Inhibition of Muscle Phosphorylase $a$ by 5-Gluconolactone*

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SUMMARY

5-Gluconolactone is a potent inhibitor of rabbit muscle phosphorylase $a$ in the presence of saturating AMP. Kinetic analysis leads to the conclusion that the inhibitor binds most strongly to the enzyme-glycogen-P$_i$ complex (K$_{AI}$ = 0.025 mm), but weaker binding is also observed with the enzyme-glycosyl-$\alpha$-d-glucopyranose 1-phosphate complex, the enzyme-glycogen complex, the enzyme-P$_i$ complex, and the free enzyme. Dissociation constants for each of the observed inhibitor complexes have been evaluated. The kinetic data is consistent with a model in which 5-gluconolactone binds to the site in the enzyme that normally binds the glucosyl residue that is transferred between polysaccharide and P$_i$ in the catalytic reaction. 5-Gluconolactone may be an analogue of the substrate part of the transition state of the phosphorylase reaction.

5-Gluconolactone has recently been reported by Tu, Jacobson, and Graves (1) to be a good inhibitor of maltodextrin phosphorylase from Escherichia coli and glycogen phosphorylase $b$ from rabbit muscle ($\alpha$-1,4-glucan:orthophosphate glucosyltransferase EC 2.4.1.1). Although the kinetics of inhibition of muscle phosphorylase $b$ were complex, their studies indicated that 5-gluconolactone binds to the central complex of enzyme with arsenate and glycogen but fails to bind to the central complex containing $\alpha$-d-glucopyranose 1-phosphate and glycogen. This led to the conclusion that the inhibitor occupies the site that normally binds the glucose residue that is transferred between P$_i$ and glycogen in the catalytic reaction. They suggested, on the basis of this and other evidence, that the transition state probably involves glucose in the half-chair conformation with some degree of positive charge at C-1.

Lactones derived from various hexoses, $N$-acetylhexosamines, and hexuronic acids have been studied extensively as inhibitors of glycosidases (2–4). Inhibition by these compounds, in which C-1 has been converted to a carbonyl group, is greatest when their configurations and ring sizes correspond to those of the natural substrates. Thus, 5-gluconolactone is a better inhibitor of both $\alpha$- and $\beta$-glycosidases than is 5-mannosanolate, while the converse is true for mannosidases. Relatively little can be learned about the nature of the inhibition by steady state kinetic methods because glycoside hydrolysis is a unisubstrate reaction and the reverse reaction can not easily be observed. Lactones generally are competitive inhibitors of the glycoside substrates, indicating that they bind at the substrate sites. In contrast, the glycogen phosphorylase reaction is quite complex, involving many distinguishable enzyme forms and the possibility of inhibitor binding to any or all of the forms.

In the present work, we have studied the inhibition of rabbit muscle phosphorylase $a$ by 5-gluconolactone in the presence of saturating 5'-AMP. The kinetic mechanism of phosphorylase $a$ is known to be rapid equilibrium random bi-bi (5–7) and the kinetic constants represent macroscopic dissociation constants of the appropriate enzyme forms. We have concluded that 5-gluconolactone is a highly specific inhibitor of phosphorylase $a$, having the property of binding to both central complexes. Evidence indicating that the inhibitor binds to the free enzyme, the enzyme-glycogen complex, and the enzyme-P$_i$ complex has also been obtained. No indication of binding to the enzyme-glucose-1-P complex has been found, but a weak affinity for this enzyme form cannot be excluded. Of the various enzyme forms which bind 5-gluconolactone, the affinity of the enzyme-glyco-1-P$_i$ central complex for the inhibitor is the greatest. This evidence conflicts with that of Tu et al. (1) with respect to the competitive relation between inhibitor and glucose-1-P$_i$; however, it is consistent with a model that differs only slightly from theirs. Our results support the idea that the glucosyl residue is transferred between P$_i$ and polysaccharide in the form of a stabilized carbonium ion or its equivalent.

EXPERIMENTAL PROCEDURE

Materials—All materials are the same as described in a previous publication (5).

Methods—Concentration of phosphorylase $a$ was determined spectrophotometrically at 279 nm using $A_{279}^{\text{em}} = 13.0$. Glyco-
gen concentration is expressed as total concentration of glucosyl residues and is based upon dry weight of glycogen. All experiments were carried out at 30° and pH 6.8.

Kinetic measurements were carried out by the isotopic assay
described previously (5). Rates in the direction of glycogen synthesis were determined by estimating the amount of \(^3\)P liberated as Pi from isotopic glucose-1-P, while rates in the direction of glycogen degradation were determined by estimating the synthesis of isotopic glucose-1-P from \(^3\)P-containing Pi. All assays were carried out in the presence of 0.025 m potassium maleate buffer (except as noted in the figures), 0.10 mm EDTA, 7.0 mm cysteine, and 0.10 mm AMP. Rather than maintain a fixed ionic strength, as was done previously (5), changes in ionic strength within particular experiments were minimized by including 0.10 m KCl in all experiments involving low concentration of glucose-1-P or Pi, regardless of whether these were the fixed or varied substrates. Experiments in which the phosphate (or buffer) was present in relatively high fixed concentration were done in the absence of added KCl.

Because of the rapid hydrolysis of 5-gluconolactone under the experimental conditions (\(t_4 = \text{approximately 30 min}\)) relatively little time was allowed to elapse between dissolution of the inhibitor and completion of the incubation with enzyme and substrates. The crystalline inhibitor was dissolved in distilled water and aliquots were distributed to each reaction mixture immediately prior to starting the reaction by addition of enzyme. Hydrolysis of the lactone in distilled water is relatively slow because of the low pH of such solutions. Enzyme incubation times were usually restricted to 2.5 min, except in a few cases (noted in the figures) in which incubation was carried out for 5 min. All experimental points were done in duplicate; reversing the order of addition of enzyme to the samples in the duplicate set did not appear to alter the results significantly. Nevertheless, greater scatter of experimental rates occurred in the presence of inhibitor than in its absence.

For the analysis of our results we have adopted the nomenclature and system of kinetic constants proposed by Cleland (8). Curve fitting was accomplished by use of a series of computer programs described by Cleland (9). Slopes and intercepts of individual lines in reciprocal plots were determined using the HYPERBOLA program without weighting factors. Parameters for parabolic reciprocal plots were computed with the PARABOLA program; in experiments carried out at high substrate concentration the velocities were used as weighting factors, while in experiments at low substrate concentrations the points were not weighted. Replots of slopes, intercepts, or parabolic parameters versus inhibitor concentration were fitted using the LINE program, with the reciprocals of the squares of the standard errors as weighting factors. No correction was made for the hydrolysis of the gluconolactone. All the programs provide standard errors for the computed parameters. In the figures, experimental points are averages of duplicate values while the lines and curves are calculated from the computer-derived parameters. All points were used individually for the computer solutions and duplicates were averaged only for representation in the figures.

**RESULTS**

The kinetic mechanism of phosphorylase \(a\) has been shown to be rapid equilibrium random bi-bi (5-7) and, in the presence of saturating AMP, approximates the model shown in Fig. 1. \(A, B, P\) represent glycogen, glucose-1-P, and Pi, respectively. The kinetically observed Michaelis constants are macroscopic dissociation constants of the designated ligands from ternary complexes; the inhibition constants are macroscopic dissociation constants of the binary complexes. Only one \(EA\) complex is indicated, although it seems reasonable that two such complexes exist, one in which glycogen functions as an acceptor and another in which it functions as a donor. Since these complexes are iso-meric they are indistinguishable by equilibrium or by steady state kinetic methods and \(K_{ia}\) represents the macroscopic dissociation constant of glycogen from the equilibrium mixture of \(EA\) complexes.

The most general case of reversible inhibition, in which inhibitor binds to each of the six enzyme forms, is characterized by another set of six equilibria.

\[
K_{ia} A + I \rightleftharpoons K_{ia}A + I
\]

Rate Equation 1 may be derived from the kinetic model and the six inhibitor equilibria. Other relationships that pertain to this model are given in Equations 2, 3, and 4.

\[
V_1^{AB} - V_1^{AP}/K_{eq} = \frac{V_1 K_{b}(1+I/K_b)+K_{A}A(1+I/K_{A})}{K_{id} K_{b}(1+I/K_{id})+K_{A}A(1+I/K_{A})} + \left[\frac{K_{p}P(1+I/K_{p})+AP(1+I/K_{AP})}{V_1/V_2 K_{eq}}\right]
\]

\[
K_{ia} K_{b} = K_{ia} K_{ib}
\]

\[
K_{ia} K_{p} = K_{ip} K_{ip}
\]

\[
K_{eq} = \frac{V_1 K_{p}}{V_2 K_{b}}
\]

It is possible to evaluate \(K_{AP}\) by determining the dependence upon inhibitor concentration of apparent maximum velocities in the direction of glycogen synthesis and degradation, respectively. However, it is not practical to do a complete initial velocity experiment at each of several concentrations of inhibitor because of the large number of individual measurements required and the instability of inhibitor and enzyme over the necessary time. We made use of a method suggested by Cleland (10), in which both reactants are varied together in fixed ratio; this allows a rapid approach to the maximum velocity as the substrate concentrations are raised. In the case of an enzyme with a sequential mechanism, such as phosphorylase \(a\), the recip-
rocal plot is a parabola and the maximum velocity can be estimated by a least squares method of curve fitting. If \( AB = p \) and \( P = 0 \) the reciprocal of the rate is expressed by Equation 5, the coefficients of which are defined in Equations 6 through 8.

\[
\frac{1}{v} = \sigma + \beta \left( \frac{1}{B} \right) + c \left( \frac{1}{B} \right)^2 \tag{5}
\]

\[
\sigma = \frac{(1 + 1/K_{AB})}{V_1} \tag{6}
\]

\[
\beta = \frac{K_a (1 + 1/K_{B1}) + \beta K_b (1 + 1/K_{A1})}{V_1 P} \tag{7}
\]

\[
c = \frac{K_a K_b (1 + 1/K_{B1})}{V_1 P} \tag{8}
\]

Clearly, parameter \( a \) depends upon binding of \( I \) to the centra complex and parameter \( c \) depends upon binding of \( I \) to the free enzyme. The parameter \( b \) is more complex and depends upon binding of \( I \) to both \( EA \) and \( EB \). Dissociation constants \( K_{AB} \) and \( K_I \) can be evaluated from replots of \( a \) and \( c \), respectively.

A representative experiment in which glycogen and glucose-1-P are varied together over a range of high concentrations is shown in Fig. 2. The intercepts are clearly dependent upon inhibitor concentration and a replot of a versus \( I \) yields a value of 3.4 ± 0.7 mM for \( K_{AB} \). A similar experiment in which glycogen and \( P_1 \) are varied together is illustrated in Fig. 3. In this case the concentrations of substrates are high enough to make the contribution from the \( (1/P)^2 \) term negligible and the curve approximates a straight line. Consequently, the best lines were computed using the HYPERBOLA program. A replot of intercepts versus \( I \) yields a value of 0.025 ± 0.007 mM for \( K_{AP} \). In order to evaluate \( K_I \), the experiment was repeated using low concentrations of glycogen and \( P_1 \). The results are illustrated in Fig. 4 and a replot of \( c \) versus \( I \) is shown in the inset; \( K_I \) is calculated to be 7.5 ± 1.2 mM.

Having values for \( K_I \), \( K_{AB} \), and \( K_{AP} \), it is possible to estimate other inhibitor dissociation constants in more conventional
experiments. If Equation 1 is rearranged to express the reciprocal of the velocity as a function of the reciprocal of the glycogen concentration at fixed concentrations of glucose-1-P and inhibitor (in the absence of P), Equation 9 is obtained. A replot of intercept versus $I$ gives rise to a straight line. The ratio of intercept of the replot to its slope is an apparent constant, $K_{in}$, the value of which is given in Equation 10. In a similar way, Equation 1 can be rearranged to express the reciprocal of the velocity as a function of the reciprocal of the glucose-1-P concentration at fixed concentrations of glycogen and inhibitor. A replot of the slope versus $I$ also gives a straight line and the ratio of intercept to slope of the secondary plot is an apparent constant, $K_{slp}$, whose value is given in Equation 11. Since $K_{AT}$ and $K_I$ are known, it is possible to evaluate $K_{AT}$ from Equation 10 in an experiment carried out at a low fixed concentration of glucose-1-P. Likewise, $K_I$ and $K_{IA}$ are known, making it possible to evaluate $K_{AT}$ from Equation 11 in an experiment carried out at a high fixed concentration of glycogen.

\[
\frac{1}{v} = \left[ \frac{1}{V_1} + \frac{1}{V_1} \left( \frac{1}{K_{AB}B + K/BK} \right) \right]
\]

\[
K_{int} = \frac{1 + K_b/B}{1/K_{AB} + K/BK_A} \quad (10)
\]

\[
K_{slp} = \frac{1 + K_{IA}/A}{1/K_{AI} + K_{IA}/K_A} \quad (11)
\]

These experiments are illustrated in Figs. 5 and 6, respectively. In Fig. 5, glycogen is varied at constant 2.0 mM glucose-1-P. $K_{in}$ is 1.01 mM and, using $K_b = 2.7$ mM (5), we obtain $K_{AT} = 0.65$ mM. In Fig. 6 glucose-1-P is varied at constant 4.0 mM glycogen. $K_{slp}$ is 0.78 ± 0.12 mM and, using $K_{IA} = 0.56$ mM (5), we calculate $K_{AT} = 0.69 ± 0.10$ mM. A similar experiment gave $E_{AT} = 0.35 ± 0.14$ mM, but the agreement must be considered satisfactory in view of the experimental errors. It is reasonable to take the larger number as the best value of $K_{AT}$. No attempt was made to evaluate $K_{AT}$ since it is likely to be quite large and technical difficulties preclude the appropriate experiment.

Similar equations can be derived for the phosphorolysis reaction. When glycogen is the variable substrate $K_{slp}$ is given by Equation 12, which can be used to evaluate $K_{PI}$ in an experiment carried out at high P. Results of an experiment carried out at 10 mM P, are shown in Fig. 7. $E_{slp} = 0.67 ± 0.22$ mM and, using $K_{slp} = 4.7$ mM (5), we obtain $K_{PI} = 0.47 ± 0.15$ mM.
Because of the rapid hydrolysis of 5-gluconolactone at pH 6.8, which significantly limits the accuracy attainable in the kinetic experiments reported here, it was necessary to establish that gluconate ion does not inhibit phosphorylase a. An experiment carried out at high concentrations of substrates (20 mM P_i and 10 mM glycogen) indicated that neither 0.050 mM nor 5.0 mM gluconate had any inhibitory effect. A similar experiment carried out at low substrate concentration (0.10 mM P_i and 0.050 mM glycogen) indicated no inhibition by 5.0 mM or 20.0 mM gluconate. It appears that gluconate ion does not have a significant affinity for either the free enzyme or the enzyme-P_i-glycogen central complex and does not interfere with the experiments reported above.

Since the effect of AMP on gluconolactone binding was not investigated, it is important to establish that the AMP concentration does not significantly influence the results developed above. In an experiment carried out at high substrate concentrations (20 mM P_i and 10 mM glycogen), 0.050 mM gluconolactone was found to produce 61% inhibition when the AMP concentration was 0.10 mM and 59% inhibition at 1.0 mM AMP. In a similar experiment carried out at low substrate concentration (0.10 mM P_i and 0.050 mM glycogen), 20 mM gluconolactone produced 76% inhibition at 0.10 mM AMP and 77% inhibition at 1.0 mM AMP. We may conclude either that AMP has little effect upon the binding of gluconolactone to free enzyme or enzyme-P_i-glycogen complex, or, more likely, that the system is saturated with AMP at a concentration of 0.10 mM. In either case, the results reported above are valid for phosphorylase a saturated with AMP.

The finding that 5-gluconolactone binds to the enzyme-glycogen-glucose-1-P complex differs from the results of Tu et al. (1) with muscle phosphorylase b. They observed complex kinetics in which the double reciprocal plot was biphasic when glucose-1-P was varied to very high concentrations in the presence of the inhibitor and saturating concentrations of glycogen and AMP. In the case, the results reported above are valid for phosphorylase a in this work. Although the data obtained at high concentrations of glucose-1-P do not qualitatively change the result obtained at low concentrations, it does effect the apparent value of the reciprocal plots reported by Tu et al. (1) for phosphorylase a. Kinetic constants have been determined under similar conditions (5), but their errors are likely to be substantial and to the errors inherent in the present experiments. K_I and K_B are probably reliable only within a factor of 2. The remaining dissociation constant, K_A, was not determined because its value is likely to be quite large and this complicates the experiment. The effect of changing P_i to glucose-1-P in the central complexes is to increase the inhibitor dissociation constant by a factor of 140; if this effect is the same in the binary complexes, we can calculate an approximate value of 70 mM for K_B. An alternate method of approximation is to consider that the effect of binding glucose-1-P to the enzyme-glycogen central complex is to increase the inhibitor dissociation constant by a factor of 5; if the effect of binding glucose-1-P to the free enzyme is similar, K_B is 40 mM. By either approximation, K_B is large compared with the other dissociation constants.

Some note must be taken of the deviations from linearity of the reciprocal plots reported by Tu et al. (1) for phosphorylase b and maltodextrin phosphorylase and observed with phosphorylase a in this work. Although the data obtained at high concentrations of glucose-1-P do not qualitatively change the result obtained at low concentrations, it does effect the apparent value of the dissociation constant K_A by a factor of 3. Since similar effects are observed with two different enzymes, it is possible that it is not a property of the enzymes themselves but is an anomaly of the method. Tu et al. (1) discuss some possible rationalizations, but no convincing explanation is apparent.

It is unusual to find an inhibitor that binds reversibly to a central complex (12), and even more extraordinary when the difference in affinity for inhibitor of the two isomeric central complexes (1, 12).

### Table I

| Apparent dissociation constants of enzyme-gluconolactone complexes |
|---------------------------------------------------------------|
| The conditions are pH 6.8, 30°, 0.10 mM AMP. The limits shown are standard errors obtained from secondary plots of linear or parabolic parameters versus inhibitor concentration. The constants are defined in the text. |
| K_I = 7.5 ± 1.2 mM |
| K_Pi = 0.47 ± 0.15 mM |
| K_A = 0.69 ± 0.10 mM |
| K_B = 3.4 ± 0.7 mM |
| K_APi = 0.025 ± 0.007 mM |
complexes is as great as in the present case. These effects are reasonable on the basis of a model in which 5-gluconolactone binds specifically to that area of the active site that normally binds the glucosyl residue that is transferred between polysaccharide and phosphate. Fig. 8 is a schematic representation of the active site of phosphorylase. In phosphorolysis the glucosyl residues in the terminal segment of a glycogen chain occupy Sites \( T, U, V, \) and \( W \), with the terminal residue (destined for transfer to \( P_i \)) in Site \( T \) and the penultimate residue in site \( U \). Inorganic phosphate occupies Site \( P \) and the terminal glucose residue is transferred from its linkage in the polysaccharide to \( P_i \) without dissociation of any groups from their binding sites. The analogous central complex for glycogen synthesis has the terminal glucose residue in Site \( U \), rather than \( T \), and the penultimate residue in Site \( V \). The phosphate group of glucose-1-P occupies the \( P \) site with the glucosyl moiety in the \( T \) site. The reaction is the reverse of phosphorolysis, with the glucosyl group leaving \( P_i \) and attaching to the C-4 hydroxyl group of the glucose residue in Site \( U \). This model is similar to that suggested by Chao, Johnson, and Graves (13) for the maltodextrin phosphorylase of \( E. \) coli.

The enzyme-glycogen-P, -inhibitor complex has gluconolactone bound to Site \( T \), the terminal and penultimate glucose residues of the polysaccharide bound to Sites \( U \) and \( V \), respectively, and \( P_i \) bound to the \( P \) site. The structure of the enzyme-glycogen-glucose-1-P-inhibitor complex is similar, except that glucose-1-P is bound only at the \( P \) site and the glucose moiety is either projecting into the aqueous phase or bound weakly at an adjacent site which is not involved in catalysis. This explains the 140-fold difference in affinity of inhibitor for the two central complexes, since in the latter complex the glucosyl residue of the glucose-1-P competes with gluconolactone for the \( T \) site and the competition is especially effective since it is intramolecular.

This interpretation is supported by the large effect of substrates on the binding of inhibitor by the enzyme. Binding of either glucose or \( P_i \) enhances the affinity of the enzyme for 5-gluconolactone by a factor of more than 10, while binding of both substrates increases the affinity by a factor of 300. It is possible that this is caused by stepwise conformational changes induced by the substrates, but it is more reasonable to assume that it indicates the inhibitor and substrate sites are immediately adjacent and there is direct interaction between ligands.

Inhibition of phosphorylase \( a \) by 5-gluconolactone is especially striking when we consider the effect of glucose. Helmreich, Michaelis, and Cory (14) observed that 50 mM glucose inhibits phosphorolysis by phosphorylase \( a \) and that the effect is principally upon the kinetic constants of substrates and AMP. There appeared to be little or no effect upon \( V_{\text{max}} \) either in the presence or in the absence of AMP. Our experiments indicated no detectable inhibition by 3 mM glucose in the presence of high concentrations of \( P_i \) and glycogen and 0.10 mM AMP. Clearly, glucose does not bind to the central complex.

A question arises as to why gluconolactone has such a high affinity for the enzyme-glycogen-P, -complex when glucose itself shows no tendency to bind, even at a concentration of 50 mM. The answer could lie in the electronic structure of 5-gluconolactone (Structure \( A \) in Fig. 9) which resembles that of glucosyl oxonium ion (Structure \( B \) in Fig. 9). The three-dimensional structure of 5-gluconolactone in the crystalline state has been determined (15) and it appears only to approximate the expected half-chair conformation. If the lactone system exhibited maximum electron delocalization, C-1, C-2, C-5, O-1, and O-5 would be coplanar. In fact, C-5 is 0.28 Å below the plane occupied by the other atoms and delocalization cannot be maximum. Nevertheless, some positive charge must reside on O-5. There is no way to know the precise conformation of gluconolactone, but it is likely that it is similar to that of gluconolactone, in spite of the fact that this would lessen the stabilization of the positive charge. Steric considerations are apparently important enough in the lactone to require the sacrifice of some of the delocalization energy, and these steric effects must be present in glucosyl oxonium ion.

The kinetic mechanism of phosphorylase \( a \) makes a covalent glucosyl enzyme very unlikely as an intermediate in the catalytic reaction. Yet it is necessary to postulate something nearly equivalent since the reaction proceeds with bond cleavage between C-1 and oxygen (16) and the configuration at C-1 of the transferred glucosyl group is retained. One possibility is that the glucosyl group is held as a stabilized carbonium ion in transit from the donor to the acceptor, with the provision that both donor and acceptor must be bound to the enzyme during the transfer. This can explain the great affinity of 5-gluconolactone for the central complex, since the quaternary complex resembles the transition state of the reaction. Wolfenden (17) has argued that transition state analogues should be bound to enzymes much more strongly than the substrates themselves. His argument cannot be pursued quantitatively in the present case, since a nonenzymatic phosphorolysis reaction has not been observed and since it is not known how closely 5-gluconolactone approximates the substrate portion of the transition state. Nevertheless, the inhibitor binds to one central complex with a much higher affinity than either substrates or substrate-analogue inhibitors show in this system and this qualitatively supports the notion that 5-gluconolactone is a transition state analogue. A similar conclusion has been reached by Tu et al. (1) on the basis of their inhibition studies and observations of secondary kinetic isotope effects with phosphorylase \( b \).
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REFERENCES

1. Tu, J.-I., Jacobson, G. R., and Graves, D. J., Biochemistry, 10, 1229 (1971).
2. Conchie, J., Gelman, A. L., and Levy, G. A., Biochim. Biophys. Acta, 91, 609 (1964).
3. Conchie, J., Hay, A. J., Strachan, I., and Levy, G. A., Biochim. Biophys. Acta, 102, 620 (1967).
4. Levy, G. A., Hay, A. J., and Conchie, J., Biochem. J., 91, 375 (1964).
5. Gold, A. M., Johnson, R. M., and Tseng, J. K., J. Biol. Chem., 245, 2584 (1970).
6. Engers, H. D., Shochosky, S., and Madsen, N. B., Can. J. Biochem., 48, 3404 (1970).
7. Engers, H. D., Bridger, W. A., and Madsen, N. B., Can. J. Biochem., 48, 755 (1970).
8. Cleland, W. W., Biochim. Biophys. Acta, 97, 104 (1963).
9. Cleland, W. W., Nature, 198, 463 (1963).
10. Cleland, W. W., Ann. Rev. Biochem., 36, 77 (1967).
11. Fiske, C. H., and Subbarow, Y., J. Biol. Chem., 66, 375 (1925).
12. Cleland, W. W., Biochim. Biophys. Acta, 67, 188 (1963).
13. Chao, J., Johnson, G. F., and Graves, D. J., Biochemistry, 8, 159 (1969).
14. Helmreich, E., Michaelides, M. C., and Cori, C. F., Biochemistry, 6, 395 (1967).
15. Hackert, M. L., and Jacobson, R. A., Chem. Commun., 1179 (1969).
16. Cohn, M., J. Biol. Chem., 180, 771 (1949).
17. Wolfenden, R., Nature, 223, 704 (1969); Biochemistry, 9, 3404 (1970).
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