeys both relationships, one cannot say in this case whether its catalysis is better represented by a general-acid type or by a Lewis-acid type mechanism.

The slope of -0.34 incorporates both binding and polarising effects and has therefore only empirical significance.

(c) *Mechanism of Co-operative Catalysis by $\text{Zn}_{\text{aq}}^{2+}$ and OH^- .*—The co-operative catalysis between $\text{Zn}_{\text{aq}}^{2+}$ and OH^- can be described by mechanisms analogous to paths (α)—(δ). In this case path (γ) cannot be ruled out, although it is naturally most economical to assume that $[\text{ZnCR}]^{2+}$ and $[\text{Zn}(\text{OH}_2)_5]^{2+}$ behave similarly, so that path (α) holds for $\text{Zn}_{\text{aq}}^{2+}$ as well as for ZnCR^{2+} . The

¹⁵ R. P. Bell, M. H. Rand, and K. M. A. Wynne-Jones, *Trans. Faraday Soc.*, 1956, **52**, 1093.

observed rate [equation (5)] in conjunction with a $\text{p}K_a$ value for $[\text{Zn}(\text{OH}_2)_6]^{2+}$ of 10.1 at 0 °C¹⁶ yields a value for k_{ZnOH} of 79 l mol⁻¹ s⁻¹.

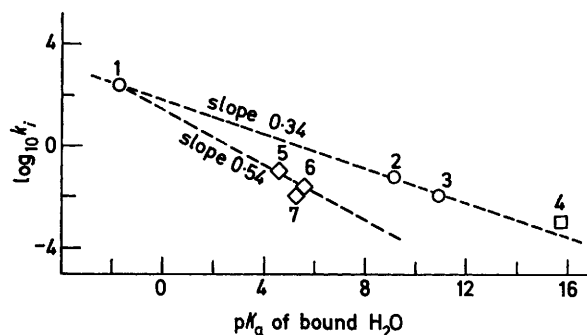


FIGURE 6 Correlation between catalytic activities of acids in acetaldehyde hydration. Data from refs. 2 and 3 and from this work. Key: 1, H^+ ; 2, ZnCR^{2+} ; 3, $\text{Zn}_{\text{aq}}^{2+}$; 4, no acid catalyst; 5, CH_3COOH ; 6, pyridinium ion; 7, picolinic acid zwitterion

(d) *Co-operative Catalysis by $\text{Zn}_{\text{aq}}^{2+}$ and Other Bases.*—Figure 7 shows two mechanisms for such catalysis involving zinc ions and an external base B (*e.g.* acetate or pyridine). These are both allowed by the kinetics,³ and the rates of the two paths are given by their contribution to k_{obs} , denoted by Δk_{obs} . Thus if Figure 7a holds, the rate is given by equation (8), and if Figure 7b holds, by (9). The values of $k_{\text{Zn},\text{B}}$ obtained by fitting experimental data³ to equation (8) are for acetate 0.33 and

$$\Delta k_{\text{obs}} = k_{\text{Zn},\text{B}}[\text{Zn}^{2+}][\text{B}] \quad (8)$$

$$\Delta k_{\text{obs}} = k_{\text{ZnOH},\text{BH}}[\text{ZnOH}^+][\text{BH}^+] \quad (9)$$

for pyridine 1.3 l² mol⁻² s⁻¹, in accordance with reasonable expectation based on the relative strengths of the two bases and on the much smaller catalysis obtained from $\text{Zn}_{\text{aq}}^{2+}$ or from acetate or pyridine alone. A similar fit to equation (9) (taking $\text{p}K_a$ values of 4.6 for acetate and 5.6 for pyridine) gives $k_{\text{ZnOH},\text{BH}}$ values of 1.0×10^5 for acetic acid and 4.1×10^4 l² mol⁻² s⁻¹ for the pyridinium

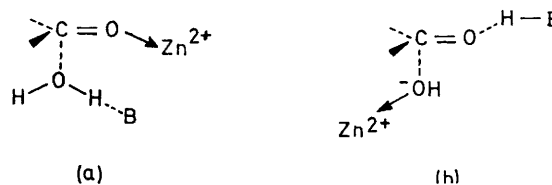


FIGURE 7 Kinetically equivalent mechanisms for the co-operative catalysis of acetaldehyde hydration with the formal participation of $\text{Zn}_{\text{aq}}^{2+}$ and a base B

ion. This is likewise in accordance with expectation based on the relative acidic strengths of acetic acid and the pyridinium ion, and on the smaller catalysis obtained from $\text{ZnOH}_{\text{aq}}^+$ or the two general acids separately. Thus, although Figure 7a may appear more reasonable than Figure 7b, it is not possible to state which of these two mechanisms is the correct one. A similar ambiguity exists for $[\text{Znpyox}^+]$ (2).⁴

¹⁶ L. A. Katsman, M. N. Vargaftik, and Ya. K. Syrkin, *Bull. Acad. Sci., U.S.S.R. (Chem. Sci.)*, 1974, 271.

Nucleophilicity and pK_a of Metal-bound Hydroxide. Comparison with Other Reactions.—The Brønsted relation

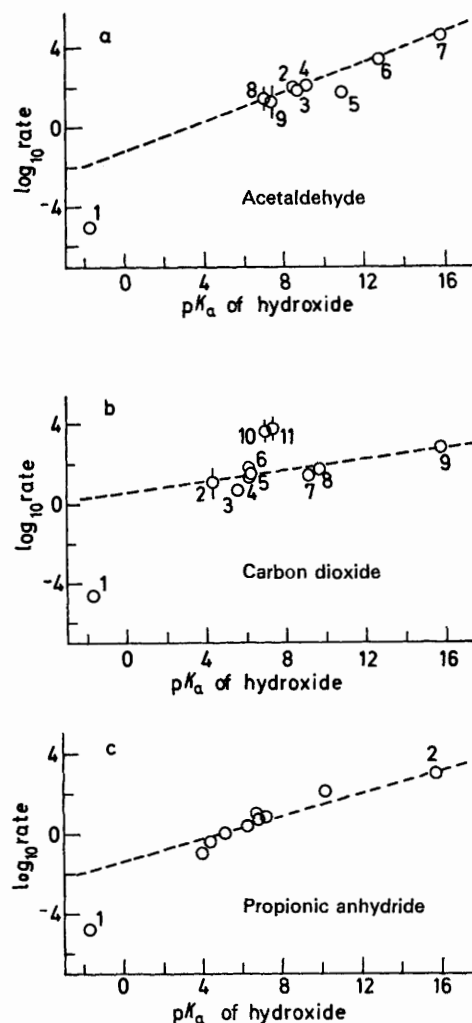


FIGURE 8 The rate of attack of the hydroxide group upon various substrates, plotted as a function of the pK_a . (a) Substrate, acetaldehyde; 1, H_2O ; 2, $[Zn(de-Me_2CR)OH]^+$; 3, $[Zn(NMeCR)OH]^+$; 4, $[Zn(CR)OH]^+$; 5, $[Zn(OH_2)OH]^+$; 6, $[Cu(CR)OH]^+$; 7, OH^- ; 8, bovine carbonic anhydrase; 9, human carbonic anhydrase, form C. Carbonic anhydrase data from ref. 27; to effect a comparison between the enzyme and the other catalysts the turnover number is divided by *ca.* 50 $mol\ l^{-1}$ in order to account for the presumed high concentration of catalyst at the active site. The Michaelis constant of the human enzyme is taken to be of similar order to that of the bovine enzyme, in accord with data from other substrates. The enzyme ordinates are therefore less certain than those for the other catalysts. All data are for 0 °C. (b) Substrate, carbon dioxide; 1, H_2O ; 2, $[(H_3N)_5CrOH]^{2+}$; 3, $[trenCoOH_2, OH]^{2+}$; 4, $[(H_3N)_5CoOH]^{2+}$; 5, $[(H_3N)_5RhOH]^{2+}$; 6, $[(H_3N)_5IrOH]^{2+}$; 7, $[ZnCR,OH]^+$; 8, $[Cu(L)OH]$ where L is glycol-glycinato; 9, OH^- ; 10, bovine carbonic anhydrase; 11, human carbonic anhydrase, form C. Data for 2 from ref. 24 (lower limit at 25 °C given); for 3–6 from Harris *et al.*,^{21–23} extrapolated to 0 °C using reported activation energies; for 7, from ref. 8 (0 °C); for 8, from ref. 18 (1.5 °C); for 9, from ref. 31 (0 °C); for 10, from refs. 28, 29; for 11, from ref. 30. 10 and 11 are converted to values expected for 0 °C using the reported activation energy of the turnover number³² (8.9 kcal mol^{-1}), and the turnover numbers are divided by *ca.* 50 $mol\ l^{-1}$ as in Figure 8a. (c) Substrate, propionic anhydride; 1, H_2O ; 2, OH^- ; other points, hydroxo-complexes of various kinetically inert transition metal ions. Redrawn from Buckingham and Engelhardt.¹⁹ Data are for 25 °C

may be applied to nucleophilic as well as to general acid-base catalysed reactions.¹⁷ Three such reactions are represented by their Brønsted plots in Figure 8. The reactions are the hydration of acetaldehyde, the hydration of carbon dioxide, and the hydrolysis of propionic anhydride; limited data are also available for the attack of oxygen at the keto carbonyl group.*

In each case the pattern is the same. The hydroxide donors fall onto a straight line, with the exception of water, which always falls *ca.* 2 log units below this line. The gradient of this line, comparable to the Brønsted β , is a measure of the amount of charge given up in the transition state by the attacking OH^- nucleophile to the carbonyl group, and for CO_2 , propionic anhydride, and acetaldehyde respectively has the approximate values 0.15, 0.25, and 0.35. The differing values may be due to the participation in the carbonyl double bond of *p* electrons on the neighbouring oxygen atom(s), which increases in the order aldehyde < anhydride < CO_2 . This could increase the difficulty of expansion of the coordination number of the carbon atom and thus make the reaction less sensitive to the strength of the attacking nucleophile.

The rates of attack upon acetylacetone by H_2O and by $[en_2CoOH_2,OH]^{2+}$ (pK_a 6) are, as expected, lower by a factor of *ca.* 10^2 than corresponding rates of attack upon acetaldehyde.²⁰

It is also informative to compare the enzyme carbonic anhydrase²⁵ with these metal hydroxides. A suggested mechanism²⁶ of hydration catalysis by carbonic anhydrase involves nucleophilic attack by zinc-bound hydroxide; rate data for the activity of carbonic

* In the case of acetaldehyde hydration only labile metal-hydroxide complexes have been studied, and the assumption is made that these catalysts act by mechanisms similar to that by which $[Zn(CR)OH]^+$ acts. The same mechanism has been put forward for CO_2 hydration by $[Zn(CR)OH]^+$ ⁶ and by $[Cu(glycyl-glycinato)OH]$.¹⁸ Buckingham *et al.*^{19,20} and Harris *et al.*^{21–23} used co-ordinatively inert metal ions, so that the products of attack on propionic anhydride and on CO_2 were isolable metal complexes, demonstrating that direct nucleophilic reaction occurs. Earley and Alexander²⁴ used isotope exchange to reach a similar conclusion.

¹⁷ W. P. Jencks, 'Catalysis in Chemistry and Enzymology,' McGraw-Hill, New York, 1969, pp. 79ff.

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²³ T. P. Dasgupta and G. M. Harris, *J. Amer. Chem. Soc.*, 1975, **97**, 1733.

²⁴ J. E. Earley and W. Alexander, *J. Amer. Chem. Soc.*, 1970, **92**, 2294.

²⁵ S. Lindskog, L. Henderson, K. K. Kannan, A. Liljas, P. O. Nyman, and B. Strandberg, in 'The Enzymes,' ed. P. D. Boyer Academic Press, New York, 3rd edn., vol. 5, pp. 587–665.

²⁶ R. P. Davis, in 'The Enzymes,' eds. P. D. Boyer, H. Hardy, and K. Myrback, Academic Press, New York, 2nd edn., vol. 5, pp. 545–562.

anhydrase in acetaldehyde²⁷ and CO₂²⁸⁻³⁰ hydration show that the enzyme falls into line with the other catalysts for acetaldehyde as a substrate (Figure 8a) but not for CO₂ (Figure 8b). The activity of [Zn(CR)OH⁺] and the enzyme have already been discussed in a more detailed comparison between ZnCR²⁺ and carbonic anhydrase.⁶ The broader context of Figures 8a and b lends weight to the contention that the postulated existence of a zinc-bound hydroxide nucleophile would indeed suffice to account for the catalytic activity of the enzyme in the hydration of acetaldehyde, but that if such a mechanism should apply for carbon dioxide

hydration some further enzymatic feature accelerates the attack on carbon dioxide only, such as a carbon dioxide-specific polarising group or groups in the protein, or a carbon dioxide-induced co-ordination expansion of the zinc.

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³¹ C. Faurholt, *J. Chim. phys.*, 1924, **21**, 400.

³² M. Kiese, *Biochem. Z.*, 1941, **307**, 400.