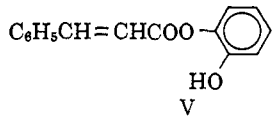
$$v = k'\alpha[\text{phenol}] = \frac{k'K_N}{K}[\text{phenolate}^-]$$

where K is the dissociation constant of the phenol, α is the fraction of the nucleophile in the anionic form, and K_N is the dissociation constant of the nucleophile, *viz.*

$$K_N = [H^+][N^-]/[HN] \cong [H^+][N^-]/[N]_{total}$$

Experiments are now in progress to distinguish between these two possible rate enhancement mechanisms. Hydroxyl groups, quite remote from a susceptible bond, when properly oriented can cooperate with a vicinal peptide group to very markedly enhance the reactivity. In some enzymes, such as pepsin[9] and carboxypeptidase,[10] where tyrosine groups are involved in the activity, the role of the phenolic group may be related to one or the other of the above-mentioned mechanisms. It should follow that the esters as well as imides of the *o*-OH phenyl derivatives should exhibit enhanced reactivity. The hydrolysis of monocinnamoylcatechol (V) was followed spectrophotometrically utilizing the change in optical density at 3000 Å. accompanying the hydrolytic reaction. The second-order specific rate of OH^- catalyzed hydrolysis is 2×10^4 M^{-1} min.$^{-1}$ near neutral pH. This is 300 times greater than the specific rate of phenyl cinnamate hydrolysis; another manifestation of the marked effect of an adjacent hydroxyl on reactivity. Similar results have been recently reported for monoacetylcatechol.[11]



V

A detailed report of these kinetic results, and of their potential pertinence to specific examples of enzymic catalysis, will be presented shortly.

Acknowledgment.—This work was supported by grants from the National Science Foundation and the Public Health Service.

(9) R. Herriott, *J. Gen. Physiol.*, **19**, 283 (1935).
(10) R. T. Simpson, J. F. Riordan, and B. L. Vallee, *Biochemistry*, **2**, 616 (1963).
(11) B. Hansen, *Acta Chem. Scand.*, **17**, 1375 (1963).
(12) On leave of absence from the Department of Biophysics, The Weizmann Institute of Science, Rehovot, Israel.

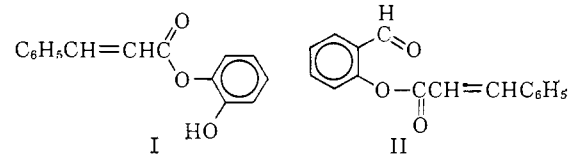
INSTITUTE OF MOLECULAR BIOLOGY AND DEPARTMENT OF CHEMISTRY
UNIVERSITY OF OREGON
EUGENE, OREGON
YECHIEL SHALITIN[12]
SIDNEY A. BERNHARD

RECEIVED MARCH 30, 1964

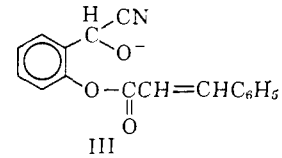
Neighboring Group Effects on Ester Hydrolysis. II. Neighboring Carbonyl Groups

Sir:

Previously,[1] and in an accompanying communication,[2] large rate enhancements in ester hydrolysis have been demonstrated in compounds containing a neighboring hydroxyl group. These results may be of pertinence to the mechanism of enzymic hydrolysis of acyl derivatives, wherever a serine hydroxyl residue is a constituent of the catalytically active site. Other neighboring functional groups may possibly exert rate enhancement effects as well. Carbonyl groups have been shown to increase the rate of hydrolysis of a neighboring ester.[3,4] Likewise, we have found that the aldehyde analog of I, cinnamoylsalicylaldehyde (II), undergoes very rapid hydrolysis. The rate is first order in OH^- concentration. The second-order specific rate is approximately 7×10^5 M^{-1} min.$^{-1}$. The activity is presumably due to the hydrated aldehyde or its conjugate base. This is indicated by the fact that in 0.002 M KCN solution, where the reactive species is probably the cyanohydrin (III), the rate of hydrolysis is increased tenfold. The catalysis may proceed similarly to either Scheme A or B of ref. 2, or *via* a nucleophilic attack on the carbonyl ester as suggested for the hydrolysis of *o*-formyl benzoate esters.[4]

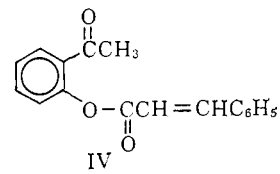


I II



III

A related ester containing a neighboring ketone, the cinnamoyl ester of *o*-hydroxyacetophenone (IV), is hydrolyzed with a specific rate of 2.6×10^3 M^{-1} min.$^{-1}$; very much slower than the hydrolysis of III,



IV

but nevertheless 40-fold faster than the hydrolysis of phenyl cinnamate. Rate enhancement by CN^- is similar to that found with II.

Carbonyl groups of aldehydes and ketones, especially the former, can have a great influence on the rate of hydrolysis of neighboring ester groups. The available evidence indicates, however, that the rate enhancement is, in reality, exerted by the hydroxylic adduct or its conjugate base.

Acknowledgment.—This work was supported by grants from the National Science Foundation and the Public Health Service.

(1) S. A. Bernhard, A. Berger, J. H. Carter, E. Katchalski, M. Sela, and Y. Shalitin, *J. Am. Chem. Soc.*, **84**, 2421 (1962).
(2) Y. Shalitin and S. A. Bernhard, *ibid.*, **86**, 2291 (1964).
(3) M. S. Newman and S. Hishida, *ibid.*, **84**, 3582 (1962).
(4) M. L. Bender and M. S. Silver, *ibid.*, **84**, 4589 (1962).
(5) On leave of absence from the Department of Biophysics, The Weizmann Institute of Science, Rehovot, Israel.

INSTITUTE OF MOLECULAR BIOLOGY AND DEPARTMENT OF CHEMISTRY
UNIVERSITY OF OREGON
EUGENE, OREGON
YECHIEL SHALITIN[5]
SIDNEY A. BERNHARD

RECEIVED APRIL 27, 1964

Search For Accidental Degeneracy in Purines

Sir:

The strong adenine absorption at 260 mμ (38.5 kk.) appears to be a single electronic transition. Mason[1]

(1) S. F. Mason, *J. Chem. Soc.*, 2071 (1954).