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A Kinetic Study of the Rate of Cleavage of the
Glycosidic Bond of Methyl- β -Glucopyranoside
in an Alkaline Medium

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A KINETIC STUDY OF THE RATE OF CLEAVAGE OF
THE GLYCOSIDIC BOND OF METHYL- β -GLUCOPYRANOSIDE
IN AN ALKALINE MEDIUM

A thesis submitted by

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SUMMARY

Two mechanisms are encountered in the over-all degradation of polysaccharides in an alkaline medium: a progressive, stepwise degradation starting at the reducing end called the "peeling" reaction and random cleavage of the glycosidic bonds. There is no study of the relative importance of these mechanisms in the literature. In the current work, methyl- β -glucoside was used as the reactant. Since this molecule is blocked at the reducing end, "peeling" cannot occur and the importance of the cleavage mechanism alone could be assessed. At the outset of the current work the effect of temperature, pH, and molecular oxygen content of the system on the rate of cleavage of the glycosidic bond were unknown and disagreement existed on the effect of molecular oxygen on over-all polysaccharide degradation.

Reaction conditions included temperatures from 140 to 170°C., several levels of molecular oxygen concentration and two levels of hydroxyl ion concentration. At varying reaction times the concentration of unused methyl- β -glucoside was determined polarimetrically and the methanol produced from the degradation was measured colorimetrically.

The Arrhenius diagram of the reaction run under nitrogen-purged conditions in 10% sodium hydroxide at temperatures from 140 to 170°C. is linear, indicating a single rate-controlling reaction mechanism over this temperature range. From the literature, polysaccharide degradation is much more severe at 170°C. than at 100°C. when both reactions are carried out to the same yield. It has been assumed that this is explained by an increase in the relative importance of random cleavage with the increase in reaction temperature. From the literature it is also found that as the temperature is increased from 140 to 170°C. the rate of over-all

polysaccharide degradation is increased about threefold. From the current work, however, a 22-fold increase in the rate of cleavage was found between 140 and 170°C. The Arrhenius activation energy for this cleavage reaction was found to be 36,000 cal./g. mol. Since the activation energy for the over-all degradation of polysaccharides, which includes both the "peeling" and cleavage mechanisms, has been found to be near 17,000 cal./g. mol. the activation energy required for the "peeling" reaction alone must be quite low. In that the activation energy for random cleavage is much greater than that for "peeling" it follows that at high temperatures, e.g., 170°C., random cleavage tends to be rate controlling while at low temperatures, e.g., 100°C., "peeling" is controlling.

In the above comparison of activation energies the assumption is made that the glycosidic bonds of polysaccharides and methyl- β -glucoside respond similarly to changes in reaction variables. In addition, the two mechanisms of degradation are not totally independent because each time a polysaccharide chain cleavage takes place a reducing end is formed which is subject to further degradation by "peeling." Therefore, an increase in the rate of random cleavage will also increase the rate of "peeling." This interaction does not affect any of the conclusions drawn but merely makes the treatment of results less quantitative than would have otherwise been possible.

When excess molecular oxygen was used a marked effect of pH and temperature was found. In 10% sodium hydroxide, pH about 14, the reaction rate was increased 18 times at 170°C. and 45 times at 150°C. by the inclusion of excess molecular oxygen. When the pH was decreased to 10.2 the presence of excess molecular oxygen increased the reaction rate 64 times at 150°C. In all cases oxygen was found to be very effective in increasing the rate of reaction, this effect increasing as the severity of the other reaction conditions was decreased. This is probably

caused by a lower activation energy for the reaction involving oxygen than for the reaction excluding oxygen.

The quantity of molecular oxygen required in the cleavage of the glycosidic bond was also estimated. At 170°C. 2.5 moles of oxygen were used per mole of methyl- β -glucoside. This ratio increased to 3.7 at 150°C., indicating that more oxygen reacts via secondary reactions at the lower temperature. This is undoubtedly caused by a lower over-all activation energy for the secondary reactions than for the reaction with oxygen causing cleavage of the glycosidic bond.

From the literature on the over-all degradation of polysaccharides, it has been demonstrated that as the pH is increased from 11 to 14 the average D.P. decreases, although the amount of recoverable polymer is the same. This was explained by assuming that "peeling" is the primary degradation mechanism at the lower pH but that cleavage increases in relative importance as the pH is increased. In the current work with methyl- β -glucoside a rate increase of about 20 times was found when the pH was increased from 10 to 14. This occurred both with and without the presence of molecular oxygen.

Primary reaction pathways which are consistent with both the current work and the literature are presented. In reaction both with and without oxygen the rate-determining step is believed to be the reaction of the carbinolate anion of methyl- β -glucoside which is in equilibrium in an alkaline medium. The current work defines unique rate-determining steps within each of the proposed pathways.

INTRODUCTION

GENERAL DISCUSSION OF THE PROBLEM

DEGRADATION OF POLYSACCHARIDES IN AN ALKALINE MEDIUM

It is widely accepted that the over-all degradation of polysaccharides in an alkaline medium is brought about by two primary reaction pathways:

1. The "peeling" reaction which may be defined as a progressive, stepwise degradation of the polysaccharide molecule starting at the reducing end and removing one sugar residue at a time. This reaction must proceed from the reducing end because the aldehyde group must be free to migrate before a β -elimination of the remainder of the molecule can take place (1).
2. Random cleavage of the glycosidic bonds (2-4).

The relative importance of the above two degradation pathways under various conditions similar to those employed in commercial alkaline pulping has not been established. Lindberg, *et al.*, (5+7) have obtained a single 4-point line characterizing the rate of decomposition of methyl- β -glucoside in 10% sodium hydroxide at 170°C.

Much work has been done, however, on the reaction of simple sugars in an alkaline medium at less severe conditions (8). From this study and from the work of MacLaurin (9), it appears that the kinetic rate characterizing the alkaline "peeling" reaction is very much greater than that of the random cleavage of glycosidic bonds under similar conditions.

The over-all degradation of 4-O-methylglucuronoxylan in a homogeneous alkaline medium at elevated temperatures has been studied by several workers (2-4).

Other investigators have studied the alkaline degradation of cellulose which is normally heterogeneous (10-13). When comparing reaction rates of these two systems it should be remembered that in the heterogeneous system diffusion rather than the chemical reaction could be rate determining. When rates were obtained in the above studies they were found to be very much greater than the rate of decomposition of the methyl glycosides obtained by Lindberg, et al., under similar conditions. It should be noted that while the rate of random cleavage of glycosidic bonds appears to be much less than the rate of "peeling" or the over-all degradation rate, this is not a basis for considering random cleavage negligible. Richards (14) has shed light upon this particular point. As he increased the reaction temperature toward 170°C. in his experiments with cellulose in an alkaline medium he noted both a markedly decreasing D.P. and an increasing carboxyl content at the same yield. He then postulated that the decrease in D.P. was caused by an increase in the rate of the random cleavage of glycosidic bonds and that the increase in carboxyl content was caused by a subsequent "peeling" of the newly created reducing polysaccharide chain ends. When viewed in conjunction with the probable subsequent "peeling" reaction, the rate of random cleavage does not need to be large to be significant to the over-all rate of degradation.

IMPORTANT VARIABLES IN POLYSACCHARIDE DEGRADATION IN AN ALKALINE MEDIUM

It appears that there are three primary variables in the degradation reaction of polysaccharides in an alkaline medium: (1) reaction temperature, (2) the concentration of molecular oxygen, and (3) the hydroxyl ion concentration.

Acidic hydrolysis proceeds via random attack at any reactive point along a polysaccharide chain resulting in a rapid decrease in D.P., as estimated by

viscosity, but not in the per cent polymer recovered* throughout a wide temperature range. The D.P. in an alkaline medium at 100°C., however, apparently remains constant though considerable polymer loss is incurred (4,15-19). This type of degradation is thought to proceed primarily via the "peeling" reaction. Also, this rather constant product average D.P. may be in part an artifact created by the loss of very short-chain polymers during the usual analysis procedures. At higher reaction temperatures the over-all degradation of polysaccharide molecules can no longer be explained solely in terms of the "peeling" reaction. At about pH 12.5 and higher both a loss in per cent polymer recovered and a decrease in average D.P. are incurred as the temperature is increased to the vicinity of 170°C. (2-4, 10-12). This effect has been attributed to an increase in the relative importance of the random cleavage reaction as the temperature is increased.

Thompson, et al., (3) have characterized an apparent change in the rate-controlling mechanism of over-all polysaccharide degradation. They have presented an Arrhenius diagram for the rate of loss of yield of 4-O-methylglucuronoxylan cooked in approximately 6% alkali through the temperature range of 120 to 170°C. At a temperature slightly greater than 150°C. a significant increase in the rate of over-all polymer degradation appears to occur. That is, a single straight line will not adequately represent the data throughout the entire temperature range. The equation for this line, the Arrhenius equation, was derived with the assumption that a single rate-controlling reaction mechanism could be represented by a single straight line (20). This apparent change in the rate-controlling mechanism of over-all degradation may reflect a change in the mechanism of random cleavage.

*In order to remove inorganic materials the polymer product solution is usually dialyzed against water during the analysis procedure. This, however, also removes low molecular weight organic materials. Therefore, the "per cent polymer recovered" includes only polymer of D.P. greater than approximately 3 to 7.

Thompson, et al., (21) have cooked spruce at pH levels ranging from 1.5 to 12.5 and have characterized the resultant pulps quite completely. It was found that the viscosity of the pulps formed a maximum for cooks at pH 11, decreasing at higher and lower pHs. This finding is consistent with the work of Meier (2) who found that at 170°C. the D.P. of 4-O-methylglucuronoxylan remained essentially constant for 100 minutes at an initial pH of 11.4, but decreased rapidly when an initial pH of 13.4 was used. It was postulated by several investigators that the increased rate of over-all polymer degradation encountered as the pH of the reaction solution was increased above 11.0 is caused by an increase in the relative importance of the random cleavage of the glycosidic bonds.

The effect of molecular oxygen on the degradation rate of polysaccharides in an alkaline medium as described in the literature by various investigators is in conflict. Prey, et al., (22,23) working with the reaction of crude hemicelluloses in an alkaline medium have concluded that the presence of small amounts of air had no effect upon over-all degradation rates. Corbett and Richards (12) observed also that small amounts of air had little or no effect upon the degradation rate of cellulose in an alkaline medium. Bottle, et al., (24), however, have reacted amylose in an alkaline medium, establishing a very large effect of molecular oxygen on the over-all degradation rate. For example, a polymer solution with an initial intrinsic viscosity of 4.20 was reacted under an atmosphere of oxygen for one hour, yielding a product with a viscosity of 0.12. A sample of the polymer with a viscosity of 4.20 was then reacted for 41 hours under the same conditions, except that it was under a hydrogen-purged atmosphere, yielding a product with a viscosity of 3.82. In other experimental runs the degradation under an atmosphere of air is also shown to proceed much faster than under a hydrogen-purged atmosphere. It should be noted that the glycosidic bonds of

amylose are arranged in an α -1,4 configuration while those of the xylans and cellulose are β -1,4. While the analogy of these two glycosidic bonds is considered fruitful, it should be used with reservation.

In reviewing the work of these three groups of investigators it should be noted that while both Prey, et al., and Corbett and Richards were exploring the effect of an oxygen-free alkaline system of the degradation rate of polysaccharides and only considered the effect of oxygen in control samples, Bottle, et al., explicitly sought and explored the effect of oxygen on the reaction. All three groups used reaction chambers which were sealed at the start of each reaction thereby allowing only a limited, though undefined, amount of oxygen to enter into the reaction. It is probable that although the rate-escalating effect of oxygen was present in all three groups of experiments, the extent of the reaction allowed with this limited supply of oxygen could not be easily determined. Referring again to the work by Corbett and Richards, polysaccharides were exposed to an alkaline medium for four hours. At 100°C. the D.P. after reaction under a nitrogen-purged atmosphere had decreased only 1%. Reaction under air at 100°C., however, decreased the D.P. 25%. At 170°C. the D.P. was decreased 75% by reaction under either air or a nitrogen-purged atmosphere. That is, no difference in product D.P. was found at 170°C. It was then concluded that there was no effect of molecular oxygen on the degradation rate. But, it is possible that the effect was merely masked when the reaction was carried out further at the higher reaction temperature.

It is, then, suspected that the random cleavage reaction becomes relatively more important in over-all polysaccharide degradation as the temperature is increased from 100 to 170°C. or the pH is increased from 11 to 14. These effects will later be clarified. No work concerning the effect of oxygen on the rate of

cleavage of the glycosidic bond was found. And, previous work on the effect of oxygen on over-all polysaccharide degradation is in conflict. The effect of oxygen on the rate of cleavage of the glycosidic bond in alkali will be explored.

METHYL- β -GLUCOSIDE AS A REACTANT

For this study a carbohydrate reactant was required which could be used to characterize the random cleavage reaction of glycosidic bonds of polysaccharides in an alkaline medium. Methyl- β -glucoside has been widely used as a model compound representing cellulose. It contains a glycosidic bond, similar to those joining polysaccharide residues, which cannot be cleaved by the "peeling" reaction in that the sugar residue is blocked against the required prior molecular rearrangements. Another advantage of using methyl- β -glucoside rather than a polysaccharide is that the rate of decrease in yield of the glycoside is the same as the rate of cleavage. Therefore, the rate of random cleavage can be measured directly by measuring the loss of yield. Methyl- β -glucoside also has the advantage of being soluble in aqueous sodium hydroxide, greatly facilitating the analysis of unused reactant.

Most analytical procedures are accurate to within only about $\pm 2\%$. If analysis of unused reactant alone is used to characterize the reaction it must therefore proceed to at least 4 or 5% reaction before sufficient analytical accuracy is obtained. It was found, however, that methanol could be determined as a product from the alkaline reaction of methyl- β -glucoside. This allowed the characterization of the cleavage reaction at short reaction times, adding greatly to the usefulness of methyl- β -glucoside as a reactant.

Methyl- β -glucoside was, then, used as a model compound; its glycosidic bond representing the nonreducing end glycosidic bonds of polysaccharides in general.

This reaction will be referred to as the "random cleavage reaction" whether speaking of the nonreducing end cleavage of a polysaccharide or cleavage of the glycosidic bond of methyl- β -glucoside. While the absolute rate of reaction of the various glycosidic bonds will most certainly vary, the same relative trends in reaction behavior are expected no matter which of the above glycosidic bonds is in question.

CLEAVAGE OF THE GLYCOSIDIC BOND IN AN ALKALINE MEDIUM

The glycosidic bonds of polysaccharides, other than at the reducing end of the molecule where rearrangement may occur (25), are usually considered stable to reaction in an alkaline medium as established by E. Fischer (29) and since confirmed many times. Only under the drastic conditions encountered in modern alkaline pulping processes, e.g., 10% sodium hydroxide and 170°C., does cleavage of the glycosidic bond appear to occur at a measurable rate. It has been found, however, that glycosides can be modified in such a manner as to greatly increase this cleavage rate. For example, with an alkyl glycoside this may be accomplished by modification of the aglycone group, the alkyl group, to include either an unsaturated bond or an electron-accepting group (27-29). Phenyl glycosides are also more easily reacted in alkali because of the electron-accepting ability of the phenyl group. Modification of the glycon group, the sugar group, can also increase the rate of reaction in alkali. Kenner (30), in a discussion of the alkaline degradation of oxycelluloses, shows that once a carbonyl group at the 2- or 3-position of an anhydroglucose unit is formed, chain cleavage can occur by the same mechanism as the "peeling" reaction. Cleavage would then occur at a relatively rapid rate. Theander (31) has reacted the 2- and 3-keto methyl glucosides in alkali and found a reaction rate many orders of magnitude greater than for the corresponding unoxidized methyl glucoside.

The mechanism of cleavage of the glycosidic bond in an oxygen-free alkaline medium proposed by McCloskey and Coleman (32) working with phenyl- β -glucoside has since found wide acceptance. This reaction pathway is illustrated in Fig. 1 using methyl- β -glucoside, (I), as the reactant. The first step in the reaction sequence is the formation of the secondary carbinolate anion, (II), which is in equilibrium with (I). Although the hydroxyl group at C-2 is much more reactive than at C-3 (33), its ionization constant is still thought to be quite small (34), i.e., in the range of 10^{-14} . The concentration of (II) will in turn be dependent upon the concentration of hydroxyl ions and the concentration of (I). The rate-determining step, noted as R.D., is postulated as the reaction of (II) to form 1,2-anhydro- α -D-glucopyranose, (III), since the rate of decomposition of (III) has been established as very rapid (35,36). The formation of (III) is thought to involve the nucleophilic displacement of the aglycon group by the neighboring secondary carbinolate anion at C2. This anion, then, attacks the trans-glycosidic oxygen intramolecularly with an inversion eliminating a methoxyl ion and forming (III). Because of its comparatively rapid rate of reaction, (III) has not been isolated from reaction products.

Evidence pointing to the existence of (III) as an intermediate has been obtained, however. Anchimeric assistance of the C-2 hydroxyl group is indicated from work by Bardolph and Coleman (35). They have shown that when phenyl 2-O-methyl- β -D-glucopyranoside is treated for 48 hours in an alkaline medium at 100°C. it can be recovered in 92% yield. When, however, the phenyl- β -D-glucopyranoside is treated under the same conditions it is almost entirely converted to levoglucosan in a much shorter reaction time (36). Work by Gasman (37) has shown similar results. All of this work indicates that cleavage of the glycosidic bond of phenyl glucosides in an alkaline medium involves anchimeric assistance from the

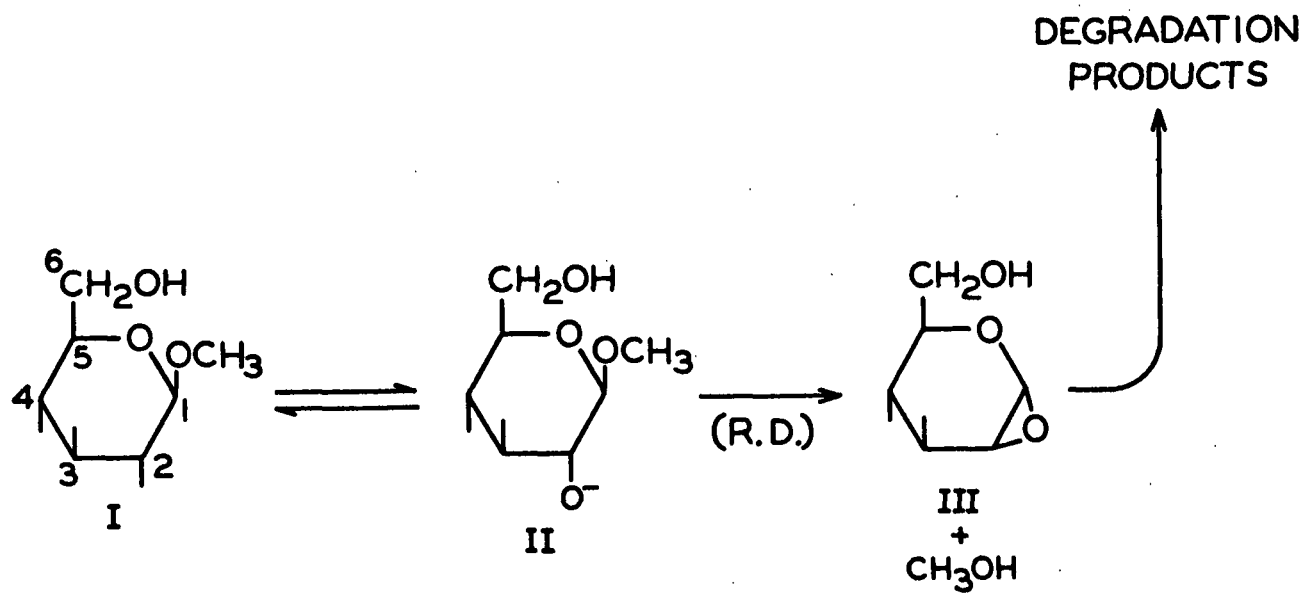


Figure 1. A Probable Reaction Pathway Leading to Cleavage of the Glycosidic Bond of Methyl-β-Glucoside in an Oxygen-Free Alkaline Medium

C-2 hydroxyl group. Janson and Lindberg (7) concluded that a similar anchimeric assistance was present in the alkaline reaction of methyl glucosides. They found that the reaction rate of methyl 2-O-methyl- β -D-glucopyranoside is less than half that of methyl- β -D-glucopyranoside in a similar medium.

Evidence confirming the inversion step in the formation of (III) has also been found. Lindberg, *et al.*, (6,38,39) have obtained kinetic data on the rate of reaction of the more common naturally occurring sugars in both the methyl- α - and the methyl- β -glycopyranoside form in an alkaline medium. In all cases the glycoside which had the glycosidic oxygen and the C-2 hydroxyl group in a trans-configuration reacted much faster than did the corresponding cis-isomer. Ballou (40) and McCloskey and Coleman (32) each have reported similar results in the alkaline degradation of phenyl glycosides. These investigations, then, both confirm the presence of an inversion step in the formation of (III) and also indicate anchimeric assistance of the C-2 hydroxyl group.

Bardolph and Coleman (35) in further work have started with a model for the postulated intermediate and obtained the normal reaction product. They have reacted 3,4,6-tri-O-acetyl-1,2-anhydro- α -D-glucopyranose in an alkaline medium at relatively low temperatures obtaining 1,6-anhydro- β -D-glucopyranose in large yield.

This review presents evidence for both the inversion mechanism and the existence of the 1,2-anhydride intermediate, (III), in random cleavage of the glycosidic bond in an oxygen-free alkaline medium.

Reaction of the glycosidic bond in an alkaline solution involving molecular oxygen has not been studied. A mechanism based on the current experimental work and incorporating work from the literature in related areas is later presented in the Discussion section.

EXPERIMENTAL

SYNTHESIS OF METHYL- β -GLUCOSIDE

The synthesis of methyl- β -D-glucopyranoside from glucose is presented in three parts: (1) Preparation of acetobromoglucose from glucose. (2) Preparation of methyl- β -glucoside tetraacetate from acetobromoglucose. (3) Deacetylation of methyl- β -glucoside tetraacetate.

PREPARATION OF ACETOBROMOGLUCOSE FROM GLUCOSE

The method of Barczai-Martos and Korozy (41) was followed in the preparation of 2,3,4,6-tetra-O-acetyl- α -D-glucosyl bromide from D-glucose. This method utilizes perchloric acid as a catalyst in the acetylation reaction. With this catalyst, acetic anhydride alone, without the commonly used glacial acetic acid, was found to be quite effective in the acetylation of glucose. Hydrogen bromide is formed in the reaction mixture by adding phosphorous, bromine, and water in that order. This compound then reacts with the previously acetylated glucose to give acetobromoglucose. Details of the synthesis are outlined in Appendix I. Barczai-Martos and Korozy state that the acetobromoglucose product is pure enough to use as a reactant in further synthesis without crystallization. In this work the acetobromoglucose was not crystallized. The over-all results were satisfactory.

PREPARATION OF METHYL- β -GLUCOSIDE TETRAACETATE FROM ACETOBROMOGLUCOSE

A method similar to that published by Reynolds and Evans (42) was used in the conversion of the acetobromoglucose to methyl 2,3,4,6-tetra-O-acetyl- β -D-glucopyranoside. The key point in this method is the exclusion of water from the reaction system. If water is present in the system, the water will react with acetobromoglucose simultaneously with the desired reaction of acetobromoglucose

with methanol, decreasing the yield of the glycoside considerably. The addition of an internal desiccant, 10/20 mesh nonindicating Drierite, to the system decreased greatly the amount of water in the system, thereby decreasing the reaction of acetobromoglucose with water. The crystalline intermediate methyl- β -glucoside tetraacetate was obtained in all syntheses. The details of this synthesis procedure are given in Appendix I.

DEACETYLATION OF METHYL- β -GLUCOSIDE TETRAACETATE

Deacetylation of the methyl- β -glucoside tetraacetate was accomplished in methanol, using a trace of sodium methylate as a catalyst (43). Methyl- β -D-glucopyranoside hemihydrate was obtained as the crystalline product. The deacetylation procedure used is also outlined in Appendix I.

PURITY OF METHYL- β -GLUCOSIDE

During the course of the experimental work, two separate batches of methyl- β -glucoside were synthesized. Each batch consisted of the product of several syntheses, recrystallized in one container in order to obtain a large amount of homogeneous material. The melting point of the first batch was 105.5 to 107.5°C. and the second 106 to 108°C. The optical rotation of the first batch was -32.2 degrees and the second batch -32.4 degrees, both in aqueous solution at 25°C. using a sodium light source. Literature values for these physical constants range from 104 to 111°C. for the melting point and from -32.2 to -32.9 degrees for the optical rotation under similar conditions (44).

The purity of this recrystallized methyl- β -glucoside mixture was established chromatographically on Whatman No. 1 filter paper. A 3:1:1 ethyl acetate:acetic acid:water developer was used. Methyl- β -glucoside moves about three times faster than glucose in this developer, giving excellent separation of expected contaminants.

Spots were developed by dipping in an acetone solution of silver nitrate, then in an alcoholic solution of sodium hydroxide, and lastly in a solution of sodium thiosulfate (45). For comparative purposes a series of known glucose spots were developed in the concentration range of 0.1 to 100 γ . The smallest detectible spot was found to be 0.3 γ . A series of known methyl- β -glucoside spots was also developed, covering a concentration range of 0.5 to 1000 γ . The 1000- γ spot was found to move compactly. A 1000- γ sample of the synthesized methyl- β -glucoside was then chromatographed to determine its purity. From this a 20- γ spot of glucose was obtained, no other impurities being visible. This concentration was obtained by comparison of the spot size to that of a series of known glucose spots of varying concentration developed in a similar manner. The synthesized methyl- β -glucoside contained, then, approximately 2% glucose. Both batches were very similar. An over-all yield of approximately 50% was obtained. Since the methyl- β -glucoside was to be reacted in an alkaline medium, the reactant was purified by dissolving it directly in the reacting medium, usually 10% sodium hydroxide, heating for 20 minutes at 90°C. and cooling to room temperature.* The optical rotation of this material was then used to characterize zero reaction time. No change in optical rotation of the methyl- β -glucoside was incurred through this purification process.

REACTION OF METHYL- β -GLUCOSIDE IN AQUEOUS SODIUM HYDROXIDE

REACTION APPARATUS

An automatically controlled oil bath of about 75-gallon capacity was used to obtain a stable reaction temperature. Quenching was accomplished by removing a

*A common purification procedure used to remove the glucose consists of heating the material for 20 minutes at 90°C. in 1N sodium hydroxide, followed by ion exchange with first acidic and then basic resins to remove the alkali and the acidic fragments formed from the destruction of the glucose (46).

reaction tube from the oil bath and placing it into a kerosene-filled tube approximately 8 inches in diameter immersed in a moving stream of water maintained at about 15°C. The measured reaction time was started with the introduction of the reaction tube into the oil bath, which was preheated to the desired temperature. The bath temperature, as regulated by the automatic controller, remained constant within the limits of $\pm 1^\circ\text{C}$. during any given experimental run. The reaction tubes were fabricated from type 316 stainless steel pipe 1/4 inch I.D. by 4 feet. Both ends of each tube were threaded and fitted with caps of the same material. Regal K heat transfer oil, a product of Texaco, was used as the heating medium. Measurements, using a thermocouple, showed that the contents of the reaction tube were within 2°C. of the bath at 170°C. in 2.9 minutes. Reaction time was stopped at the time the reaction tube was placed into the quenching apparatus. The quenching system was found to lower the temperature of the contents of the reaction tube to room temperature in 2.0 minutes.

REACTION CONDITIONS

Most of the experiments were run in 10% aqueous sodium hydroxide calculated as 10% by volume at the reaction temperature so that the concentrations at the various temperatures would be the same (Appendix III). This concentration level provided a large excess of alkali (Appendix III). Two runs were made in a buffered system of pH 10.2 measured at 20°C. The buffered system consisted of 1 g./l. sodium hydroxide, 9.6 g./l. sodium bicarbonate, and 60.5 g./l. sodium carbonate. The pH remained constant throughout a given run.

A methyl- β -glucoside concentration of 22.2 g./l. at 20°C. was used in all runs. No dilution was incurred in removing the solution from the reaction tubes. Each data point was obtained from the analysis of the entire contents of one reaction tube, i.e., aliquots were not taken from a single reaction tube at various

reaction times. In runs where a comparatively oxygen-free system was desired a system of nitrogen purging was established (Appendix IV).

In reactions involving molecular oxygen, the molar ratio of molecular oxygen to methyl- β -glucoside at zero reaction time is used to estimate the amount of oxygen present. The oxygen content of the system was calculated by measuring the void volume in the reaction tube after the reactant solution was charged and calculating the number of moles present. Several molar ratios of oxygen to methyl- β -glucoside were used. This was done to get a better understanding of the effect of oxygen on the reaction rate. To obtain the molar ratio of 0.063 a solution volume of approximately 45 ml. was used with a reaction atmosphere of about 40 ml. of air. The exact volume of solution used was governed by the exact size of the reaction tube. To obtain the molar ratio of 0.24, the solution volume was decreased to about 25 ml., the reaction atmosphere still being air. In order to further increase this ratio to 1.1, the solution volume was maintained at 25 ml. but the reaction atmosphere was changed to pure oxygen by purging the reaction tube with oxygen after the reaction solution had been added.

ANALYSIS OF REACTION PRODUCT SOLUTIONS

Reaction solutions were not neutralized but left in the alkaline condition throughout the analysis procedure. It was found that methyl- β -glucoside is stable in 10% sodium hydroxide at room temperatures for a four-week interval as measured by both methanol analysis and optical rotation. Neutralization was then not required since the analytical procedures did not require heating.

POLARIMETRIC ANALYSIS

It is important that any optically active products be accounted for in that the concentration of unused reactant was measured by polarimetric analysis of the

raw product solution. When the glycosidic bond of methyl- β -glucoside is cleaved in an alkaline medium the most probable immediate product from the glycone group is either one or more saccharinic acids or an internal glycoside. Drysilius, et al., (6) found that the neutral products from the reaction of methyl- β -glucoside at 170°C. consisted of unchanged starting material, with only traces of other components such as 1,6-anhydroglucopyranose and 1,6-anhydrogalactopyranose, as demonstrated by comparative paper chromatography.

The alkaline reaction products from various hexoses have been characterized in some detail (8,28) and found to be very predominantly acidic, and to cleave into shorter chains as the reaction conditions become harsher. Green (47) formed the anilide of the saccharinic acids from kraft black liquor and separated them by paper chromatography. He found that only about 10% were six-carbon acids, the remainder being shorter-chained. The largest single spot obtained was that of lactic acid.

As illustrated in Tables IV and V of Appendix V the specific rotation of the product solutions did not change significantly after deionization with both acidic and basic ion-exchange resins. This result coupled with the fact that only a trace amount of the fragmentation products are neutral provides strong evidence that polarimetric analysis of the raw product solution is a valid measure of the concentration of unused methyl- β -glucoside. Also, the concentrations so obtained agree stoichiometrically with the product methanol concentrations, further illustrating the validity of this measurement.

The specific rotation of methyl- β -glucoside was found experimentally to be constant in aqueous sodium hydroxide from a pH of 7 to 14. Salt concentration, using sodium chloride, was also found to have no effect on the specific rotation. These findings are in accord with those of Reeves (48).

The unused methyl- β -glucoside concentration was determined polarimetrically, including samples at zero reaction time. The accuracy of this procedure was within $\pm 2\%$ of the unused reactant as determined from repeated statistical analysis of multiple observations.

METHANOL ANALYSIS

Because the polarimetric analysis is accurate to within only $\pm 2\%$ of the methyl- β -glucoside present, the reaction must proceed to at least 4 or 5% before the rate can be characterized by this method. Reaction rates at small concentration change were obtained, then, only by methanol analysis.

The colorimetric methanol analysis procedure of Boos (49) was adapted for use. It was found that salt interfered with light transmission through the sample and that unused reactant and/or other products tended to produce extraneous color during the methanol analysis. Therefore, all samples were distilled under a high vacuum prior to methanol analysis. The colorimetric methanol analytical procedure employed in the experimental program and a drawing of the distillation equipment used are presented in Appendix VI. Confidence limits for the procedure and for the distillation followed by colorimetric analysis are also included in Appendix VI. For the total methanol analytical procedure, including distillation, the confidence is $\pm 5\%$ of the methanol present in the reaction mixture. It was found that 1 mole of methanol was produced per each mole of methyl- β -glucoside reacted. This stoichiometric relationship was then used to calculate the equivalent moles of unreacted material plotted in Fig. 2 through 23. The actual amount of methanol produced may be seen in the tables of Appendix VIII. Methanol was found to be stable under the reaction conditions employed as shown in Fig. 14 of Appendix VI.

PRESENTATION OF RESULTS

From kinetic studies of phenyl- β -glucoside in an alkaline medium (50) and similar reactions (51) it was anticipated that the alkaline reaction of methyl- β -glucoside would exhibit second-order kinetics. The rate of reaction would then depend upon the concentration of reactant and of hydroxyl ions. However, because hydroxyl ion concentration in the current experimental work does not vary appreciably throughout a given reaction, this concentration can be combined with the second-order rate constant into what is normally called a pseudo first-order rate constant. All of the reactions run in a nitrogen-purged system were found to be pseudo first order, i.e., when the log of the unused reactant concentration was plotted vs. the reaction time, a straight line was obtained. The reactant concentration at zero reaction time was converted to a basis of 10 moles per liter. All later reactant concentrations were obtained by multiplying the per cent unreacted material by 10 and dividing by 100. This process in no way changed the experimental results and facilitated the calculation of logarithms.

The rate at which the glycosidic bond of methyl- β -glucoside is cleaved in 10% sodium hydroxide under nitrogen-purged conditions from 170 through 140°C. is illustrated in Fig. 2. The reaction rate constants which characterize these lines are listed in Table I. No rate constants are presented for the reactions run with initial oxygen to methyl- β -glucoside ratios of 0.063 and 0.24 because the loss of reactant with time is not linear on a semilog plot (Fig. 5), i.e., pseudo first-order conditions are not met. Data points are omitted from Fig. 2 in the interest of clarity. These lines with data points included are shown in Fig. 7; 15, 18, 19, 20, 22, and 23; all except Fig. 7 are in Appendix VIII. In each of these figures the lines are determined by both methanol and polarimetric analysis. Since a straight line running through zero product concentration at zero reaction

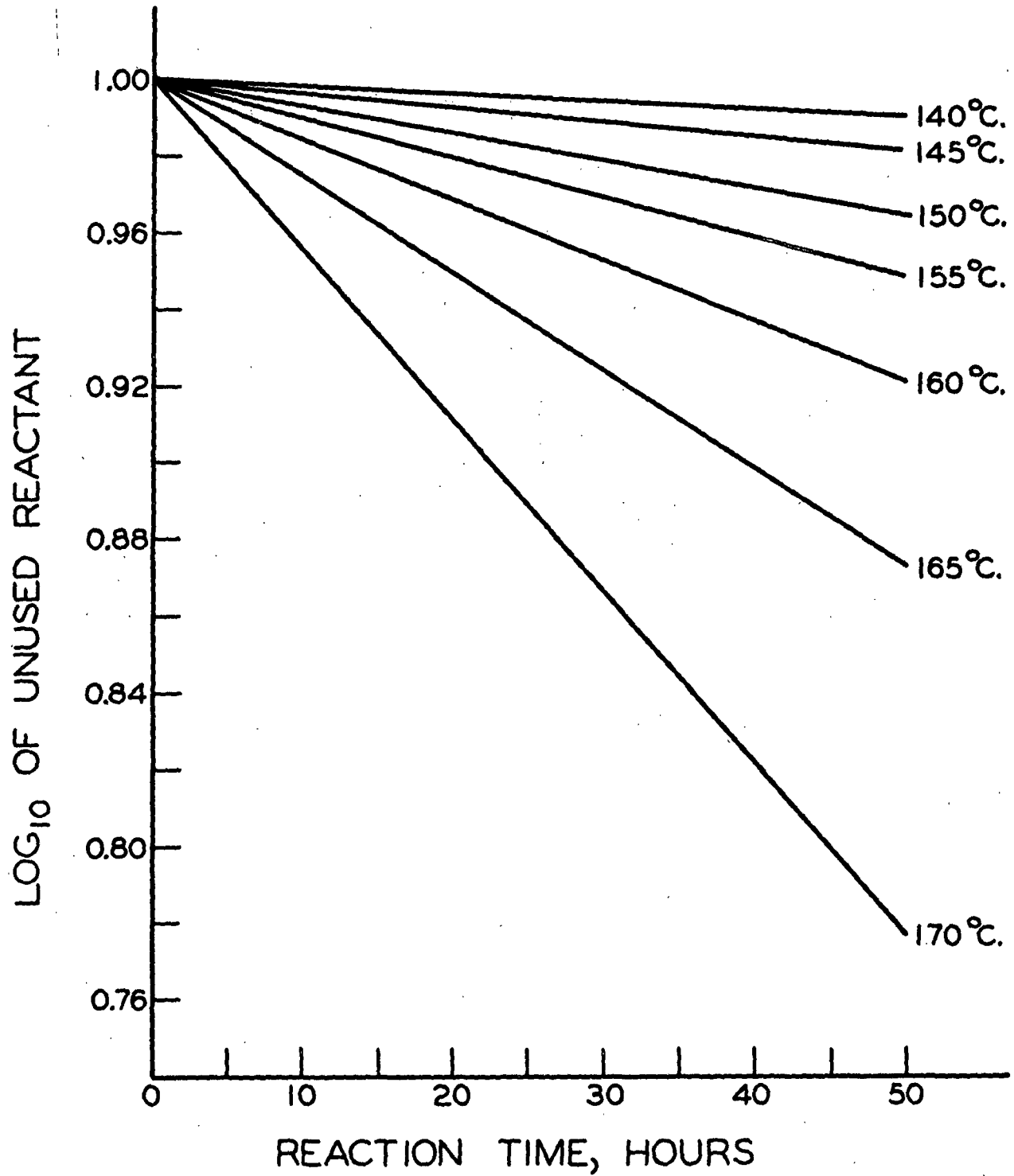


Figure 2. Summary of the Reaction of Methyl- β -Glucoside in 10% Sodium Hydroxide Under Nitrogen-Purged Conditions at All Reaction Temperatures Used

time was obtained in all runs with prior nitrogen-purging, the effects of residual oxygen are considered negligible. The rate of reaction varies with reaction temperature in a predictable manner.

TABLE I
SUMMARY OF PSEUDO FIRST-ORDER REACTION RATE CONSTANTS^a

Reaction Temperature, °C.	N ₂ -Purged Conditions in 10% NaOH	N ₂ -Purged Conditions with pH = 10.2	O ₂ /MBG = 1.1 at Zero Time in 10% NaOH	O ₂ /MBG = 1.1 at Zero Time with pH = 10.2
140	0.00020	-	-	-
145	0.00038	-	-	-
150	0.00073	0.000033	0.033	0.0021
155	0.0010	-	-	-
160	0.0016	-	-	-
165	0.0026	-	-	-
170	0.0044	-	0.080	-

^aRate constants are expressed in common logs with units of reciprocal hours.

This is also illustrated in the corresponding Arrhenius plot, Fig. 3. The significance of this plot will be discussed later. The linear correlation coefficient for this line establishes its linearity with a very high degree of confidence, as shown in Appendix VII.

Figure 4 illustrates the effect of molecular oxygen on the rate of cleavage of the glycosidic bond of methyl- β -glucoside in 10% sodium hydroxide at 170°C. This shows that the reaction rate is dependent not only upon the concentration of methyl- β -glucoside and hydroxyl ion, but also upon the concentration of molecular oxygen. When oxygen is present initially the reaction proceeds at a much faster rate at short reaction times, decreasing to a rate equal to that obtained

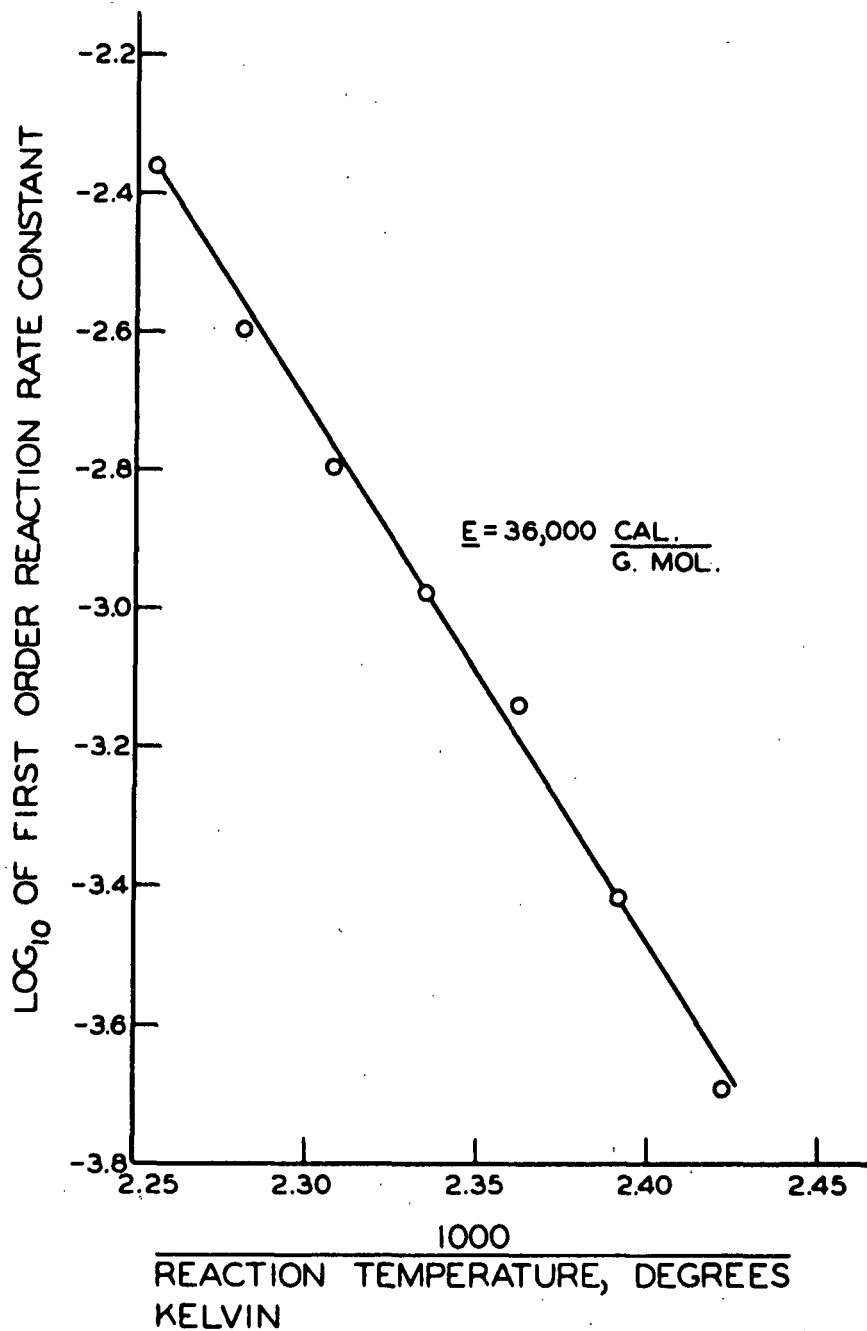


Figure 3. Arrhenius Plot for the Reaction of Methyl- β -Glucoside in 10% Sodium Hydroxide Under Nitrogen-Purged Conditions

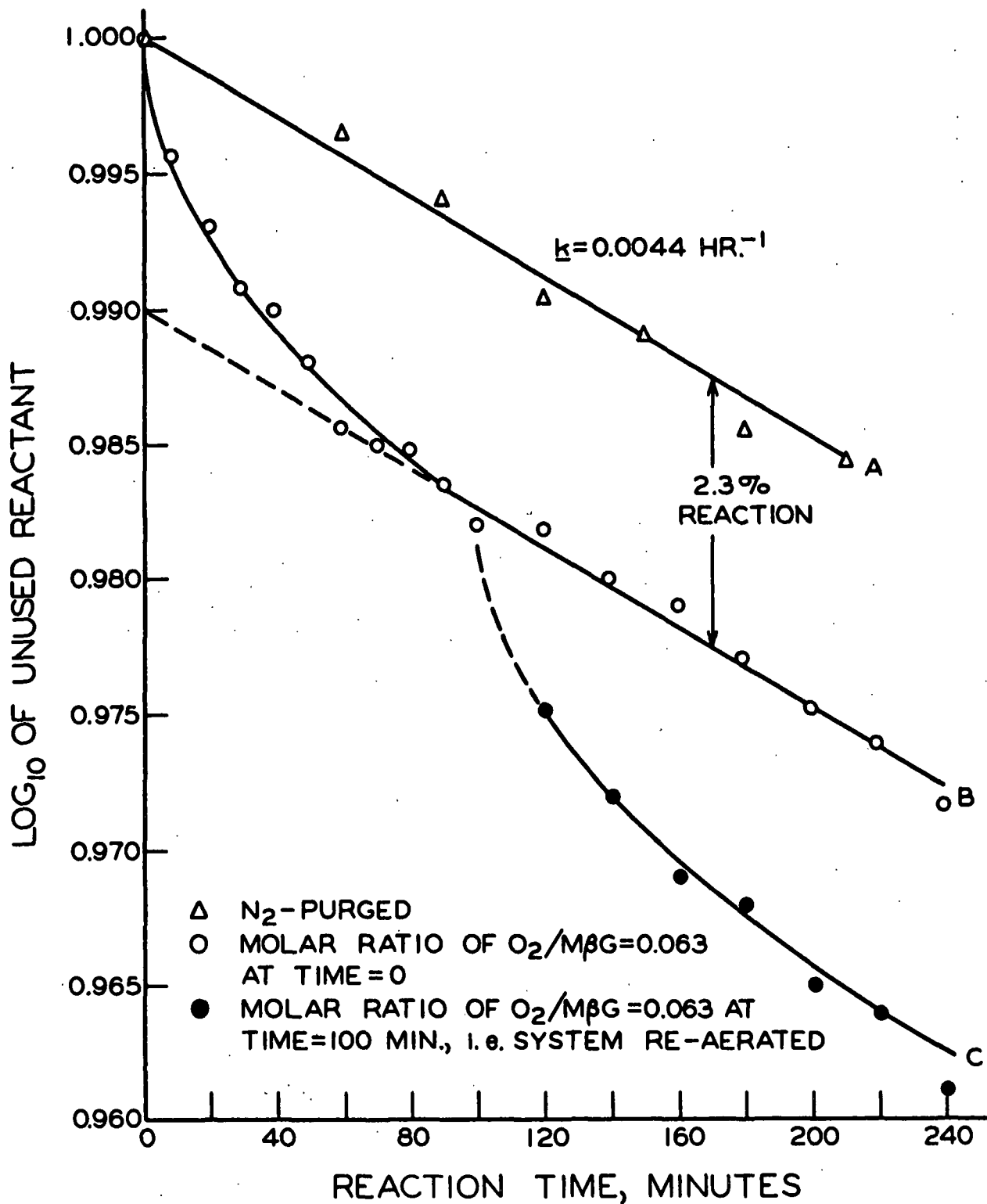


Figure 4. The Initial Reaction of Methyl- β -Glucoside in 10% Sodium Hydroxide at 170°C. Characterized by Methanol Analysis

under the corresponding nitrogen-purged conditions at long reaction times. The immediate effect of oxygen is demonstrated throughout the experimental work, e.g., the more rapid rate of reaction characterized by line B of Fig. 4 at less than 100 minutes' reaction time, when compared to the nitrogen-purged system characterized by line A. Figures 15 and 16 of Appendix VIII form parallel extensions of lines A and B, respectively, of Fig. 4 out to 50 hours' reaction time. The only plausible explanation of this phenomenon is that the molecular oxygen was used up at about 100 minutes' reaction time.

In Fig. 4 an estimate of the moles of molecular oxygen used per mole of methyl- β -glucoside reacted by a pathway involving oxygen was obtained by extrapolating back to the ordinate the flat portion of the curve, which occurs after the free molecular oxygen has apparently been used up. This extrapolation was also carried out in Fig. 5, 6 and 8.

To confirm that the rate increase was caused by the presence of oxygen in the reaction system and to more firmly establish the reproducibility of this phenomenon, seven identical paired samples of methyl- β -glucoside in 10% sodium hydroxide were reacted at 170°C. for 100 minutes, each sample in a separate reaction tube. All samples were then quenched to room temperature. One reaction tube of each pair was opened, emptied, purged with fresh air, refilled with its original contents and resealed. The paired tubes, one now re-aerated and one not, were then cooked for various lengths of time and analyzed for methanol. The results shown by lines B and C of Fig. 4 show that the effect of molecular oxygen on the rate of cleavage can be repeated.

The large effect of molecular oxygen concentration on the reaction rate is illustrated in Fig. 5. The general reaction conditions are the same as those of Fig. 4, but here four different concentrations of molecular oxygen were used;

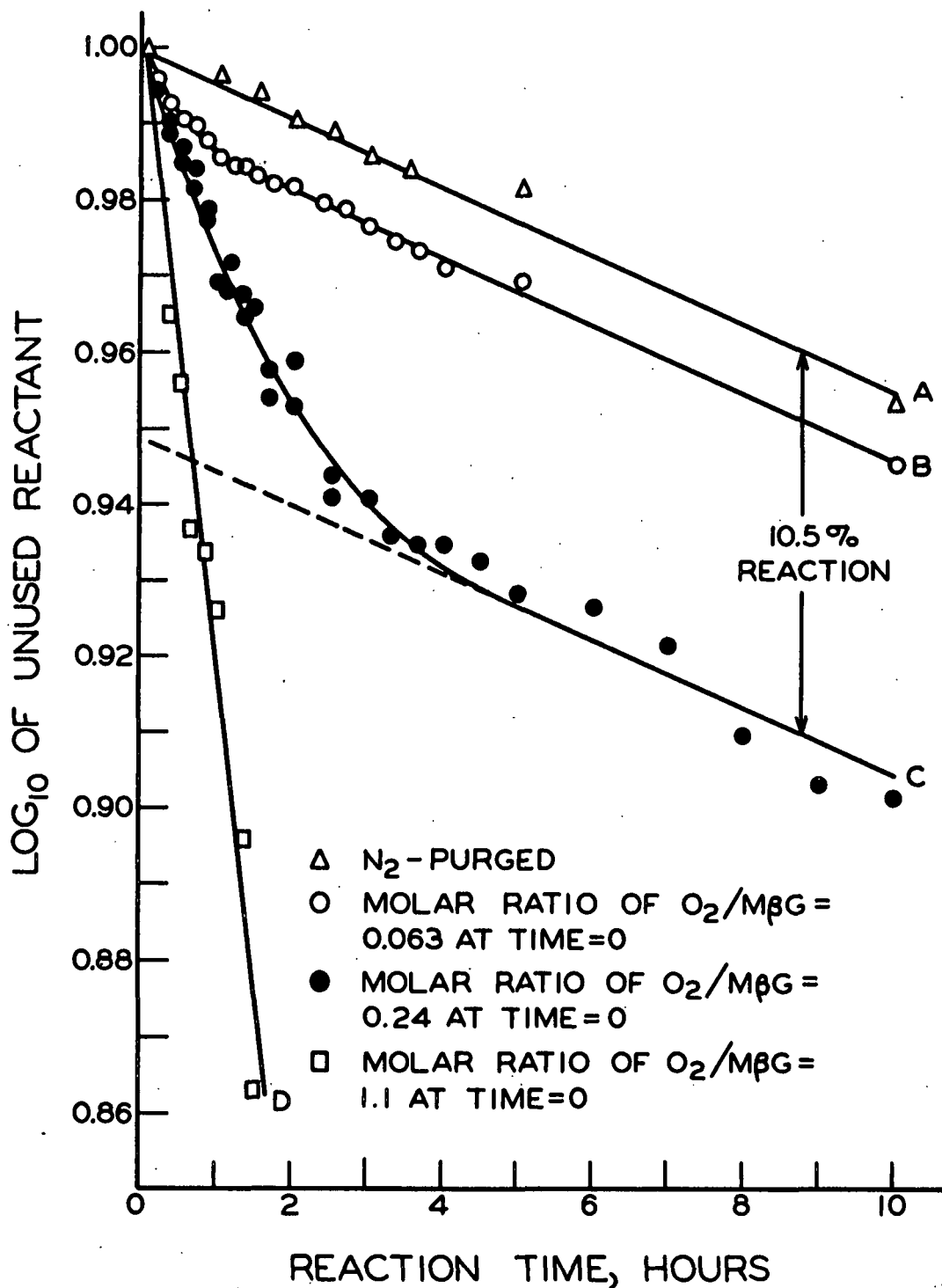


Figure 5. The Reaction of Methyl-β-Glucoside in 10% Sodium Hydroxide at 170°C. Characterized by Methanol Analysis

a nitrogen-purged system, Curve A, and systems containing 0.063, 0.24, and 1.1 moles of molecular oxygen per mole of methyl- β -glucoside at zero reaction time, Curves B, C, and D, respectively. In the reaction characterized by Curve C, molecular oxygen appears to have been consumed in about 4 hours. Curve D appears to be a straight line over the reaction time interval used. This can better be seen in Fig. 17 of Appendix VIII. The pseudo first-order rate constant calculated from this line includes constants representing both the hydroxyl ion and the molecular oxygen concentrations. The rate calculated from Curve D is approximately 18 times that calculated from Curve A.

The effect of molecular oxygen on the reaction rate of methyl- β -glucoside at 150°C. in 10% sodium hydroxide at short reaction times is illustrated in Fig. 6. In general, the same effect of molecular oxygen on the rate of reaction is seen at 150°C. as was seen at 170°C. in Fig. 4.

The effect of hydroxyl ion concentration on the rate of the reaction of methyl- β -glucoside at 150°C. under nitrogen-purged conditions is shown in Fig. 7. In the reaction system represented by Line A, the pH was buffered to a constant value of 10.2. The rate constant obtained under these conditions is also pseudo first order since the hydroxyl ion concentration is held constant by the action of the buffer. In the system represented by Line B the concentration of sodium hydroxide was 10% at zero time, equivalent to a pH of about 14, and remained essentially constant throughout the reaction. The effect of hydroxyl ion concentration on the reaction rate is large, the rate at the higher hydroxyl ion concentration being about 22 times that at the lower concentration.

The effect of hydroxyl ion concentration on the rate of reaction at 150°C. with an oxygen to methyl- β -glucoside molar ratio of 1.1 at zero reaction time is shown in Fig. 8. Again the hydroxyl ion concentration has a pronounced effect

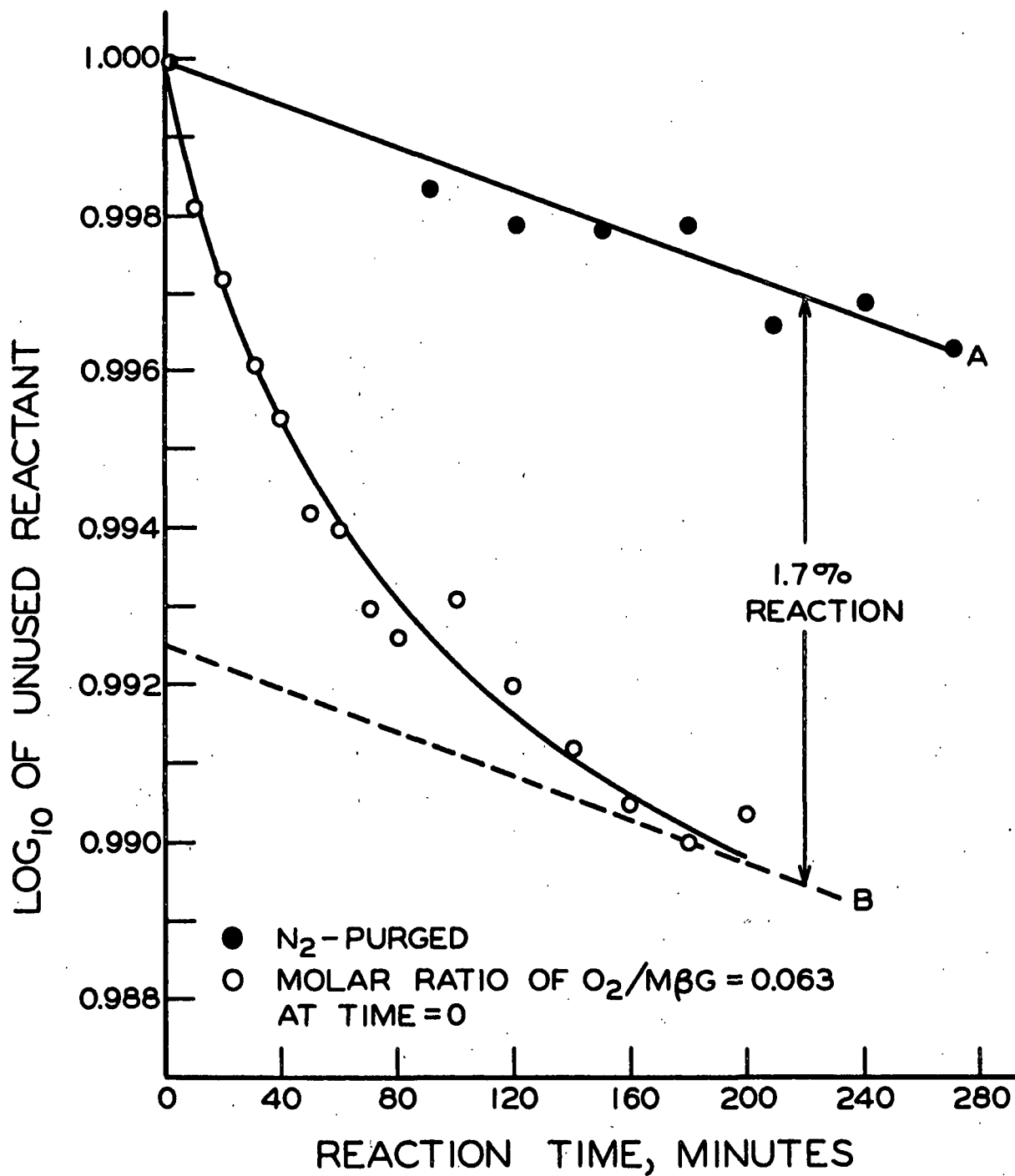


Figure 6. The Initial Reaction of Methyl-β-Glucoside in 10% Sodium Hydroxide at 150°C. Characterized by Methanol Analysis

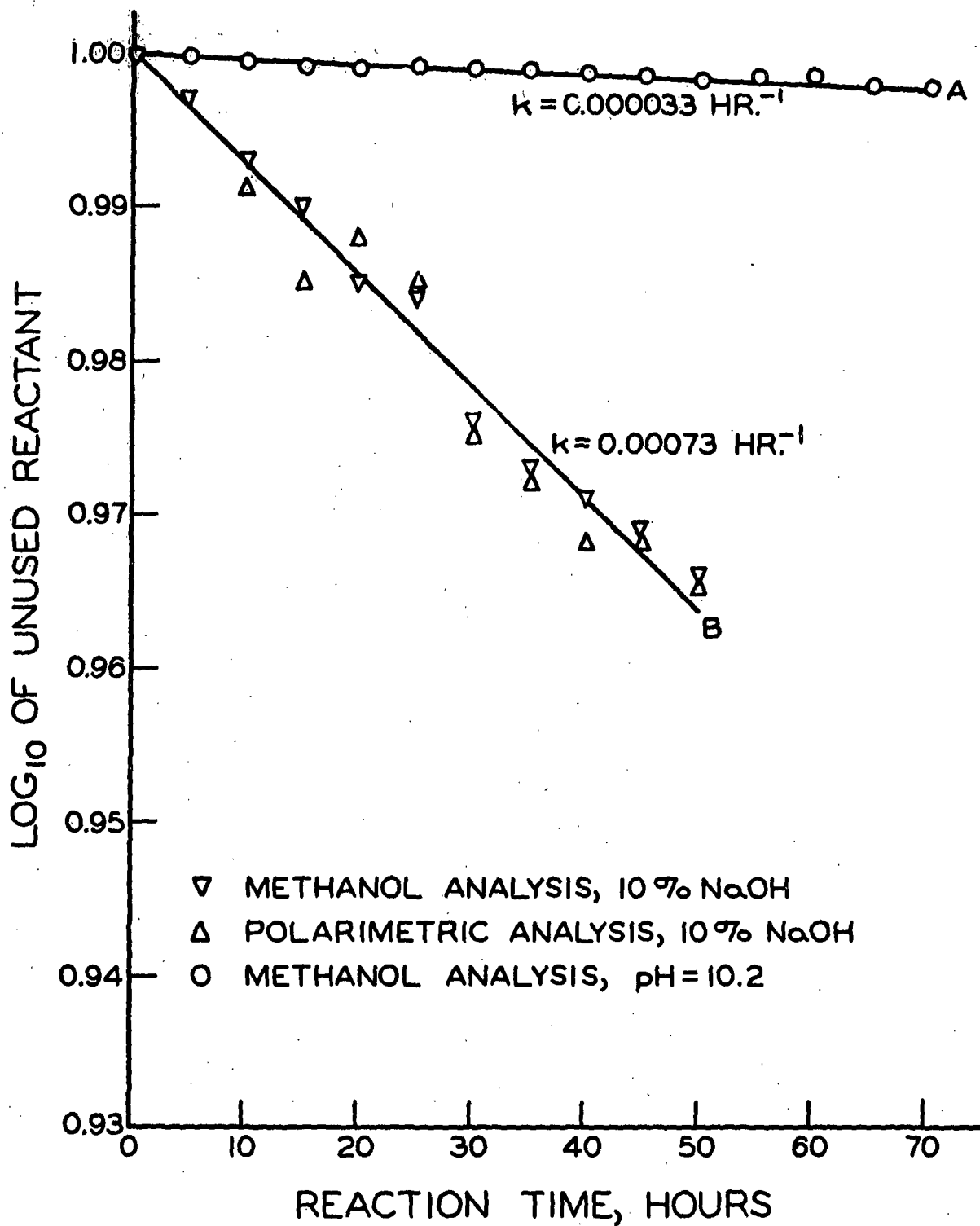


Figure 7. The Reaction of Methyl-β-Glucoside at 150°C. Under Nitrogen-Purged Conditions

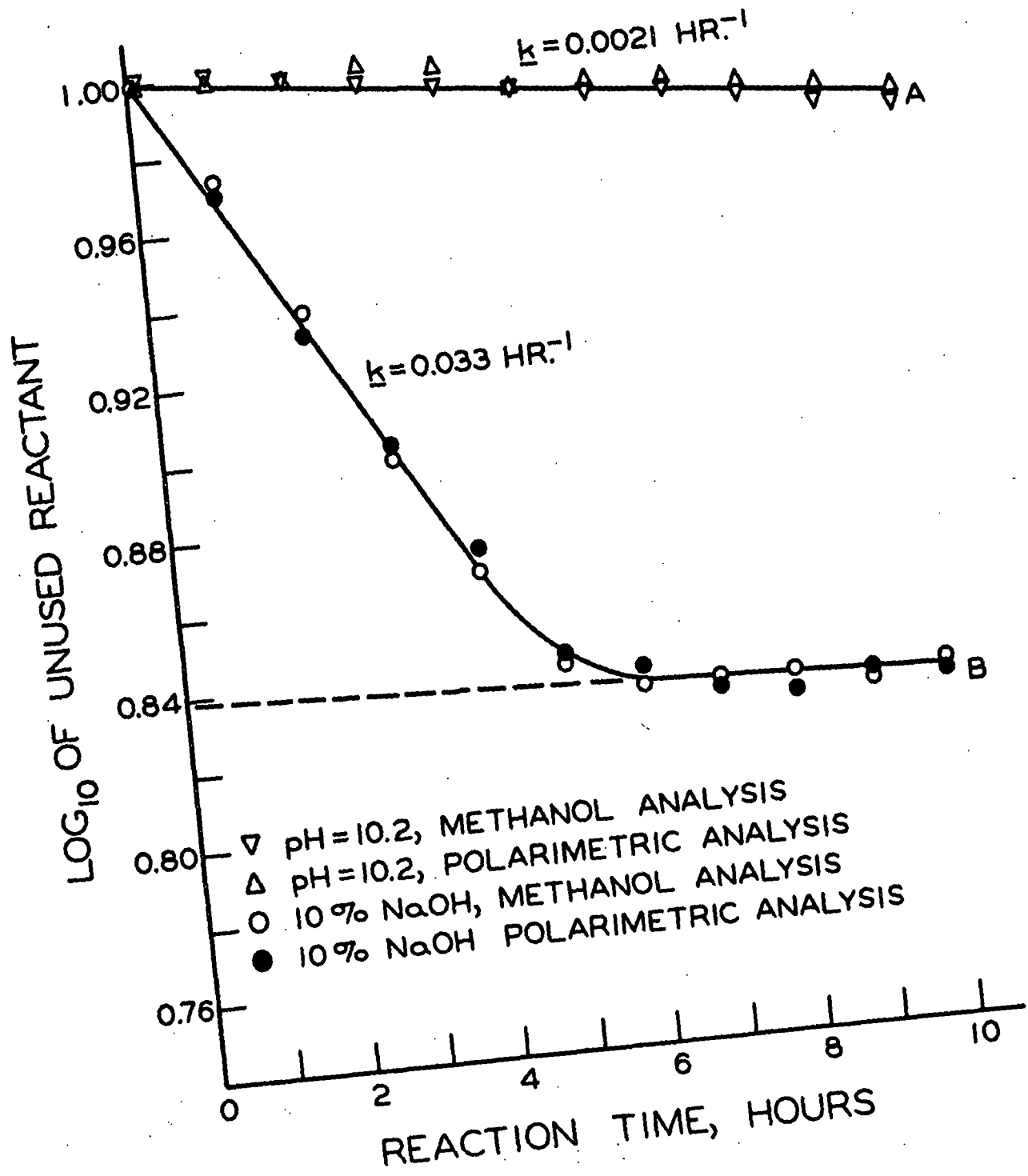


Figure 8. The Reaction of Methyl- β -Glucoside at 150°C. with a Molar Ratio of Molecular Oxygen to Methyl- β -Glucoside of 1.1 at Zero Reaction Time

upon the reaction rate, the rate with 10% sodium hydroxide present (Line B) being about 16 times faster than the rate at pH 10.2 (Line A). Apparently, the reaction illustrated by Line B consumed all of the available molecular oxygen in about 5 hours.

In Fig. 9, which is a combination of Fig. 7 and 8, the relative effect of the concentrations of hydroxyl ion and molecular oxygen on the rate of cleavage of the glycosidic bond is illustrated. Lines A and B represent reactions in a nitrogen-purged atmosphere and Lines C and D represent the reaction in an atmosphere which includes molecular oxygen. The point of interest is that the reaction represented by Line C has proceeded at a rate approximately 3 times as fast as that represented by Line B, while the former reaction was run at a pH of 10.2 and the latter in 10% sodium hydroxide, approximately pH 14. This finding is consistent with the postulated reaction pathways and again points out the very large effect of molecular oxygen on the rate of cleavage of the glycosidic bond.

Figure 9 also shows that the effects of hydroxyl ion concentration and oxygen on the reaction rate are additive. Line A represents reaction of methyl- β -glucoside at pH 10.2 under a nitrogen-purged atmosphere at 150°C. The rate of the reaction represented by Line B has been increased over Line A by increasing the hydroxyl ion concentration. The rate of the reaction represented by Line C has been increased over Line A by changing the reaction atmosphere to oxygen. Line D represents a reaction incorporating both the higher hydroxyl ion concentration, Line B, and an atmosphere of oxygen, Line C. The reaction represented by Line D is more than 10 times faster than the reaction represented by either Lines B or C. The effects of hydroxyl ion concentration and oxygen on the rate of reaction of methyl- β -glucoside are, then, additive in nature.

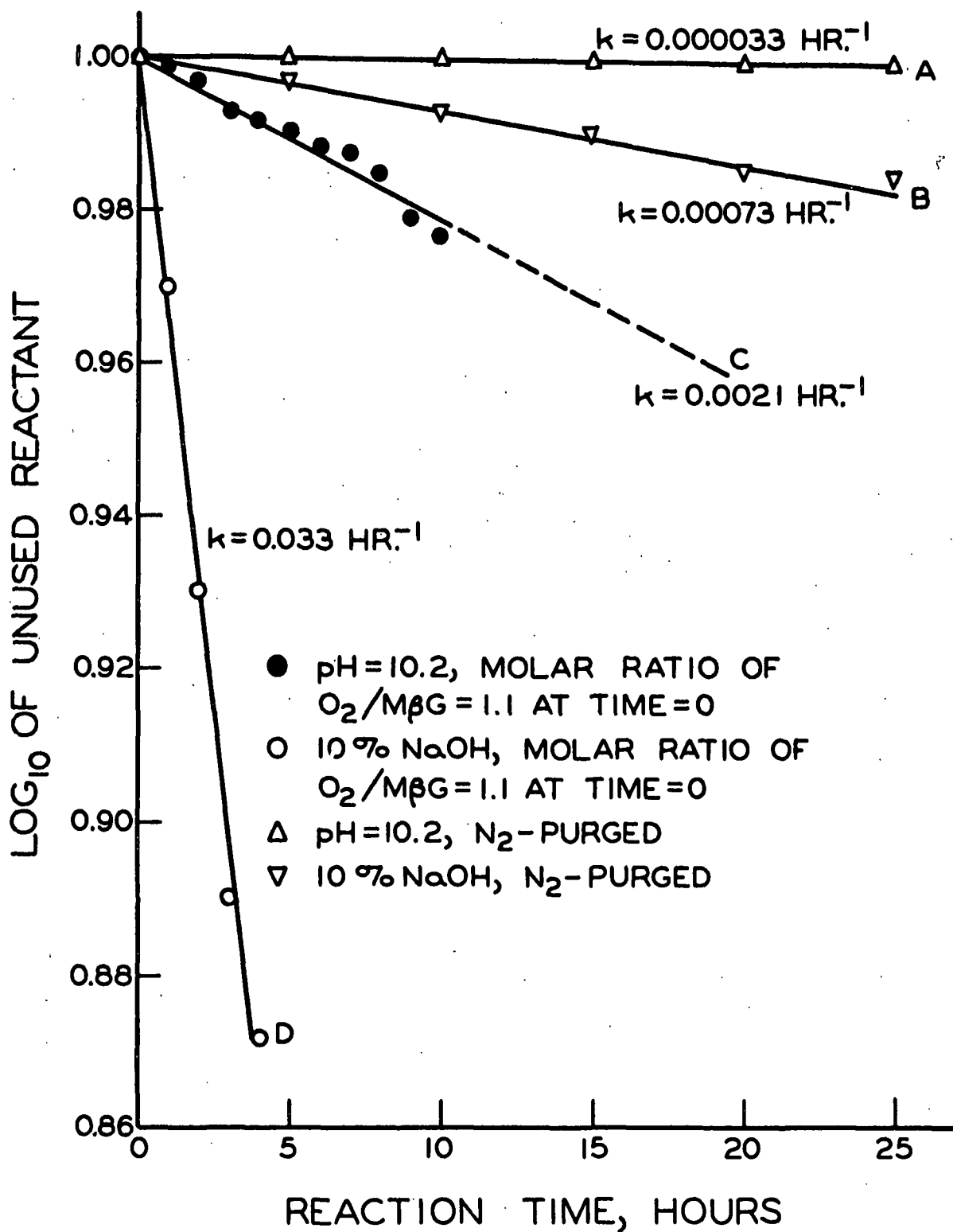


Figure 9. The Reaction of Methyl-β-Glucoside at 150°C., Varying Both Hydroxyl Ion and Molecular Oxygen Concentration

DISCUSSION

In the following discussion the assumption is made that the glycosidic bonds of methyl- β -glucoside and polysaccharides respond similarly to changes in reaction variables in an alkaline medium. This does not, however, involve the assumption that the rate constant for the cleavage of the glycosidic bonds are the same. It should also be noted that the cleavage and "peeling" mechanism of degradation are not totally independent because each time a polysaccharide chain cleavage takes place a reducing end is formed which is subject to further degradation by "peeling." Therefore, an increase in the rate of random chain cleavage will also increase the rate of the "peeling" reaction. This interaction does not affect the reaction of methyl- β -glucoside because the "peeling" reaction cannot take place. When drawing an analogy between the glycosidic bonds of methyl- β -glucoside and the polysaccharides, however, this interaction between chain cleavage and "peeling" should be realized.

The effect of temperature on the rate of random cleavage of glycosidic bonds in an alkaline medium is illustrated in Fig. 3, the Arrhenius plot. As was discussed in the introduction, the literature concerning the over-all degradation rates of polysaccharides in an alkaline medium (3) suggests a possible change in the over-all rate-determining reaction mechanism at about 150°C. This change at 150°C. either may or may not have reflected a change in the mechanism of the random cleavage of glycosidic bonds. The data shown in Fig. 3, though, form a straight line from 140 to 170°C. indicating that no change in the cleavage reaction mechanism occurs within this temperature range.

The random cleavage reaction, however, may still justifiably be used to explain the apparent change in mechanism of the over-all degradation reaction. As

illustrated in Fig. 2, the rate of random cleavage of the glycosidic bond is very small at 140°C. being about 0.3% reaction in 5 hours. This amount reacted, however, increases to 0.8% at 150°C., and to 5.5% at 170°C. This represents a 22-fold increase in the rate of cleavage between 140 and 170°C. In work with 4-0-methylglucuronoxylan, experiments conducted under similar conditions resulted in only a 3-fold increase in the rate of over-all polysaccharide degradation (4). This difference is accompanied by a corresponding difference in activation energy. The Arrhenius activation energy for the random cleavage reaction (shown in Fig. 3) was found to be 36,000 cal./g. mol. Since the activation energy for the over-all degradation of polysaccharides, which includes both the "peeling" and cleavage mechanisms, has been found to be near 17,000 cal./g. mol. (4,10) the activation energy required for the "peeling" reaction alone must be quite low. Because the activation energy for random cleavage is much greater than that for "peeling" it follows that at high temperatures, e.g., 170°C., random cleavage tends to be rate controlling while at low temperatures, e.g., 100°C., "peeling" is rate controlling (52).

Turning now to the effect of pH, the decrease in D.P. noted by both Thompson, et al., (21) and Meier (2) as the pH of the polysaccharide reaction system was increased above 11.0 is consistent with the corresponding large increase in the rate of random cleavage of glycosidic bonds established in this experimental work as shown in Fig. 7 and 8. The hypothesis put forth by these investigators that the decrease in polysaccharide viscosity was caused by an increase in the relative importance of the random cleavage reaction is, then, substantiated.

In the Introduction the conflicting views on the effect of molecular oxygen on the over-all alkaline degradation of polysaccharides presented in the literature were outlined. From the current experimental work it is seen that molecular

oxygen has a very large effect on the rate of cleavage of the glycosidic bond. The amount of molecular oxygen present in the reaction system was not defined by previous investigators. This knowledge is considered very important since the rate of reaction is greatly affected by the presence of oxygen, as measured by a pseudo first-order rate constant. This point is well illustrated in Fig. 4, 5, 6 and 8.

Two probable reaction pathways leading to cleavage of the glycosidic bond in an alkaline medium with molecular oxygen present are illustrated in Fig. 10, again using methyl- β -glucoside as the reactant. It should be noted that Pathway A is the same as that shown in Fig. 1. Pathway B represents that part of the reaction assisted by the presence of oxygen and is proposed on the basis of the current experimental work. The common first step to both pathways is the formation of the secondary carbinolate anion, II. This may be visualized as a required "activation" step, needed before either the inversion with elimination as in Pathway A or autoxidation as in Pathway B will take place.

Theoretically, methanol could ionize to the methoxyl ion and also react via Pathway B, although it was earlier shown to be experimentally stable. Hine and Hine (53) have shown that the relative acidity of ethylene glycol is about 11 times that of methanol and that the acidity of glycerol is about 44 times that of methanol. Since alkaline autoxidation is dependent upon anion formation and methanol forms a relatively small amount of anion, methanol should be reacted relatively slowly. This would account for the relative stability of methanol in reaction via Pathway B.

Theander (31) has followed much of Pathway B. Starting with IV and with VI he has characterized IX, actually isolating IX in several experiments. The synthesis of IV and VI was accomplished in an acidic medium where these keto glycosides

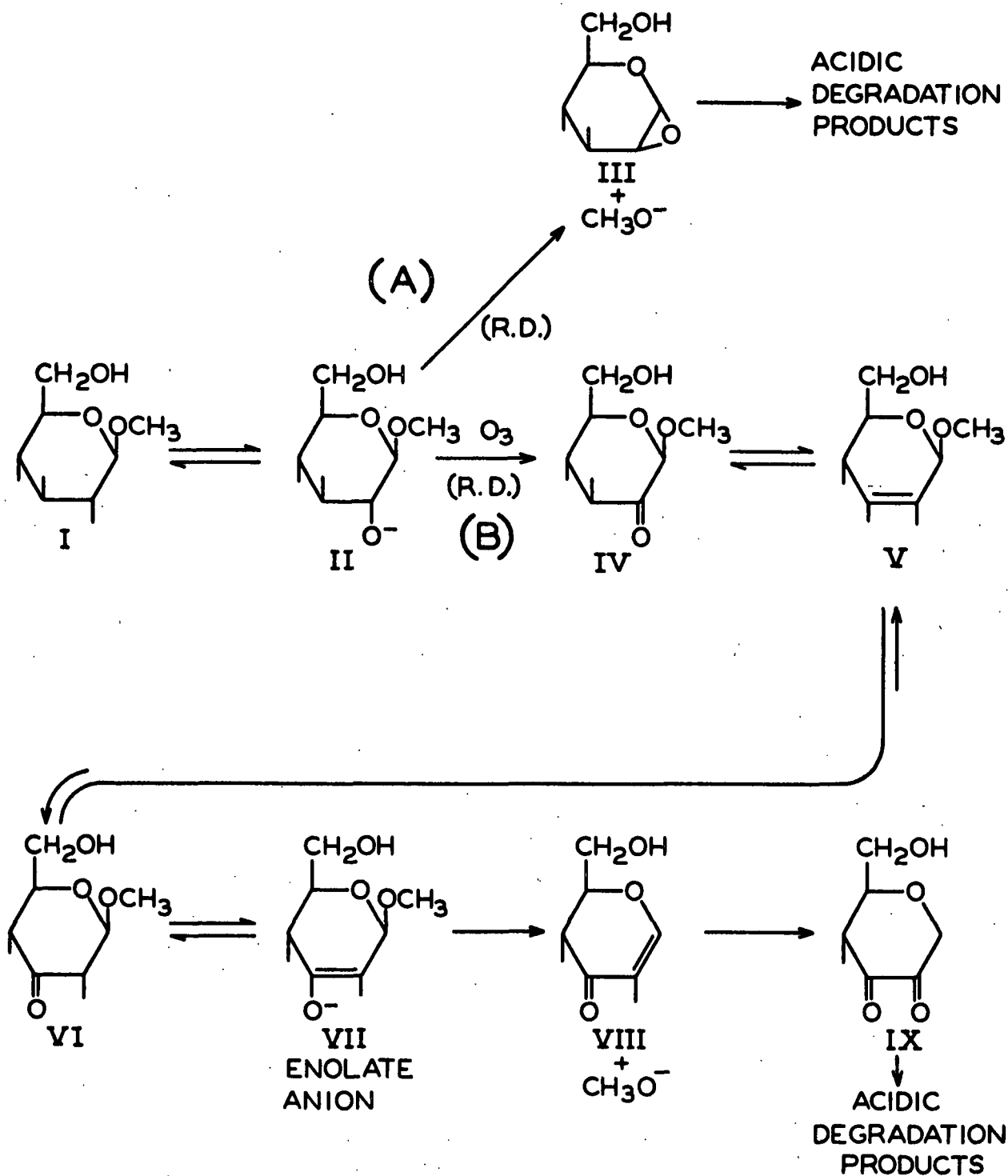


Figure 10. Two Probable Reaction Pathways Leading to Cleavage of Glycosidic Bond of Methyl-β-Glucoside in an Alkaline Medium with Molecular Oxygen Present

are relatively stable. He established that the reaction of IV to IX in alkali is very rapid by decomposing IV in lime water at 25°C. He obtained a rate of decomposition which is more than 100 times faster than that found for the decomposition of I in 10% sodium hydroxide at 170°C. in the current work. This means that the rate-determining step must precede the reaction of IV. By either pathway this leaves as the probable rate-determining step the reaction of II to form less stable products. The reaction rate by either of the postulated pathways should then be dependent upon the concentration of II, which in turn is dependent upon the concentrations of hydroxyl ion and I. In reaction via Pathway B, however, there should be an additional dependence of the rate-determining step upon the concentration of molecular oxygen.

The formation of methyl 2-keto- β -D-glucopyranoside, IV, entails reaction of molecular oxygen with the required precursor, II, which is highly susceptible to autoxidation in an alkaline medium (54). This oxidation may proceed via a free radical mechanism (55-57) or an ionic mechanism (58).

The most probable course of Pathway B after oxidation to IV involves migration of the keto group via the enediol form, V, to the C-3 position to give methyl 3-keto- β -D-glycopyranoside, VI. This keto migration is an example of the well-known Lobry de Bruyn-Alberda van Ekenstein transformation (27,59). In an alkaline medium VI will be in equilibrium with its enolate form, VII (1,31,54). The enolate anion is then in a position beta to the alkoxy group and cleavage of the glycosidic bond proceeds by the β -elimination of a methoxy ion (1,31,33), forming the 3-keto-1,2-enol, VIII, which is in turn thought to be rapidly fragmented at temperatures in the vicinity of 170°C. with hydroxyl ion concentration.

By Pathway A the reaction of II and the elimination of the methoxy ion are the same reaction. Therefore, the rate of production of methanol was a direct

measure of the rate-determining step. This was not the case in reaction by Pathway B, however, where several reaction steps are thought to take place between the rate-determining step and the actual elimination of the methoxyl ion.

Reaction Pathways A and B of Fig. 10 are meant to illustrate what are hypothesized as the main routes of reaction. Several alternate routes based on these two pathways are plausible. For example, a carbinolate anion could be formed at C-3, allowing direct oxidation at this site. Migration of the keto group would then not be required. With methyl- β -glucoside, but not cellulose, the carbinolate anion could be formed at C-4 allowing oxidation at this site with subsequent migration of the keto group to C-3. Although the C-6 hydroxyl is thought to be ionized to an extent approximating that of the C-2 hydroxyl (33) subsequent keto migration is inhibited because of the predominantly pyranose form of the glycon group. Also, in a concentrated alkaline medium, this primary hydroxyl is thought to be in large part complexed with the alkali (60) which would further inhibit reaction.

It should be noted that Pathway B after oxidation to IV is very similar to the "peeling" reaction. Bearing in mind that in the "peeling" reaction the elimination takes place from C-4 and in Pathway B the elimination takes place from C-1, all of the reaction steps of Pathway B after the oxidation (IV to VIII) are the same as those postulated in the "peeling" reaction, relative to the site of elimination. Since the "peeling" reaction is known to proceed at a relatively rapid rate in an alkaline solution, (9) this further substantiates the hypothesis that II to IV is the rate-determining step in Pathway B.

Table I shows the effect of temperature on the ability of oxygen to increase the reaction rate. In 10% sodium hydroxide the reaction rate is increased 1.9 times at 170°C. and 4.5 times at 150°C. by the inclusion of excess oxygen.

Therefore, not only is the effect of molecular oxygen on the reaction rate very large, but also reaction by Pathway B of Fig. 10 is less affected by changes in temperature than is reaction by Pathway A, since at the higher temperature more of the reaction is proceeding via Pathway A than at the lower temperature. That is, the activation energy for reaction by Pathway B is undoubtedly less than that for reaction by Pathway A.

Since the cleavage of the glycosidic bond is of primary interest in this study, all other reactions will be termed "secondary reactions." These secondary reactions in the alkaline degradation of methyl- β -glucoside are thought to consist primarily of fragmentation of the glycon after cleavage of the glycosidic bond, as indicated in Fig. 1 and 10. The presence of molecular oxygen could easily alter the mechanisms and rates of side reactions, just as it has done in the cleavage reaction. Since reaction by Pathway B of Fig. 10 requires one mole of oxygen per mole of methyl- β -glucoside, it appears as though products formed through secondary reactions consume molecular oxygen. From extrapolation of Fig. 4, 5, 6 and 8 the apparent number of moles of methyl- β -glucoside reacted with molecular oxygen by Pathway B of Fig. 10 was calculated. The moles of molecular oxygen in the system at zero reaction time, which was totally consumed, was known. From these two numbers the ratio of the moles of oxygen consumed per mole of methyl- β -glucoside reacted via Pathway B was calculated. At 170°C. ratios of 2.7 and 2.2 were obtained from Fig. 4 and 5, respectively, the average being 2.5. From Fig. 6 and 8, these reactions being run at 150°C., equal ratios of 3.7 were obtained. The effectiveness of a given quantity of molecular oxygen in promoting cleavage of the glycosidic bond, then, is seen to decrease as the reaction temperature is decreased. Most likely more of the molecular oxygen reacts via secondary reactions at lower temperatures. This is very plausible and could be caused

by a difference in average activation energy in reaction via primary and secondary reaction.

The rate-determining step of the postulated reaction pathways illustrated in Fig. 1 and 10 has been determined. In reaction by either Pathway A or B the concentration of II is dependent upon the equilibrium constant between I and II, K_1 , and the concentrations of I and hydroxyl ion, as shown in Equation (1).

$$[\text{II}] = K_1 [\text{I}][\text{OH}^-] \quad (1)$$

The rate-determining step by either pathway is postulated as the reaction of II, as represented by Equation (2) for an oxygen-free system and Equation (3) for reaction with molecular oxygen.

$$-d [\text{II}]/dt = k_A [\text{II}] \quad (2)$$

$$-d [\text{II}]/dt = k_A [\text{II}] + k_B [\text{II}][\text{O}_2] \quad (3)$$

In all cases the rate is dependent upon the concentration of II, the concentration of II being dependent upon the hydroxyl ion concentration. In this manner the hydroxyl ion concentration does have an effect upon the reaction rate. This is easily demonstrated by combining Equations (1) and (2) for an oxygen-free system as done in Equation (4), and combining Equations (1) and (3) for reaction with molecular oxygen as is done in Equation (5).

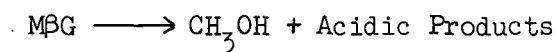
$$-d [\text{II}]/dt = k_A K_1 [\text{I}][\text{OH}^-] \quad (4)$$

$$-d [\text{II}]/dt = k_A K_1 [\text{I}][\text{OH}^-] + k_B K_1 [\text{I}][\text{OH}^-][\text{O}_2] \quad (5)$$

If the reaction of II is the rate-determining step in reaction by either pathway, and if the concentration of II is dependent upon the hydroxyl ion concentration, then the over-all rate of reaction by either pathway should vary with the hydroxyl

ion concentration. This was found to be so as is illustrated in Fig. 7 and 8 for reactions excluding and including molecular oxygen, respectively. These experimental results are qualitatively consistent with the initial formation of II from I followed by reaction of this anion as the rate-determining step in reaction by either pathway. Also, if reaction of II is the rate-determining step and if II reacts with molecular oxygen by Pathway B, then the concentration of molecular oxygen will affect the rate of reaction. This was also found to be the case. Reaction of II by Pathways A and B is the only reaction step which can be rate-determining and still be consistent with all of the experimental facts.

The reaction of one mole of methyl- β -glucoside in alkali was found to yield one mole of methanol, i.e.,



This establishes that the glycosidic bond of methyl- β -glucoside was cleaved and indicates that cleavage of a polysaccharide in alkali probably occurs at the glycosidic bond. No change in mechanism of the random cleavage reaction was noted in the range of 140 to 170°C. based on the Arrhenius plot. A change in the rate-determining mechanism of over-all polysaccharide degradation is strongly indicated, however, because the Arrhenius activation energy for the cleavage reaction is much greater than that for the "peeling" reaction. The "peeling" reaction is rate determining up to 140 to 150°C. and the cleavage reaction appears to become rate determining as the temperature is increased further. A large effect of hydroxyl ion concentration on the rate of cleavage of methyl- β -glucoside in alkali was found. A large effect of molecular oxygen on this rate of cleavage was also established. A reaction pathway, Pathway B, for the reaction of methyl- β -glucoside in an alkaline medium containing oxygen is proposed on the basis of the current work. The rate-determining step of this pathway is

defined. The Arrhenius activation energy for reaction of oxygen via secondary reactions appears to be appreciably lower than for reaction of oxygen by Pathway B. The Arrhenius activation energy for cleavage of the glycosidic bond in the presence of oxygen was also found to be much less than that for cleavage in a nitrogen-purged system. The current work, then, not only increases considerably the understanding of the relative importance of the cleavage and "peeling" mechanisms in over-all polysaccharide degradation in alkali, but also establishes large effects of hydroxyl ion concentration and oxygen on the rate of the cleavage reaction.

SUGGESTIONS FOR FUTURE RESEARCH

Using a cellulose model compound such as methyl- β -glucoside, a study of the rate of cleavage of the glycosidic bond throughout the entire pH spectrum could be made, preferably using a high temperature pH monitoring system such as described by Ingruber (61,62). A very interesting phase of this work could be the inclusion of oxygen in some of the reactions. At pH 7.0 the effect of molecular oxygen on the rate of cleavage of the glycosidic bond should be greatly reduced from the rate at pH 10.2 because of the decreased carbinolate anion concentration.

By obtaining the reaction rates at several temperatures, 140 through 170°C., at each of several pHs, e.g., 8.0, 9.5, 11.0, 12.5, a comparison of the entropy and enthalpy of activation at each level could be made. This would indicate whether or not the mechanism of glycosidic bond cleavage remained constant with changing pH. The work could be done both with and without atmospheric oxygen. Methyl- β -glucoside would again serve as the reactant.

A study of the total degradation rates of cellulose could be made. A hydrocellulose can be made soluble in an alkaline medium by lowering the temperature to about -5°C. (15). Other work (63) indicates that the hydrocellulose will stay in solution when it is reacted at higher temperatures. Through studying similar homogeneous and heterogeneous systems, possibly the decrease in pulp viscosity obtained by Thompson, *et al.*, (21) as the pH of the pulping medium was changed from 11.0 to 7.0 could be explained. Linear polymer degradation theories could be used to calculate chain-cleavage rates as discussed by Spiegelberg (64).

By extending the concept of model compounds of cellulose from methyl- β -glucoside, methyl- β -cellobioside and possibly methyl- β -cellotrioside, the effect of chain length and leaving groups on the rate of cleavage of the glycosidic bond could be obtained (65-67).

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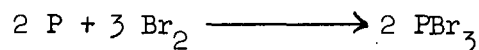
APPENDIX I

PROCEDURE FOR THE PREPARATION OF
METHYL- β -GLUCOSIDE FROM GLUCOSE

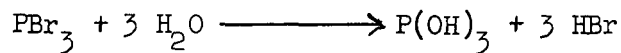
PREPARATION OF ACETOBROMOGLUCOSE FROM GLUCOSE

The following outline was used in the synthesis of 2,3,4,6-tetra-O-acetyl- β -D-glucosyl bromide from glucose (41):

1. Add 2.4 ml. of perchloric acid to 400 ml. of acetic anhydride in a 2000-ml. flask reaction vessel.
2. Add 100 g. of glucose slowly over a 1/2-hour period, keeping the temperature between 30 and 40°C.
3. Add 30 g. of amorphous red phosphorus.
4. Cool the reaction mixture in an ice bath to 10°C.
5. With the reaction vessel still in the ice bath, slowly add 180 g. (61.3 ml.) of bromine, keeping the temperature below 20°C. The reaction taking place can be represented as:



6. Leaving the reaction vessel in the ice bath, add 36 ml. of water (slightly less than the stoichiometric amount) slowly over a 1/2-hour period, avoiding local temperature rise. The reaction is:



HBr is the brominating species.

7. Allow the reaction mixture to stand at room temperature for 2 hours in a closed vessel. This step completes the synthesis of acetobromoglucose.

8. Add 300 ml. of chloroform to the reaction mixture, which dissolves the acetobromoglucose.
9. Pour this chloroform solution into 800 ml. of ice water. The water phase attracts the excess acetic acid.
10. Filter both phases through Celite.
11. Transfer the filtrate to a 2000-ml. separatory funnel and extract according to the scheme outlined in Fig. 11.
12. Dry the chloroform solution of acetobromoglucose by adding 2 g. sodium bicarbonate, number 4 mesh calcium chloride to equal 1/2 volume of the solution, and 5 g. activated carbon. Let stand 1/2 hour with occasional stirring.
13. Filter through Celite.
14. Evaporate the filtrate to dryness in vacuo from a 60°C. bath.
15. Dissolve the sirup in 800 ml. of prepurified chloroform (42). Ethanol must be removed from the chloroform before it is used in the Koenigs-Knorr reaction in the synthesis of methyl- β -glucoside tetraacetate as outlined in the next section.

PREPARATION OF METHYL- β -GLUCOSIDE TETRAACETATE
FROM ACETOBROMOGLUCOSE

The following outline was used in the preparation of methyl-2,3,4,6-tetra-O-acetyl- β -D-glucoside from 2,3,4,6-tetra-O-acetyl- β -D-glucosyl bromide (42).

1. To a light-tight 3000-ml. reaction vessel, equipped with a motor-driven stirrer, add 555 ml. of purified chloroform and 139 g. of silver oxide which has been predried over phosphorous pentoxide.

2. Add 555 g. of 10/20 nonindicating Drierite which has been heated for 2 hours at 240°C. and cooled over Drierite.
3. Add 45 ml. of methanol.
4. Add 27.8 g. of iodine.
5. Stir the mixture 1 hour to insure the absence of moisture at the beginning of the reaction.
6. Add the previously synthesized acetobromoglucose solution slowly over a 1-hour period. This must be done slowly so that the water formed by the methylation reaction will be present only in small quantity and therefore more effectively taken up by the internal desiccant.



7. Stir the reaction mixture for 24 hours. About 1/2 hour before the reaction time is completed add 5 g. activated carbon.
8. Filter the entire product mixture through a Celite pad.
9. Concentrate the filtrate to dryness in vacuo from a 60°C. bath.
10. Dissolve the sirup in a minimal amount of hot absolute ethanol, about 300 ml.
11. Cool the solution at room temperature to obtain the crystalline methyl-β-glucoside tetraacetate product. Concentrate the mother liquor to obtain a second crop of crystals....
12. Crops 1 and 2 are combined and used as the reactant in the synthesis of methyl-β-glucoside as outlined in the next section.

DEACETYLATION OF METHYL- β -GLUCOSIDE TETRAACETATE

The following outline was used in the deacetylation of methyl-2,3,4,6-tetra-O-acetyl- β -D-glucose to yield methyl- β -D-glucopyranoside (43).

1. Dissolve the previously synthesized methyl- β -glucoside tetraacetate in 750 ml. of hot methanol.
2. Add 30 ml. of 0.1 molar sodium methylate in methanol.
3. Boil for 15 minutes. Add 5 g. of activated carbon after the solution has boiled 10 minutes (so that it is present for the last 5 minutes of the reaction time).
4. Filter through Celite.
5. Concentrate to about 200 ml. in vacuo from a 60° bath.
6. Refrigerate for 36 hours to obtain the first crop of methyl- β -glucoside. Concentrate the mother liquor and refrigerate for 36 hours to obtain the second crop.
7. Combine crops one and two to obtain the product, which will be about 98% methyl- β -glucoside and 2% glucose with no other impurities present in amounts analyzable by standard paper chromatography techniques.

APPENDIX II

SODIUM HYDROXIDE CONCENTRATIONS USED
IN REACTION SOLUTIONS

The thermal expansivity data for water (68) were used to calculate the appropriate sodium hydroxide concentration at 20°C. which would provide a concentration of 10% at the reaction temperature desired. The sodium hydroxide concentrations, acid-base titrated at 20°C., used are listed in Table II.

TABLE II

SODIUM HYDROXIDE CONCENTRATIONS USED WITH
THE VARIOUS REACTION TEMPERATURES^a

Reaction Temp., °C.	Concentration of Sodium Hydroxide at 20°C., %
170	11.12
165	11.06
160	11.00
155	10.94
150	10.89
145	10.84
140	10.79

^aAll concentrations are calculated to be 10% in sodium hydroxide concentration at the reaction temperature indicated.

APPENDIX III

TITRATION OF PRODUCT SOLUTIONS FROM THE
REACTION OF METHYL- β -GLUCOSIDE IN 10% SODIUM HYDROXIDE

The data shown in Table III were obtained by subtracting the concentration of sodium hydroxide product solution from the concentration of sodium hydroxide in the unreacted solution. All concentrations were obtained by acid-base titration. This procedure is meant to give only an approximate value for the amount of sodium hydroxide neutralized during the reaction.

TABLE III

AMOUNT OF SODIUM HYDROXIDE NEUTRALIZED DURING THE REACTION OF
METHYL- β -GLUCOSIDE IN 10% SODIUM HYDROXIDE UNDER NITROGEN-PURGED CONDITIONS

Reaction Time, hours	Per Cent of Sodium Hydroxide Neutralized Which Was Present at Zero Reaction Time			
	145°C. Reaction Temp.	155°C. Reaction Temp.	165°C. Reaction Temp.	170°C. Reaction Temp.
0	0	0	0	0
5	0.4	0.5	1.2	2.0
10	0	0	0.9	—
15	-0.2	0.9	0.9	—
20	0	0.7	1.4	—
25	0.2	0.7	1.4	3.8
30	0	0.2	2.5	—
35	-0.2	0.9	2.4	—
40	0.4	0.9	3.1	—
45	0.2	0.7	3.6	—
50	0.2	0.9	3.6	5.5

APPENDIX IV

PROCEDURE FOR THE PREPARATION
OF A NITROGEN-PURGED REACTION ATMOSPHERE

The following procedure was used to greatly reduce the availability of oxygen during the reaction of methyl- β -glucoside in aqueous sodium hydroxide. By this procedure the oxygen in the atmosphere above the reaction solution is almost entirely removed and the oxygen present in the reaction solution itself is greatly reduced.

1. Boil a known volume of the caustic solution and a separate flask of water for 15 minutes at atmospheric pressure.
2. Purge both vessels with nitrogen, close loosely and cool to 20°C. in a cold-water bath.
3. Repurge both vessels as they cool.
4. Purge all vessels into which the solutions are poured.
5. Dilute the cooled solution to its original volume with the boiled water.
6. Purge the reaction vessel by inserting a glass tube which is fed by nitrogen all the way into the vessel and slowly withdraw it.
7. Add the proper volume of reactant solution to the vessel.
8. Repurge the remaining gas volume of the reaction vessel with a glass tube of the proper length and seal the tube immediately.

APPENDIX V

DEIONIZATION OF PRODUCT SOLUTIONS OF METHYL- β -GLUCOSIDE
REACTED IN 10% SODIUM HYDROXIDE UNDER NITROGEN-PURGED CONDITIONS

Tables IV and V show the results of deionization of product solutions. In all cases one-inch inside diameter columns were used. The bed depth was approximately 8 inches using 100 ml. of resin. The liquid level in the column was maintained approximately even with the top resin surface. The Amberlite IR-120 acidic resin was thoroughly prewashed with distilled water to minimize the leaching of color from the resin upon passing the caustic solution through. No problem with color leaching from the Amberlite MB-3 mixed bed resin was encountered so that these columns were not prewashed to any great extent. The product solution was removed from the resin by washing with approximately six times the liquid capacity of the column using distilled water (250 ml. distilled water per 40 ml. bed capacity for 100 ml. of resin). The effluent from the column was then condensed back to its original volume (25 ml.) using a Renco low-pressure evaporator with a 60°C. water bath. The optical rotation, using a sodium light source, α_2 , was then taken and compared with the rotation of the unprocessed sample, α_1 .

TABLE IV

DEIONIZATION OF PRODUCT SOLUTIONS OF METHYL- β -GLUCOSIDE
 REACTED IN 10% SODIUM HYDROXIDE AT 165°C.^a

Reaction Time, hours	Specific Rotation of Unprocessed Product Solution, α_1	Specific Rotation of Deionized Product Solution, α_2	$\frac{\alpha_2}{\alpha_1}$ (100)
0	1.32	1.25	94.8
0	1.32	1.27	96.2
0	1.32	1.30	98.5
0	1.32	1.29	97.7
5	1.30	1.25	96.2
10	1.23	1.17	95.2
15	1.21	1.17	96.8
20	1.18	1.15	97.5
25	1.15	1.15	100.0
30	1.07	1.02	95.4
35	1.06	1.06	100.0
40	1.05	1.03	98.2
45	1.02	0.98	96.0
50	0.98	0.98	100.0

^aA 25-ml. aliquot of each product solution at this temperature was run through a column containing 100 ml. of Amberlite MB-3 ion-exchange resin.

TABLE V

DEIONIZATION OF PRODUCT SOLUTIONS OF METHYL- β -GLUCOSIDE
 REACTED IN 10% SODIUM HYDROXIDE AT 155°C.^a

Reaction Time, hours	Specific Rotation of Unprocessed Product Solution, α_1	Specific Rotation of Deionized Product Solution, α_2	$\frac{\alpha_2}{\alpha_1}$ (100)
0	1.27	1.27	100.0
0	1.26	1.23	97.7
5	1.24	1.22	98.4
10	1.23	1.18	95.9
15	1.22	1.19	97.6
20	1.20	1.16	96.7
25	1.18	1.17	99.2
30	1.17	1.17	100.0
35	1.16	1.13	97.5
40	1.14	1.10	96.2
45	1.12	1.12	100.0
50	1.11	1.07	96.5

^aA 25-ml. aliquot of each product solution at this temperature was run in series through a column of 75 ml. of Amberlite IR-120 ion-exchange resin followed by a column of 100 ml. of Amberlite MB-3.

APPENDIX VI
METHANOL ANALYSIS

The following procedure was adapted from the method of Boos (49) and used for the analysis of product methanol from the reaction of methyl- β -glucoside in aqueous sodium hydroxide. Confidence limits for the method are shown in Fig. 12 and Tables VI and VII. The distillation apparatus is shown in Fig. 13. This apparatus was operated at approximately 730 mm. of mercury vacuum. The stability of the methanol formed was ascertained by attempting to react it under the same conditions used to react the methyl- β -glucoside as shown in Fig. 14, data in Table VIII.

1. Place a 10-ml. sample of reaction product solution in the distillation apparatus shown in Fig. 13 and distill seven minutes at a bath temperature of 65°C.
2. Transfer the distillate (about 9 ml.) to a 10-ml. volumetric flask and dilute to the mark.
3. Transfer a known portion of the distillate (usually 1.0 ml.) to another 10-ml. volumetric flask, the test flask.
4. To the test flask, add 4 drops of 5% phosphoric acid.
5. Add 5 drops of 5% potassium permanganate.
6. Leave the test flask unstoppered for 10 minutes, swirling occasionally.
7. Add a saturated solution of sodium bisulfate dropwise until the solution is colorless.
8. Add 5 ml. of concentrated sulfuric acid.

9. Cool in an ice bath.
10. Add 4 drops of chromotropic acid (1,8-dihydroxynaphthalene-3,6-disulfonic acid).
11. Place the stoppered test flask in a boiling water bath for 15 minutes.
12. Dilute to the mark and read the per cent transmission at 570 m μ .
13. Obtain the equivalent amount of methanol from the precalibrated curve, Fig. 12.

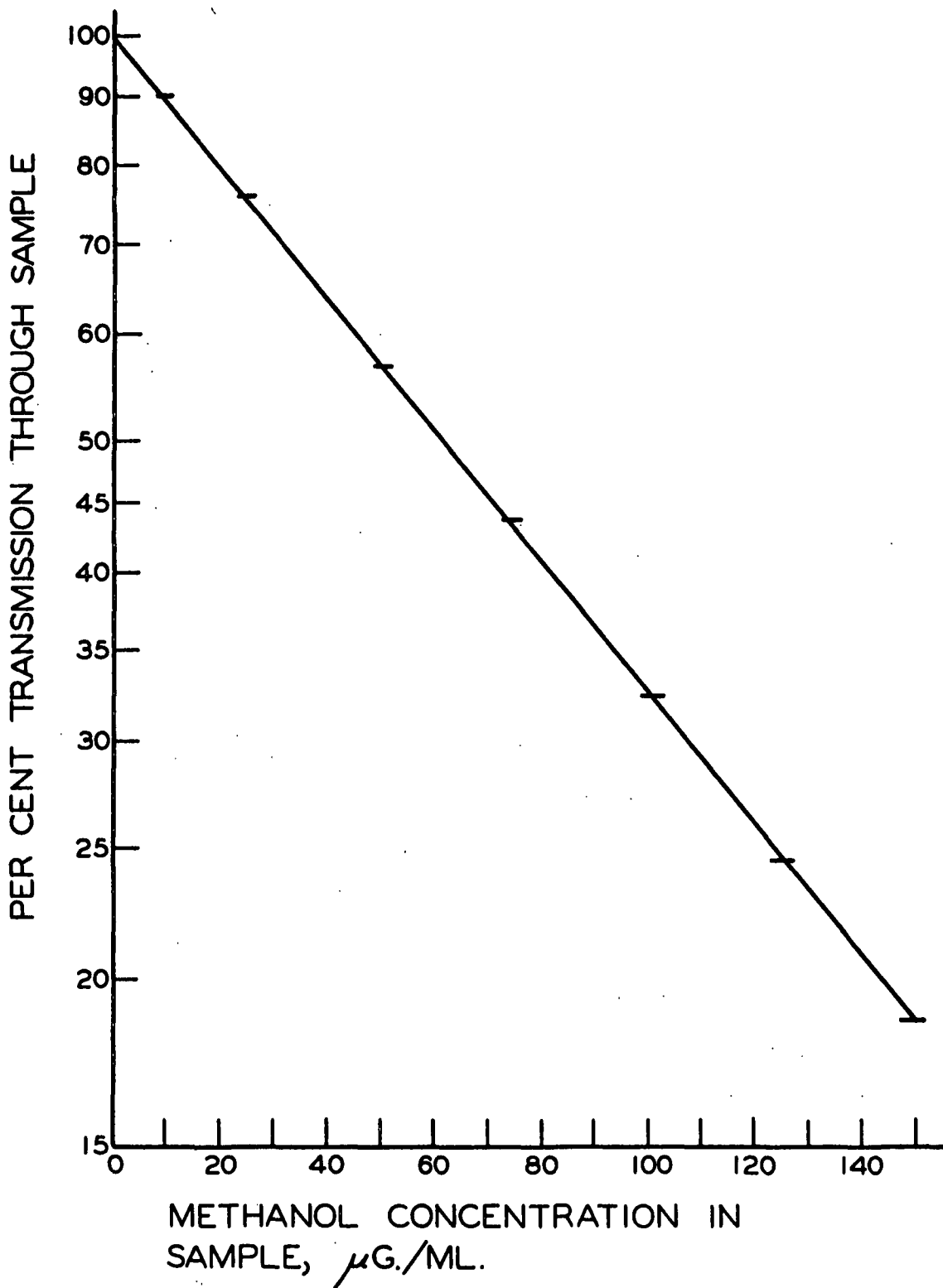


Figure 12. Calibration of Methanol Analysis Technique Showing the 95% Confidence Limits

TABLE VI

CONFIDENCE LIMITS FOR THE METHANOL ANALYSIS CALIBRATION CURVE

Known Amount of Methanol, μg./ml.	95% Confidence Limits Using 4 Samples per Concentration	Average Per Cent Transmission on the 4 Samples
10	10.3 ± 0.9	89.90
25	24.8 ± 0.7	76.24
50	50.5 ± 1.6	56.91
75	74.3 ± 1.8	43.60
100	101.0 ± 1.3	32.29
125	125.3 ± 2.3	24.46
150	149.6 ± 2.7	18.61

TABLE VII

CONFIDENCE LIMITS FOR THE DISTILLATION OF
AQUEOUS METHANOL FROM A SODIUM HYDROXIDE SOLUTION^a

Known Amount of Methanol in Distilled Solution, μg./ml.	Amount of Methanol Calibrated to be in Distillate, μg./ml.	95% Confidence Limits Using 6 Samples per Concentration Level	Maximum Per Cent Error
25	18.8	18.7 ± 1.4	8.2
100	75.0	74.3 ± 2.7	4.5
1000	750.0	742 ± 26	4.5

^aThese confidence limits necessarily include error encountered in methanol analysis as characterized in Table VI.

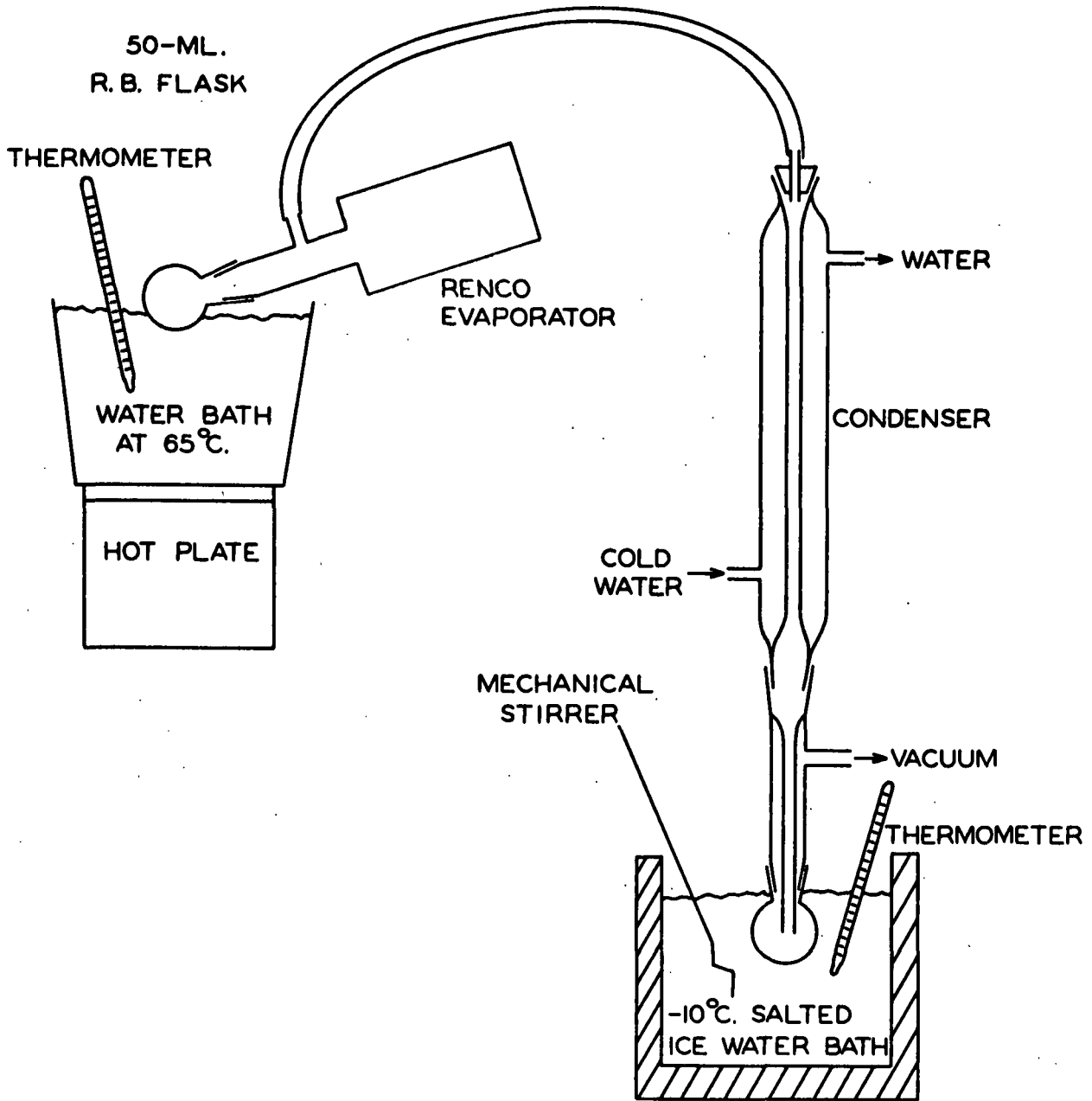


Figure 13. Distillation Apparatus

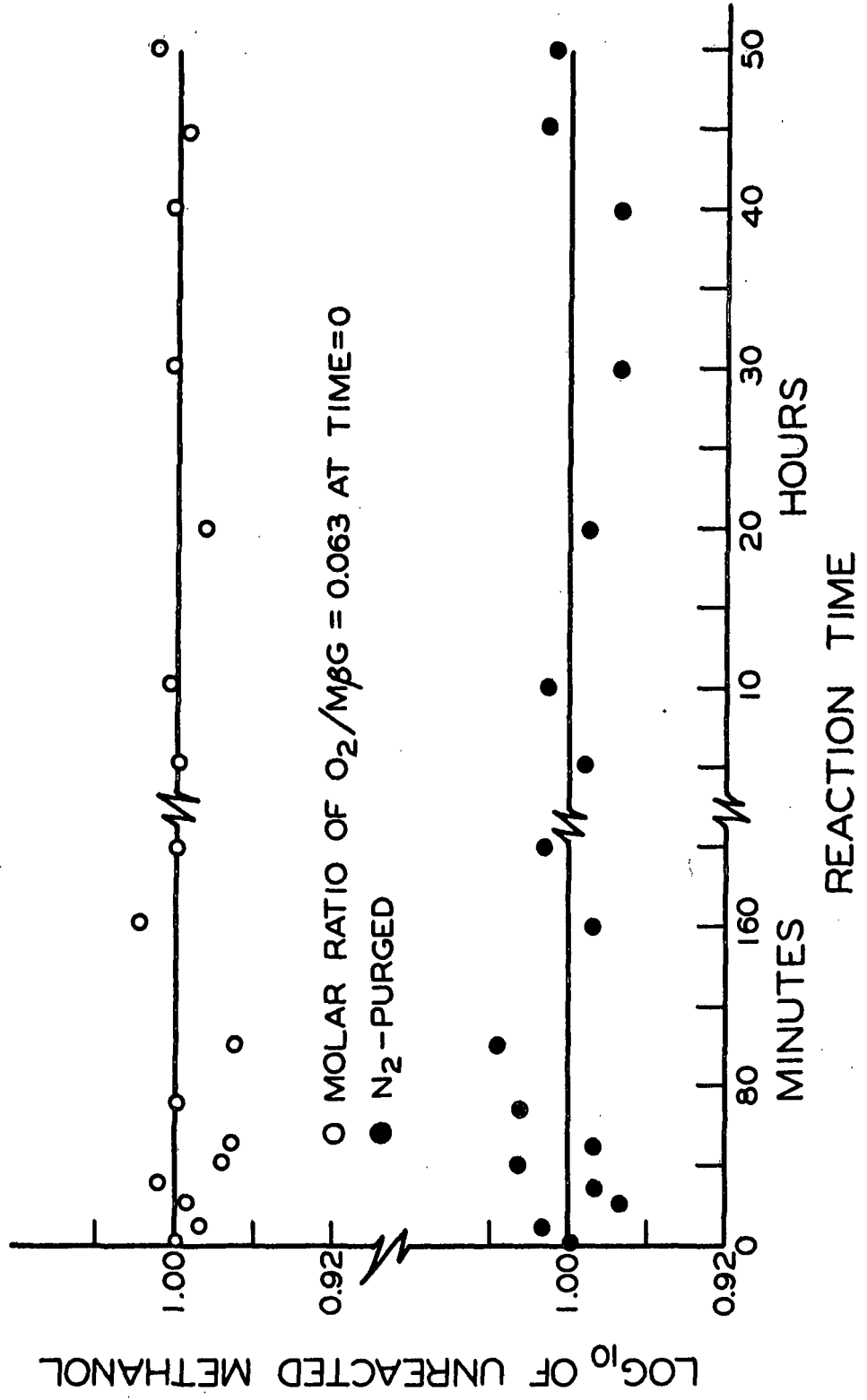


Figure 14. The Reaction of Methanol in 10% Sodium Hydroxide at 170°C.

TABLE VIII

THE REACTION OF METHANOL IN 10% SODIUM HYDROXIDE AT 170°C.

Reaction Time	Per Cent of Charged Methanol Present; N ₂ -Purged Conditions, µg./ml.	Per Cent of Charged Methanol Present, Molecular Oxygen Present ^a , µg./ml.
0	100.0	100.0
10 min.	97.2	103.0
20 min.	98.5	94.2
30 min.	101.4	97.1
40 min.	94.4	106.0
50 min.	93.0	97.1
70 min.	100.0	106.0
100 min.	93.0	109.0
160 min.	104.3	97.1
200 min.	100.0	103.0
5 hr.	98.8	98.3
10 hr.	101.3	102.2
20 hr.	97.0	98.2
30 hr.	100.4	94.7
40 hr.	100.4	94.7
45 hr.	99.2	103.0
50 hr.	102.2	102.2

^aThe ratio of molecular oxygen to methyl-β-glucoside was 0.063 at zero reaction time.

APPENDIX VII

STATISTICAL TREATMENT OF THE ARRHENIUS DIAGRAM

The linear correlation between the two variables of the Arrhenius Diagram, Fig. 3, was tested by calculating the correlation coefficient, r , between them.

$$r = \sqrt{\frac{\text{explained variation}}{\text{total variation}}}$$

If there is zero explained variation, i.e., the total variation is all unexplained, the ratio is zero. If there is zero unexplained variation, i.e., the total variation is all explained, the ratio is one. The data from Fig. 3 were used in a standard computer program, linear multiple regression, to calculate the correlation coefficient. A coefficient of 0.9941 was computed. In order to have 95% confidence that the data form a straight line, a correlation coefficient of 0.7545 or greater is required (69). Therefore, it can be said with a high degree of confidence that these data do form a straight line.

The F test may be used to determine whether or not the variation in observed data can be explained through experimental error. The Null Hypothesis is that the slope and intercept are meaningless numbers. An F ratio, a ratio indicating goodness of fit, was then calculated. If this calculated F value is greater than the tables F value, the slope and intercept have meaning (70). For the Arrhenius Diagram, Fig. 3, the calculated F value is 838. Using 95% confidence limits, the required value of F is 230 or greater. Therefore, it may be said with 95% confidence that the data form a straight line with slope = -7.874 and intercept = 15.41.

APPENDIX VIII

DATA AND SUPPLEMENTARY FIGURES

Figures 15 through 23 are presented as elaborations of figures presented in the main body. All of these figures are characterized both by polarimetric analysis of unused reactant and by-product methanol analysis. These reactions were also, in general, run for longer periods of time. Therefore, although these data cannot be represented in as compact a form as initial rate data, they are possibly more fully characterized and form the basis for the good analytical precision obtained in this experimental work. Figures 15, 18, 19, 20, 22, and 23, along with the appropriate part of Fig. 7 in the main body, illustrate the data which form the lines shown in Fig. 2. Figures 15 and 16 are extensions of Lines A and B, respectively, of Fig. 4. Figure 17 is an extension of Line D, Fig. 5. And Fig. 21 forms an extension of Line B, Figure 6.

All data shown in Tables IX through XXI are represented in figures shown either in the main body or in Fig. 15 through 23 of this appendix.

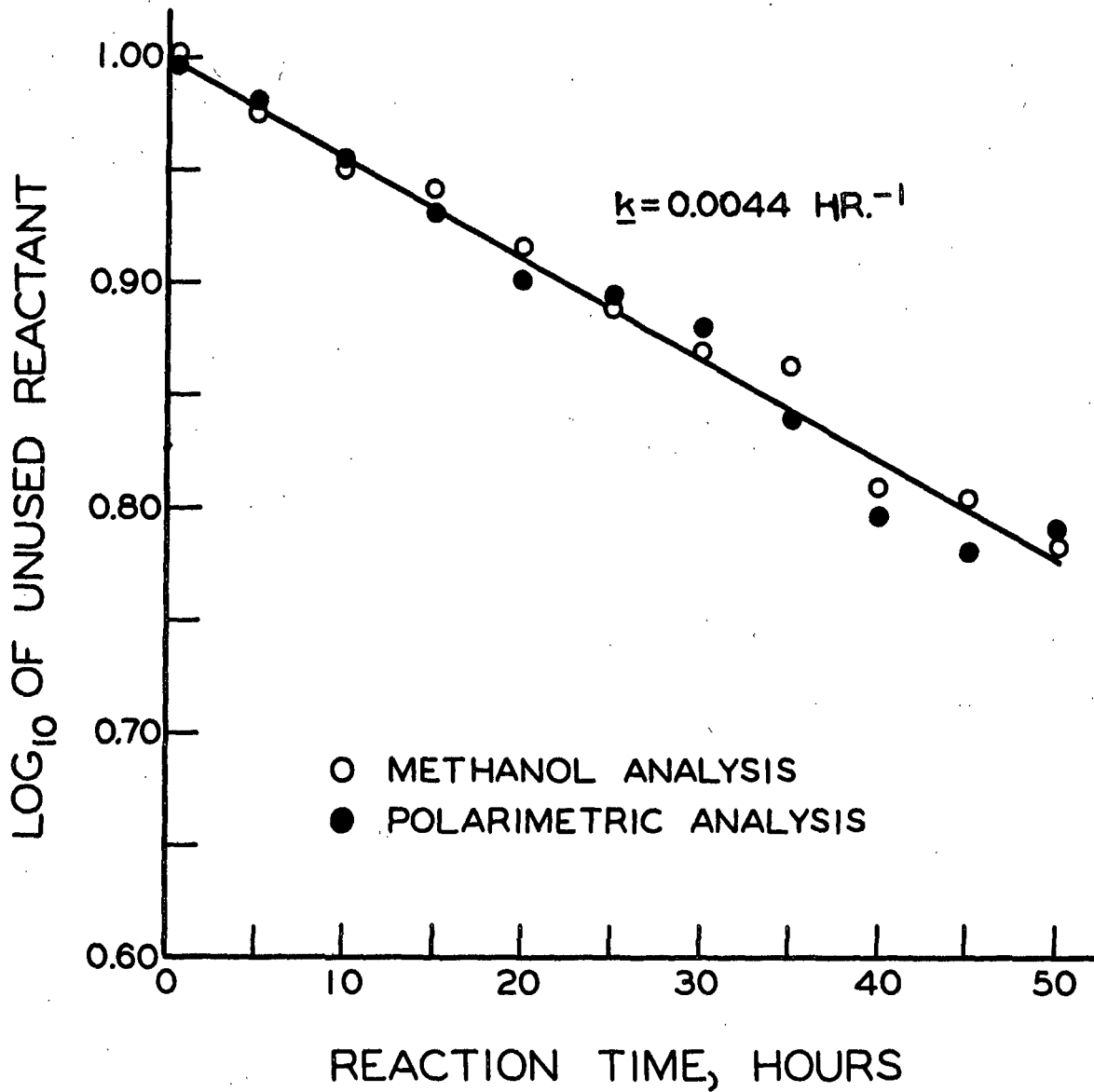


Figure 15. The Reaction of Methyl-β-Glucoside in 10% Sodium Hydroxide at 170°C. Under Nitrogen-Purged Conditions

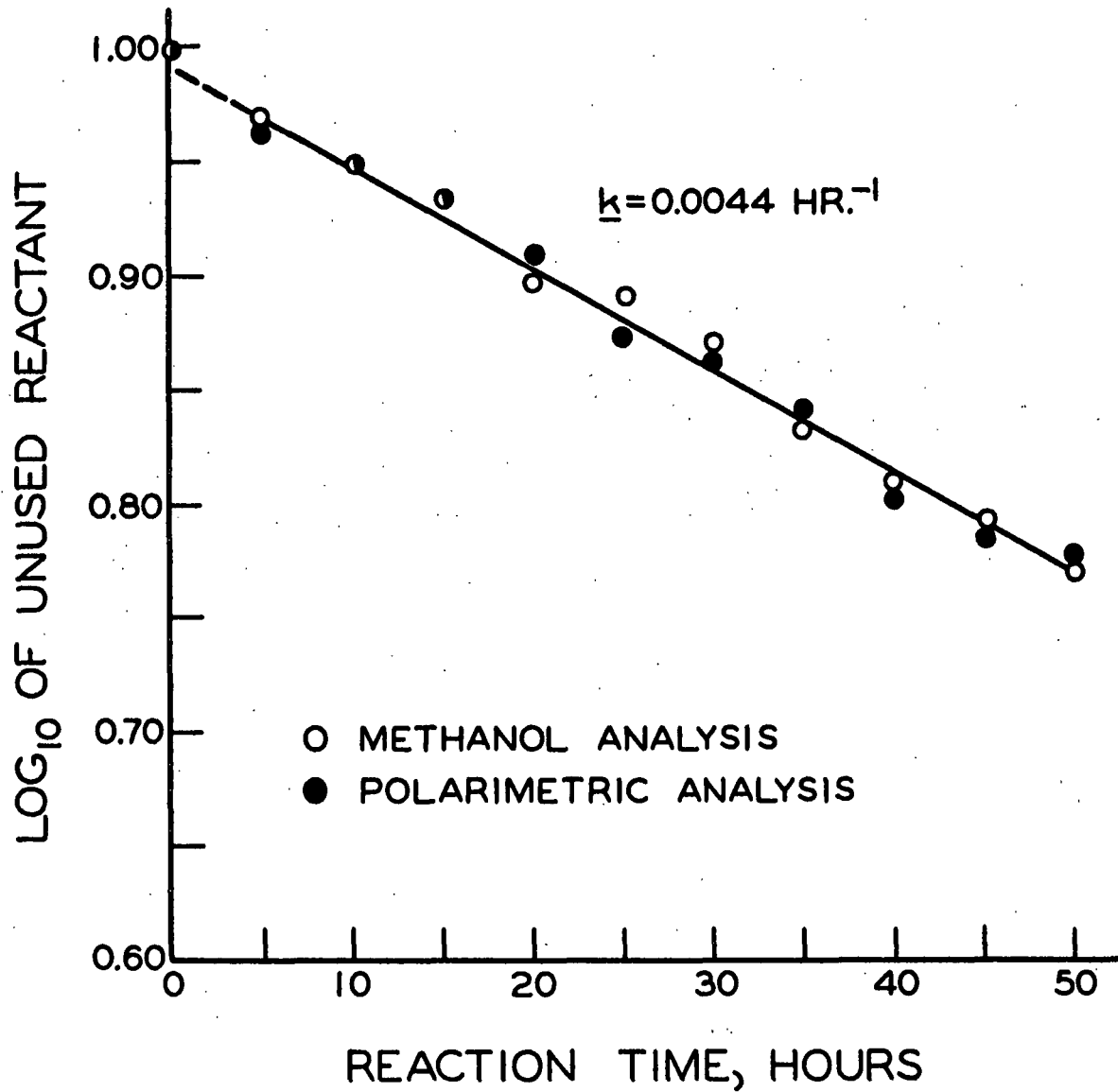


Figure 16. The Reaction of Methyl- β -Glucoside in 10% Sodium Hydroxide at 170°C. with a Molar Ratio of Molecular Oxygen to Methyl- β -Glucoside of 0.063 at Zero Reaction Time

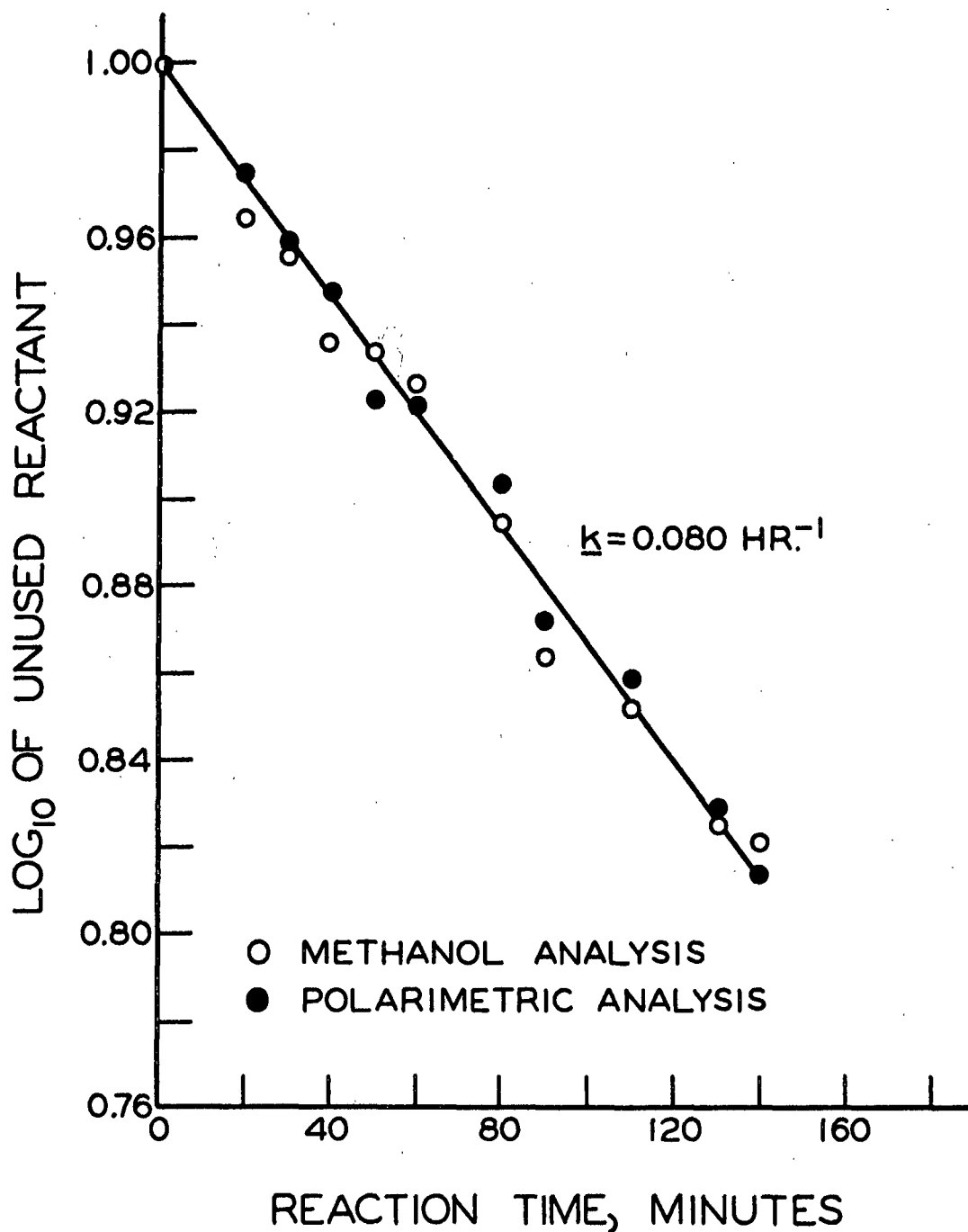


Figure 17. The Initial Reaction of Methyl- β -Glucoside in 10% Sodium Hydroxide at 170°C. with a Molar Ratio of Molecular Oxygen to Methyl- β -Glucoside of 1.1 at Zero Reaction Time

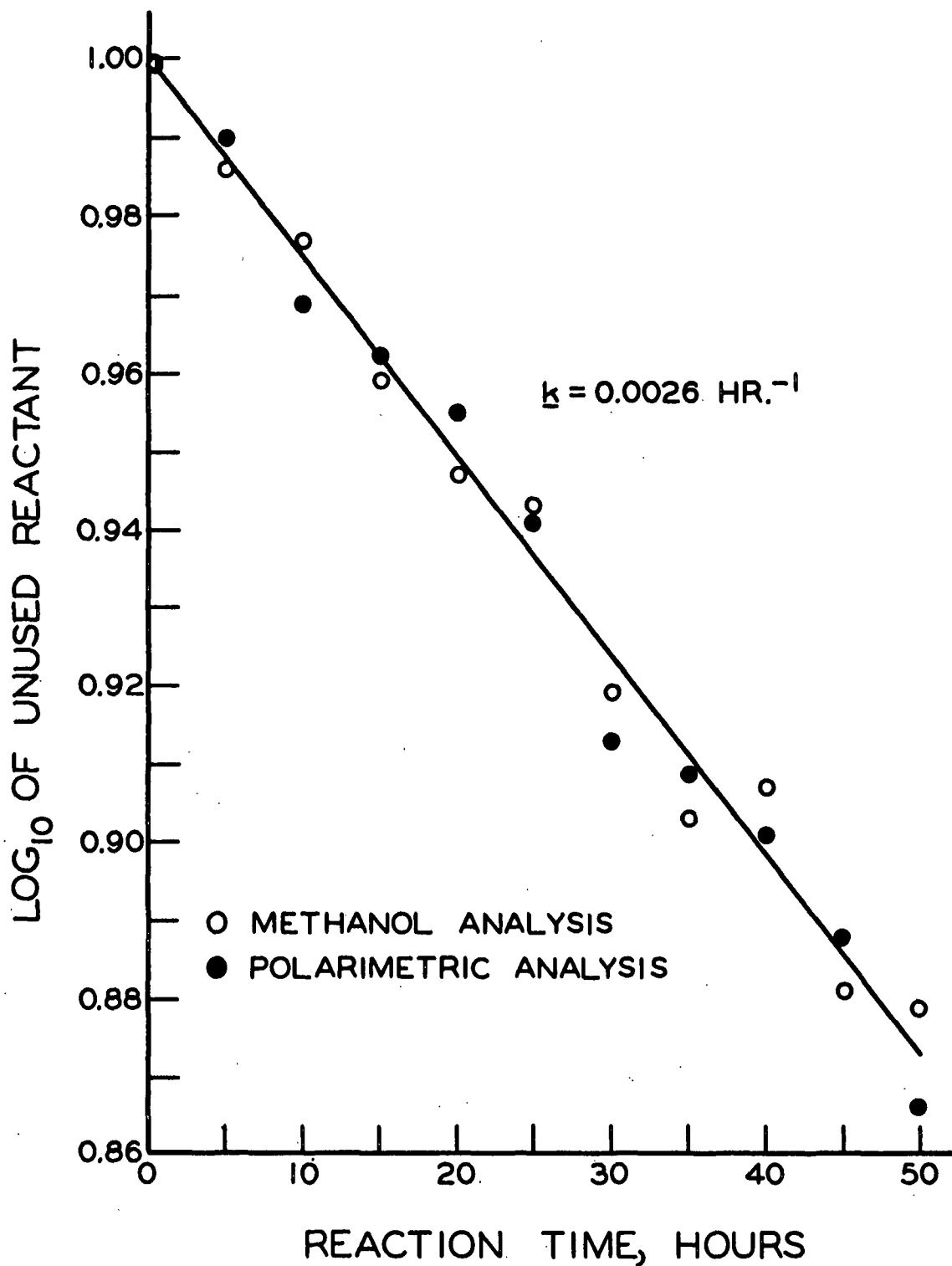


Figure 18. The Reaction of Methyl-β-Glucoside in 10% Sodium Hydroxide at 165°C. Under Nitrogen-Purged Conditions

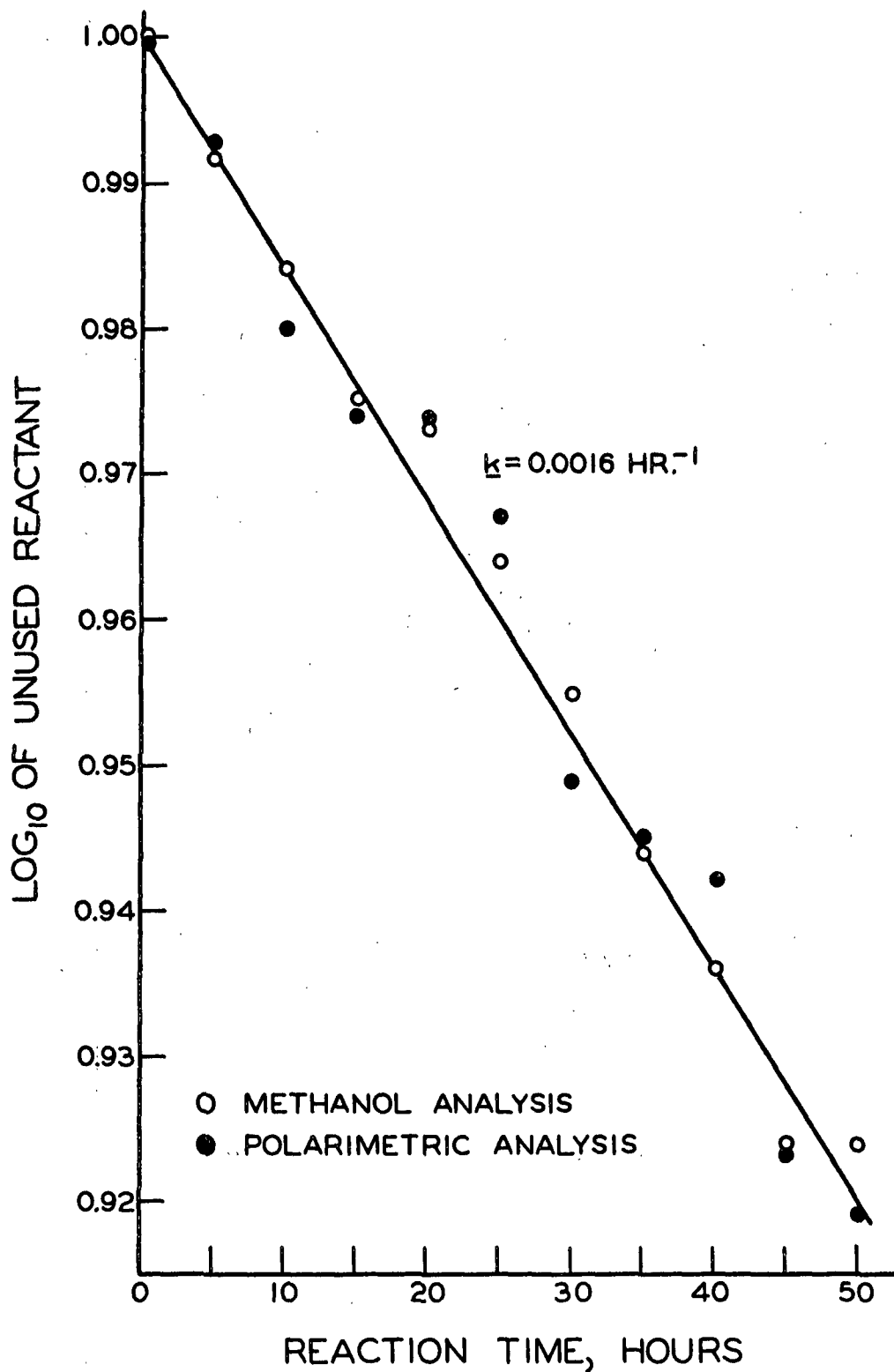


Figure 19. The Reaction of Methyl-β-Glucoside in 10% Sodium Hydroxide at 160°C. Under Nitrogen-Purged Conditions

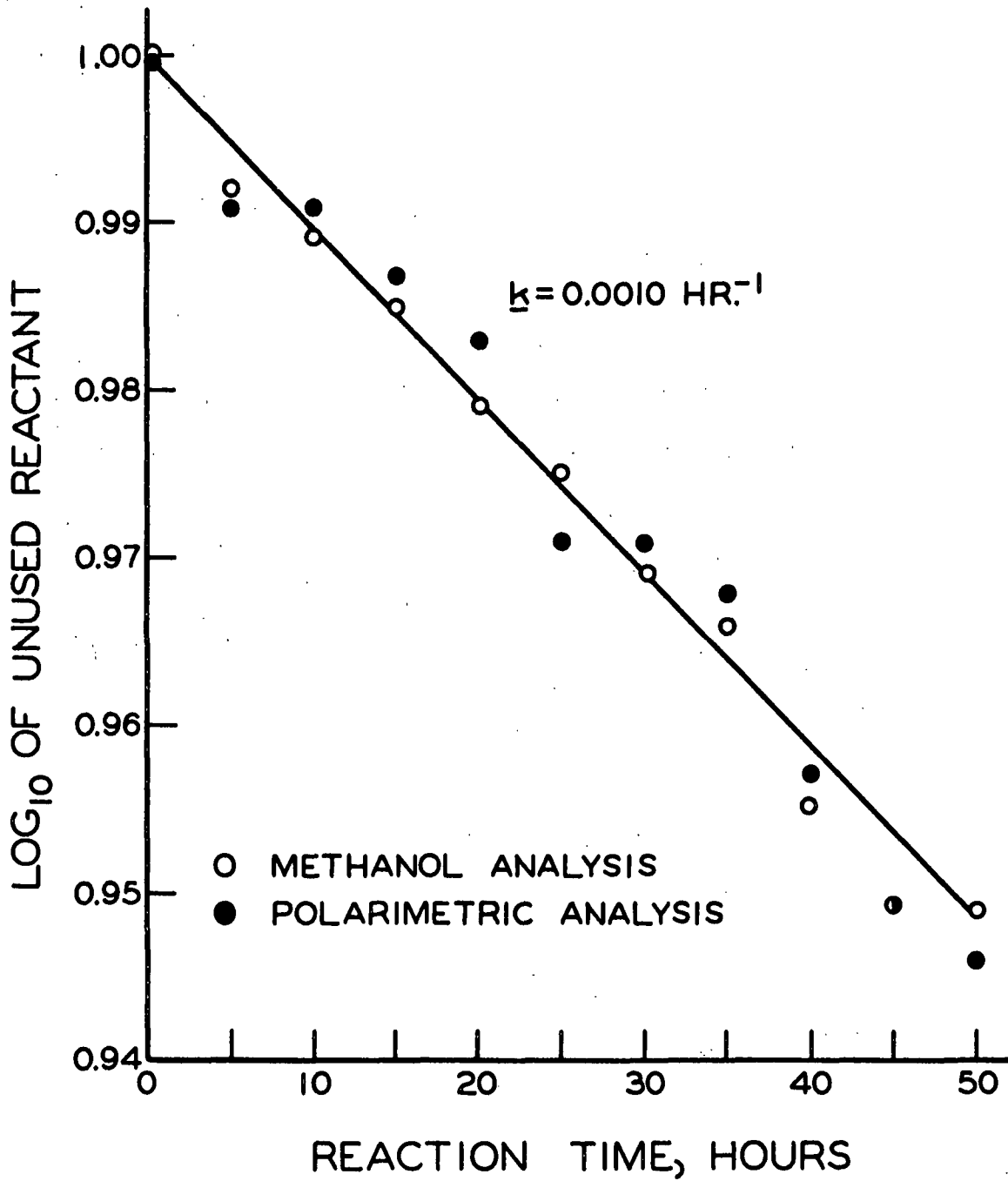


Figure 20. The Reaction of Methyl- β -Glucoside in 10% Sodium Hydroxide at 155°C. Under Nitrogen-Purged Conditions

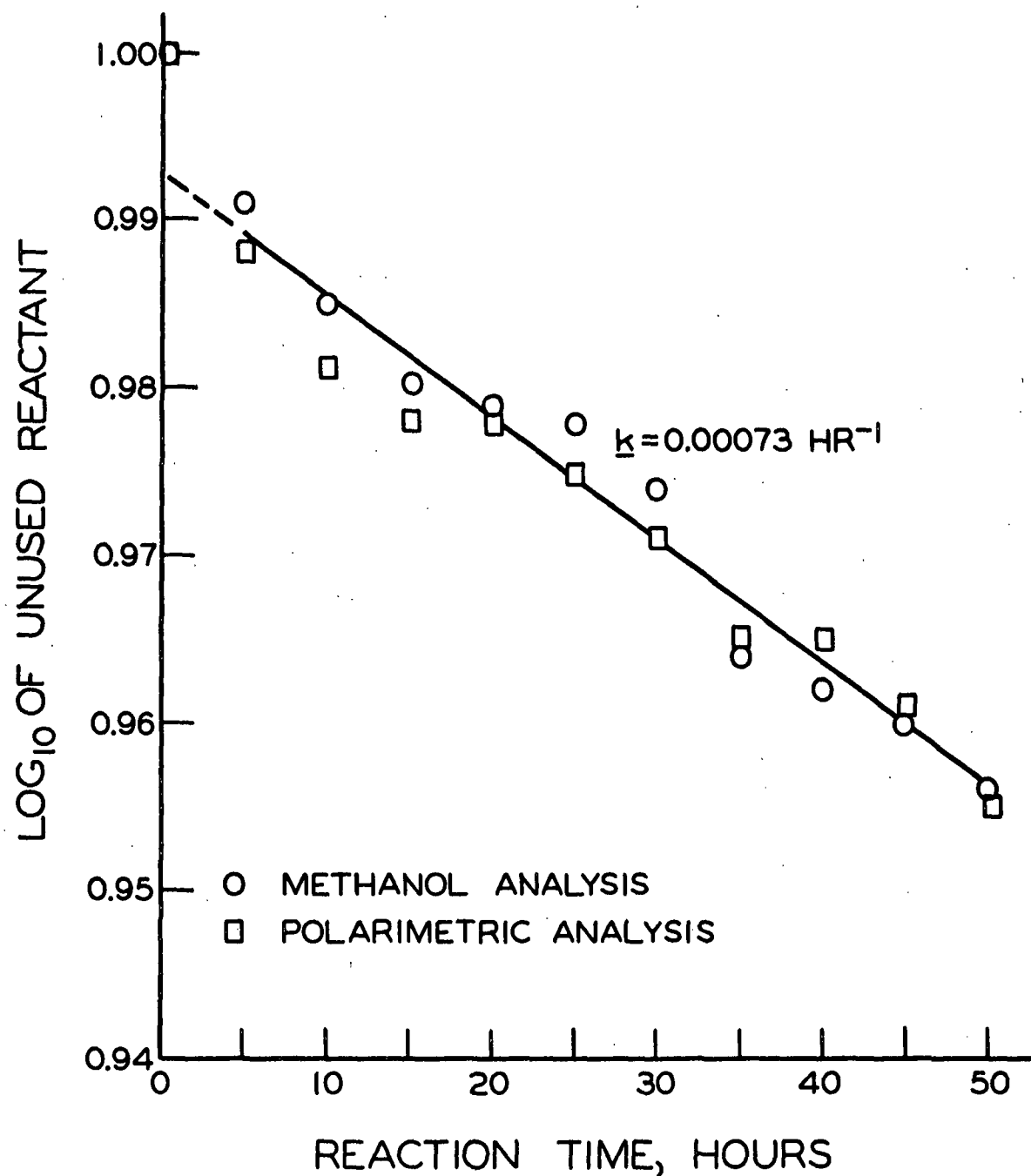


Figure 21. The Reaction of Methyl-β-Glucoside in 10% Sodium Hydroxide at 150°C. with a Molar Ratio of Molecular Oxygen to Methyl-β-Glucoside of 0.063 at Zero Reaction Time

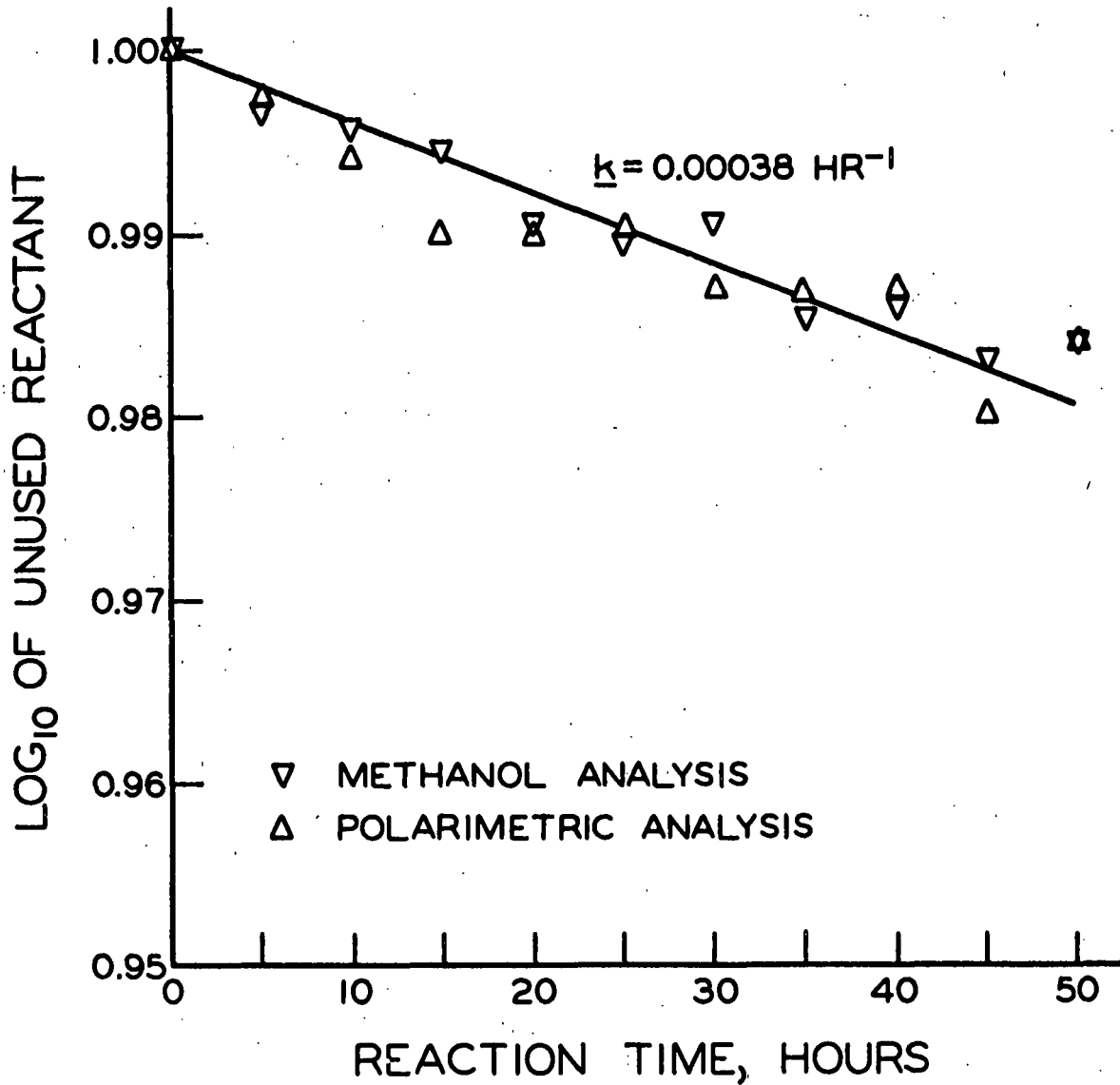


Figure 22. The Reaction of Methyl-β-Glucoside in 10% Sodium Hydroxide at 145°C. Under Nitrogen-Purged Conditions

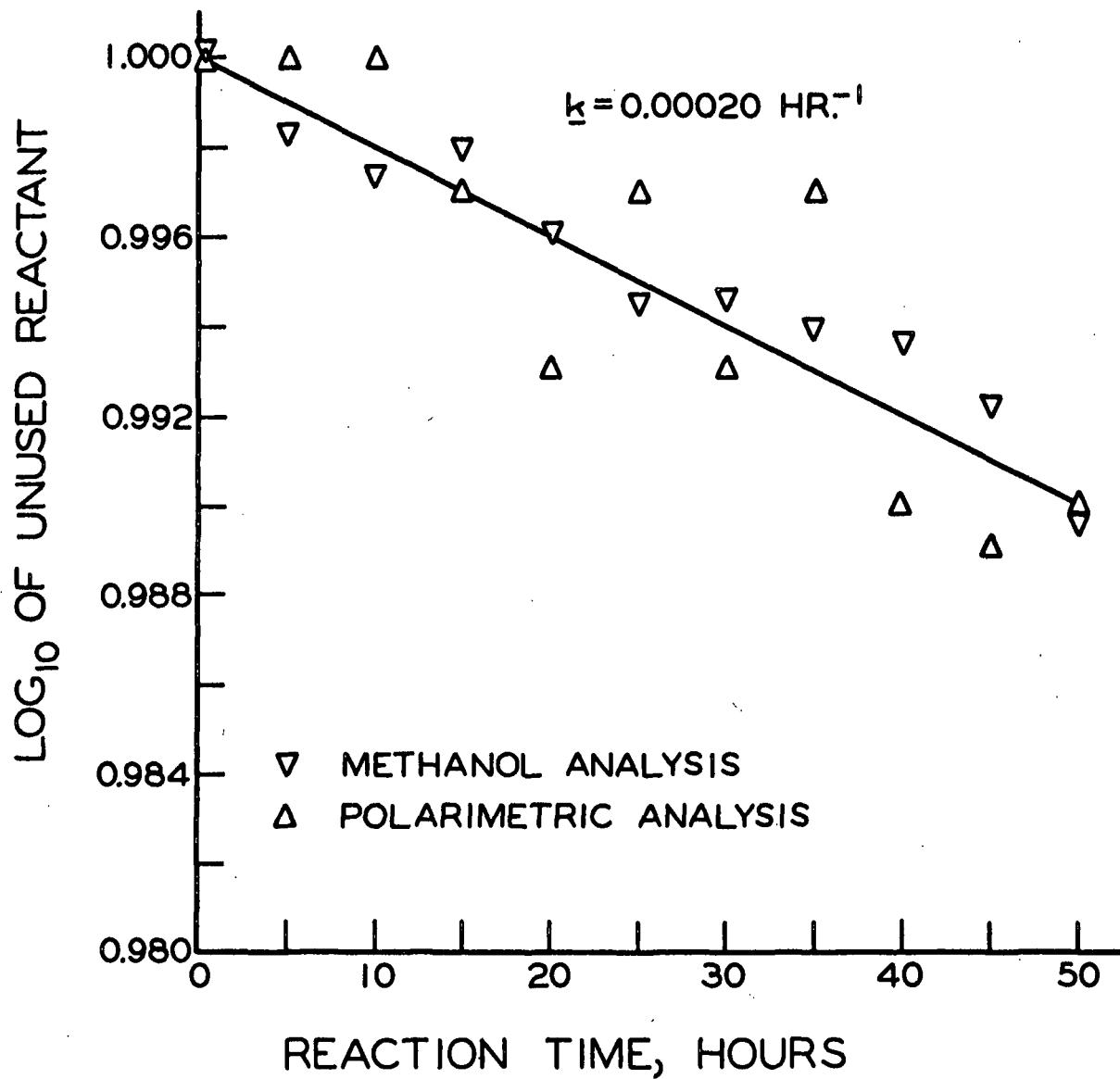


Figure 23. The Reaction of Methyl- β -Glucoside in 10% Sodium Hydroxide at 140°C. Under Nitrogen-Purged Conditions

TABLE IX

THE REACTION OF METHYL- β -GLUCOSIDE IN 10% SODIUM HYDROXIDE AT 170°C.

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %		Methanol Formed, $\mu\text{g.}/\text{ml.}$	
	O ₂ Initially Present ^a	N ₂ -Purged Conditions	O ₂ Initially Present ^a	N ₂ -Purged Conditions
0	100.0	100.0	0	0
5	92.2	96.0	250	200
10	88.8	89.7	430	380
15	86.7	85.5	510	470
20	81.1	79.4	770	640
25	74.8	78.0	810	830
30	73.4	76.0	960	950
35	69.2	69.0	1170	990
40	63.7	62.8	1290	1320
45	61.1	60.0	1380	1340
50	60.2	62.0	1500	1450

^aThe molar ratio of molecular oxygen to methyl- β -glucoside was 0.063 at zero reaction time.

TABLE X

THE INITIAL REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE AT 170°C.

Reaction Time, min.	Methanol Formed Under N ₂ -Purged Conditions, μ g./ml.	Methanol Formed with Molecular Oxygen Present ^a , μ g./ml.
0	0	0
10	-	36
20	-	57
30	-	77
40	-	83
50	-	99
60	29	119
70	-	125
80	-	126
90	50	135
100	-	148
120	79	-
150	91	-
180	119	-
210	129	-

^aThe molar ratio of molecular oxygen to methyl- β -glucoside was 0.063 at zero reaction time.

TABLE XI

THE INITIAL REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE AT 170°C.^a

Reaction Time, min.	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %	Methanol Formed, μ g./ml.
0	100.0	0
20	94.3	290
30	90.7	350
40	88.6	500
50	83.5	520
60	83.5	580
80	80.0	780
90	74.3	1000
110	72.2	1060
130	67.2	1220
140	65.0	1240

^aThe molar ratio of molecular oxygen to methyl- β -glucoside was 1.1 at zero reaction time.

TABLE XII

THE REACTION OF METHYL- β -GLUCOSIDE IN 10% SODIUM HYDROXIDE AT 170°C.

Reaction Time	Methanol Formed with Molecular Oxygen/Methyl- β -Glucoside = 0.063 at Zero Reaction Time, $\mu\text{g.}/\text{ml.}$		Methanol Formed with Molecular Oxygen/Methyl- β -Glucoside = 0.24 at Zero Reaction Time ^a , $\mu\text{g.}/\text{ml.}$	
	Standard Reaction	Reaction Vessels and Solutions Re-aerated at 100-Min. Reaction Time		
0	0	0	0	0
10 min.	-	-	45	43
20 min.	-	-	79	96
30 min.	-	-	111	123
40 min.	-	-	132	150
50 min.	-	-	177	190
60 min.	-	-	250	-
70 min.	-	-	260	230
80 min.	-	-	260	280
90 min.	-	-	280	-
100 min.	-	-	370	340
120 min.	150	200	370	330
140 min.	170	230	430	-
150 min.	-	-	-	450
160 min.	170	250	460	-
180 min.	190	260	460	-
200 min.	200	280	500	-
220 min.	210	290	510	-
240 min.	230	310	-	-
4 hr.	-	-	510	-
4-1/2 hr.	-	-	520	-
5 hr.	-	-	550	-
6 hr.	-	-	570	-
7 hr.	-	-	600	-
8 hr.	-	-	690	-
9 hr.	-	-	730	-
10 hr.	-	-	740	-

^aThe two columns of data below were derived from the reaction under the same conditions run at two different times, not from two analyses of the same data run.

TABLE XIII

THE REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE UNDER NITROGEN-PURGED CONDITIONS AT 165°C.

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %	Methanol Formed, μ g./ml.
0	100.0	0
5	97.8	117
10	93.2	190
15	91.7	330
20	90.2	420
25	87.2	450
30	81.8	630
35	81.1	730
40	79.6	710
45	77.2	880
50	73.5	890

TABLE XIV

THE REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE UNDER NITROGEN-PURGED CONDITIONS AT 160°C.

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %	Methanol Formed, μ g./ml.
0	100.0	0
5	98.5	66
10	95.5	132
15	94.1	200
20	94.1	220
25	92.6	290
30	89.0	360
35	88.2	440
40	87.4	510
45	83.8	590
50	83.0	590

TABLE XV

THE REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE UNDER NITROGEN-PURGED CONDITIONS AT 155°C.

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %		Methanol Formed, μ g./ml.
0	100.0		0
5	97.7		66
10	97.7		92
15	96.8		124
20	96.1		172
25	93.6		205
30	93.6		250
35	92.9		280
40	90.5		360
45	88.9		400
50	88.1		400

TABLE XVI

THE REACTION OF METHYL- β -GLUCOSIDE IN 10% SODIUM HYDROXIDE AT 150°C.

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %		Methanol Formed, μ g./ml.	
	O ₂ Initially Present ^a	N ₂ -Purged Conditions	O ₂ Initially Present ^a	N ₂ -Purged Conditions
0	100.0	100.0	0	0
5	97.2	100.0	73	29
10	95.8	98.0	124	55
15	95.0	96.5	165	84
20	95.0	97.2	176	124
25	94.3	96.5	183	132
30	93.6	94.4	210	194
35	92.2	93.7	290	220
40	92.2	92.9	310	240
45	91.5	92.9	320	250
50	90.2	92.2	360	270

^aThe molar ratio of molecular oxygen to methyl- β -glucoside was 0.063 at zero reaction time.

TABLE XVII

THE REACTION OF METHYL- β -GLUCOSIDE AT 150°C.
IN A CARBONATE-BICARBONATE BUFFERED SYSTEM OF pH 10.2
UNDER NITROGEN-PURGED CONDITIONS

Reaction Time, hours	pH	Methanol Formed, μ g./ml.	Unused Methyl- β -Glucoside Characterized Polarimetrically, %
0	10.2	0	100.0
5	10.2	3	100.0
10	10.2	5	99.3
15	10.2	8	100.0
20	10.2	9	100.0
25	10.2	7	100.0
30	10.2	9	100.7
35	10.2	8	100.0
40	10.2	11	100.0
45	10.2	12	100.0
50	10.2	14	99.3
55	10.2	12	99.3
60	10.2	12	100.7
65	10.2	18	100.7
70	10.2	19	100.7

TABLE XVIII

THE INITIAL REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE AT 150°C.

Reaction Time, min.	Methanol Formed Under N ₂ -Purged Conditions, $\mu\text{g./ml}$	Methanol Formed with Molecular Oxygen Present ^a , $\mu\text{g./ml}$.
0	0	0
10	-	16
20	-	24
30	-	33
40	-	38
50	-	49
60	-	50
70	-	59
80	-	62
90	14	-
100	-	58
120	18	67
140	-	74
150	18	-
160	-	80
180	18	75
200	-	80
210	28	-
240	26	-
270	31	-

^aThe molar ratio of molecular oxygen to methyl- β -glucoside was 0.063 at zero reaction time.

TABLE XIX

THE REACTION OF METHYL- β -GLUCOSIDE AT 150°C.
WITH A MOLAR RATIO OF MOLECULAR OXYGEN TO METHYL- β -GLUCOSIDE OF 1.1
AT ZERO REACTION TIME

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %		Methanol Formed, μ g./ml.	
	Buffered to pH 10.2 ^a	10% NaOH	Buffered to pH 10.2 ^a	10% NaOH
0	100.0	100.0	100.00	100.0
1	100.0	93.3	99.73	94.0
2	99.3	85.2	99.29	86.6
3	100.0	77.7	98.36	79.0
4	99.3	74.7	98.06	73.5
5	97.8	69.4	97.76	69.4
6	97.8	68.7	97.32	68.1
7	97.8	67.9	97.13	68.3
8	96.3	67.2	96.56	68.3
9	96.3	67.9	95.22	67.5
10	95.5	67.9	94.71	68.1

^aThe pH was exactly 10.2 at all reaction times as previously noted in Table XV in a prior run.

TABLE XX

THE REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE UNDER NITROGEN-PURGED CONDITIONS AT 145°C.

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %	Methanol Formed, $\mu\text{g./ml.}$
0	100.0	0
5	99.3	27
10	98.6	37
15	97.8	45
20	97.8	82
25	97.8	87
30	97.0	77
35	97.0	120
40	97.0	112
45	95.6	138
50	96.3	130

TABLE XXI

THE REACTION OF METHYL- β -GLUCOSIDE
IN 10% SODIUM HYDROXIDE UNDER NITROGEN-PURGED CONDITIONS AT 140°C.

Reaction Time, hours	Unreacted Methyl- β -Glucoside Characterized Polarimetrically, %	Methanol Formed, $\mu\text{g./ml.}$
0	100.0	0
5	100.0	16
10	100.0	22
15	99.3	17
20	98.5	32
25	99.3	45
30	98.5	43
35	99.3	50
40	97.8	53
45	97.0	65
50	97.8	87