compound separated by crystallization from the above ethyl acetate-alcohol mixture in the same way as the dimethylpiperidinium bromide. There was thus obtained 0.59 g. of pure quaternary ammonium salt from 1.57 g. of the amine.

Summary

1. Halogenated aliphatic tertiary amines of the type $Br(CH_2)_nN(CH_3)_2$, in which n has the values 4, 5, 6 and 7 have been prepared.

2. These amines react with themselves to form cyclic quaternary ammonium salts in yields which indicate that ring formation is the principal reaction.

URBANA, ILLINOIS

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF THE OHIO STATE UNIVERSITY]

THE MECHANISM OF CARBOHYDRATE OXIDATION. XI. THE ACTION OF POTASSIUM HYDROXIDE ON MALTOSE

BY WILLIAM LLOYD EVANS AND MARJORIE PICKARD BENOY

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The fact that maltose is a disaccharide makes a study of its behavior toward aqueous solutions of potassium hydroxide a much more complex problem than is the case with the monosaccharides for the reason that it may react in any one or all of three general ways. (a) The molecule of maltose may undergo hydrolysis[1] into two molecules of glucose. In this case one should expect the experimental data obtained from equivalent solutions of glucose and maltose to be quantitatively the same. (b) Since maltose is a reducing sugar like glucose, there exists the possibility of the disaccharide molecule undergoing enolization[2] and subsequent decomposition without hydrolysis having taken place previously. (c) It is evident that both reactions may take place simultaneously, the extent to which the maltose is changed in either direction being probably dependent on such experimental factors as alkali concentration and temperature. To obtain data with reference to the behavior of maltose in alkaline solutions, when examined from this general standpoint, was the first objective of these experiments.

If maltose is found to react in accordance with possibility (b) or (c), the mechanism of the alkaline decomposition of the disaccharide might depend upon the presence of an equilibrated mixture of six glucosido-hexoses, maltose and its isomers, analogs of the sugars in the well-known de Bruyn and van Ekenstein systems. These would be maltose (4-glucosido-*d*-glucose), 4-glucosido-*d*-mannose, 4-glucosido-*d*-fructose, 4-glucosido-pseudo-*d*-fructose, 4-glucosido-*d*-α-glutose and 4-glucosido-*d*-β-glutose.[3] Under the

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[1] Lewis and Buckborough, THIS JOURNAL, **36**, 2385 (1914).

[2] Nef, *Ann.*, **403**, 381 (1914).

[3] Nef, *ibid.*, **403**, 382 (1914).

conditions employed in our experiments, these glucosido sugars should react to give a series of 4-glucosido-hexose enediols. It is conceivable that these maltose isomers together with the 4-glucosido-hexose enediols and also the hexose sugars arising from any possible hydrolysis of the 4-hexosido-hexose sugars might form in these reaction mixtures a more general equilibrated condition which could be influenced by such experimental factors as the alkali concentration and the temperature.[4] To obtain some experimental data with reference to this latter point of view was the second objective of this study.

Experimental Part

The general procedures used in these experiments were exactly the same as those described in previous reports[5] from this Laboratory concerning the action of aqueous solutions of potassium hydroxide on the trioses and certain hexoses.

Materials. (a) **Carbohydrates.**—Maltose-monohydrate, *d*-glucose and *d*-mannose were purchased on the market and were found to be of the highest degree of purity.

(b) **Reagents.**—All reagents used in the various experimental operations were examined by the usual laboratory procedures as to their fitness for this study.

Manipulations. (a) **Maltose and Potassium Hydroxide.**—A 0.25 molar solution of maltose monohydrate was made by dissolving 2.251 g. of the disaccharide in 25 cc. of potassium hydroxide solution of the desired normality. This was agitated for forty-eight hours in a thermostat at the desired temperature. During this time the flask was kept closed with a well-fitting rubber stopper. At the end of the reaction period the solution was worked up for formic, acetic and lactic acids.

(b) **Analytical Methods.**—The necessary qualitative and quantitative methods have been described previously.

(c) **Determination of Mannose Phenylhydrazone.**—A portion of mannose weighing 2.251 g. was dissolved in a mixture of 25 cc. of potassium hydroxide solution of such strength as to give the final desired normality, 17.5 cc. of 95% alcohol and 5 cc. of phenylhydrazine. After this reaction mixture had been agitated in a thermostat at 25° for two hours, a light-colored precipitate was observed to separate out. At the end of forty-eight hours the solid was filtered, washed with acetone to remove any pyruvic aldehyde osazone, dried *in vacuo* at 55° and then weighed. The product was of a pale buff color. Samples were also similarly prepared using an equivalent weight of *d*-glucose. These were arranged into two groups: (1) the first group consisted of samples all of the same alkalinity (0.105 *N*), which were used as a time run, specimens being removed at intervals of several days, filtered, washed and weighed; (b) the second group of samples of varying alkali normalities was removed from the thermostat at the end of fourteen days, filtered, washed with acetone, dried and weighed. An exactly similar procedure was carried out with maltose, the time being twenty-six days.

The material was identified as mannose hydrazone by the known solubility of the compound in various reagents and by a comparison of its

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[4] *Cf.* Evans, Edgar and Hoff, THIS JOURNAL, **48**, 2666 (1926).

[5] (a) Evans, Edgar and Hoff, *ibid.*, **48**, 2665 (1926); (b) Evans and Hass, *ibid.*, **48**, 2703 (1926); (c) Evans and Cornthwaite, *ibid.*, **50**, 486 (1928); (d) Evans and Hutchman, *ibid.*, **50**, 1496 (1928); (e) Evans and O'Donnell, *ibid.*, **50**, 2543 (1928); (f) Evans, *Chem. Reviews*, **6**, 281 (1929).

melting point with that of mannose hydrazone prepared from pure *d*-mannose in cold acetic acid solution. Our results on the latter method of identification are the following.

Mannose hydrazone from glucose	178–180°
Mannose hydrazone from mannose	178–180°
Mixture of the above two	182°
Mannose hydrazone from mannose	181°
Mixture of mannose hydrazone from both mannose and maltose	192°
Mannose hydrazone from mannose	190°

The brackets indicate that the samples were heated simultaneously. Because of the known variation in melting points of hydrazones and osazones with a change in the rate of heating, all such determinations recorded in our experiments were made by direct comparison with known material. The agreement between these two was considered much more significant than that with values recorded in the literature.[6] The melting point of mannose hydrazone recorded by Reiss, Tollens and Gans[7] is 186–188°, and that by Fischer is 196–200°.

The effect of varying the rate of heating is clearly shown in the following test carried out on some pure mannose-hydrazone used as a standard of comparison.

Rate of heating, degrees/minute	4	7	8
Melting point, °C.	178–180	183–186	190

The mannose hydrazone was a pale cream color when first precipitated, a pale buff after one crystallization from water and a decided yellow after a second crystallization. The color deepened to an orange if the material was allowed to stand overnight in contact with water. It came out of alcohol solution much lighter in color. Corrections were made for the solubility of the mannose phenylhydrazone in the data given in Fig. 6.

(d) **Pyruvic Aldehyde.**—Pyruvic aldehyde was determined as an osazone by the general method described by Evans and Hass.[5b]

Experimental Results.—The quantitative data obtained from our reaction mixtures are shown in graphical form in Figs. 1–6.

Theoretical Part

In attempting to outline the decomposition mechanism of maltose in alkaline solutions, one is confronted with an appalling lack of knowledge with reference to the possible complex reaction products. More particularly, the difficulties in dealing with the experimental data obtained from the action of aqueous solutions of potassium hydroxide on maltose lie in the following facts: (a) the five theoretically possible 4-glucosido-hexoses,

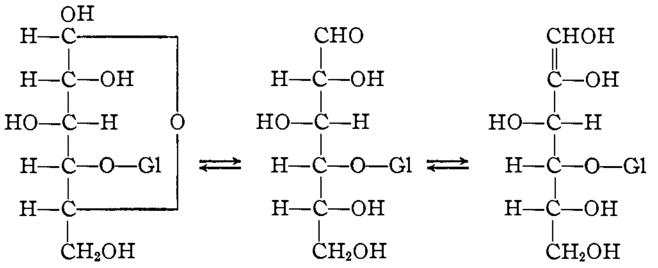
[6] Fischer, *Ber.*, **21**, 987 (1888); **22**, 1156 (1889).

[7] Reiss, Tollens and Gans, *ibid.*, **22**, 610 (1889).

isomers of maltose, are unknown; (b) there is also much work to be done in the development of sharp methods for the quantitative separation of many known organic products which are obtained by the action of alkalies on the sugars. The recently established proof[8] that maltose is 4-glucosido-*d*-glucose increases the complexity of the problem. Maltose can no longer be regarded as glucose with an inactive glucosido section attached in such a way as not to interfere with the activity of the glucose portion of the molecule. The attachment at the fourth carbon atom makes a vital difference because it limits the extent of the enediolization. The yields of pyruvic aldehyde osazone, mannose hydrazone, lactic and acetic acids obtained by the interaction of maltose and potassium hydroxide at 25 and 50° are smaller than those obtained from equivalent solutions of glucose under exactly the same experimental conditions, while those of formic acid are greater. An[8a] examination of these facts seems to show that their quantitative aspects are best understood on the assumption that maltose, in alkaline solution, reacts to form a series of 4-glucosido-glucose enediols which may rupture at the double bond. The products obtained by such a fission may then react with the alkali present much after the well-known behavior of the hexose sugars under similar experimental conditions.

The following explanation is offered to account for the relative yields of reaction products found in our experiments on the alkaline decomposition of maltose when compared to those from glucose.

(a) **Maltose-1,2-enediol (4-Glucosido-glucose-1,2-enediol).**—The formation of maltose-1,2-enediol may be shown by the following equation



(1)

Maltose Maltose, aldehydo form[9] Maltose-1,2-enediol
Gl = Glucosido

A rupture of the maltose-1,2-enediol (4-glucosido-glucose-1,2-enediol) would result in the formation of 3-glucosido-arabinose and hydroxymethylene, as is shown in the reaction

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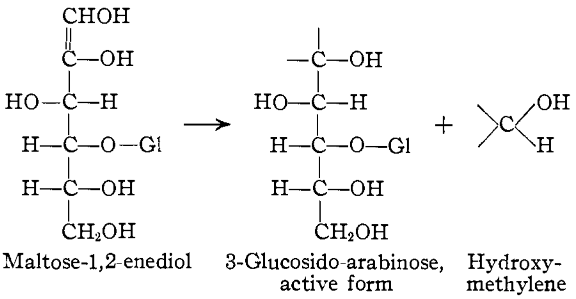
[8] Irvine and Black, *J. Chem. Soc.*, **129**, 862–875 (1926); Cooper, Haworth and Peat, *ibid.*, **129**, 876 (1926); Levene and Sobotka, *J. Biol. Chem.*, **71**, 471–475 (1927); Zemplén, *Ber.*, **60B**, 1555–1564 (1927).

[8a] *Cf.* K. Bernhauer, *Biochem. Z.*, **210**, 175, 1929.

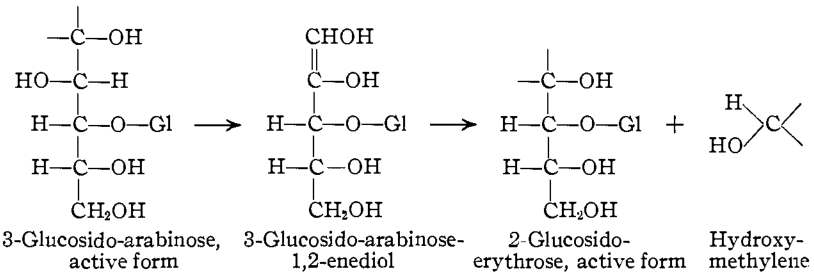
[9] Wolfrom, This Journal, **51**, 2188 (1929).



(2)

If sufficiently mild oxidizing agents were present, it is clear that 3-glucosido-arabonic and formic acids would be the reaction products. In the presence of potassium hydroxide maltose is oxidized by potassium permanganate to oxalic acid and carbon dioxide together with traces of acetic acid. The general relationship of the quantities of reaction products to the alkali concentration in the oxidation is in general similar to that of glucose.[10]

The 3-glucosido-arabinose thus formed may then in turn undergo an enediol formation with subsequent splitting into hydroxymethylene and 2-glucosido-erythrose. This change is shown in the equation



(3)

Under appropriate oxidizing conditions 2-glucosido-erythronic acid should be formed at this point. In connection with the possible formation of the 3-glucosido-arabinose and 2-glucosido-erythrose, it is of much interest to point out that Glattfeld and Hanke[11] oxidized maltose in an alkaline solution with air and with hydrogen peroxide. When the glucosido acids which they obtained as reaction products were hydrolyzed, they identified erythronic acid lactone and arabonic acid, the latter being recognized by means of its phenylhydrazide.

If the enediol formation can proceed no further, *i. e.*, if the CHO-Gl group remains intact, then it is possible for the 2-glucosido-erythrose to undergo hydrolysis with the formation of erythrose and glucose. This change is shown in the equation

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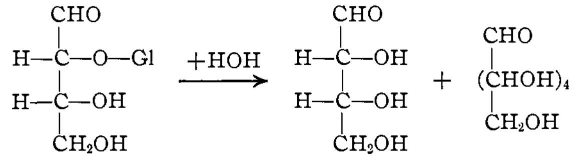
[10] Unpublished results.

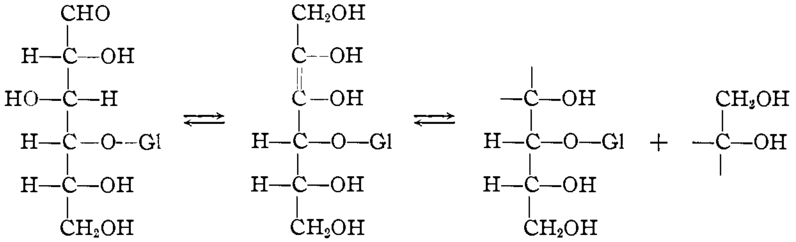
[11] Glattfeld and Hanke, THIS JOURNAL, **40,** 992 (1918).



$$\begin{array}{c}\mathrm{CHO}\\|\\\mathrm{H{-}C{-}O{-}Gl}\\|\\\mathrm{H{-}C{-}OH}\\|\\\mathrm{CH_2OH}\end{array}\xrightarrow{+\mathrm{HOH}}\begin{array}{c}\mathrm{CHO}\\|\\\mathrm{H{-}C{-}OH}\\|\\\mathrm{H{-}C{-}OH}\\|\\\mathrm{CH_2OH}\end{array}+\begin{array}{c}\mathrm{CHO}\\|\\\mathrm{(CHOH)_4}\\|\\\mathrm{CH_2OH}\end{array}\qquad(4)$$

2-Glucosidoerythrose Erythrose Glucose

(b) **Maltose-2,3-enediol (4-Glucosido-glucose-2,3-enediol).**—As the temperature of the reaction and the concentration of the alkali are increased, there should be more of a tendency for the general equilibrated condition present in the maltose solution to shift in the direction of forming a maltose-2,3-enediol, *i. e.*, 4-glucosido-glucose-2,3-enediol. Its behavior in this respect would be represented by the reaction



$$\begin{array}{c}\mathrm{CHO}\\|\\\mathrm{H{-}C{-}OH}\\|\\\mathrm{HO{-}C{-}H}\\|\\\mathrm{H{-}C{-}O{-}Gl}\\|\\\mathrm{H{-}C{-}OH}\\|\\\mathrm{CH_2OH}\end{array}\rightleftharpoons\begin{array}{c}\mathrm{CH_2OH}\\|\\\mathrm{C{-}OH}\\\|\\\mathrm{C{-}OH}\\|\\\mathrm{H{-}C{-}O{-}Gl}\\|\\\mathrm{H{-}C{-}OH}\\|\\\mathrm{CH_2OH}\end{array}\rightleftharpoons\begin{array}{c}|\\\mathrm{{-}C{-}OH}\\|\\\mathrm{H{-}C{-}O{-}Gl}\\|\\\mathrm{H{-}C{-}OH}\\|\\\mathrm{CH_2OH}\end{array}+\begin{array}{c}\mathrm{CH_2OH}\\|\\\mathrm{{-}C{-}OH}\\|\end{array}\qquad(5)$$

Maltose, aldehydo form Maltose-2,3-enediol 2-Glucosido-erythrose, active form Glycol-aldehyde, active form

Then it is clear that the 2-glucosido-erythrose thus formed would also yield glucose and erythrose as shown in Equation 4. If the mechanism of the alkaline decomposition of maltose is correctly set forth in these equations, then it is possible to offer a satisfying explanation for the relative amounts of lactic, acetic and formic acids and pyruvic aldehyde when this disaccharide is treated with aqueous solutions of potassium hydroxide under the same conditions of alkali normality and temperature as the monosaccharide, glucose.

Lactic Acid.—From the above-outlined decomposition of the maltose molecule it is seen that one molecule of the disaccharide may yield the following general results: (a) two molecules of hydroxymethylene, one molecule of erythrose and one molecule of glucose (Equations 1, 2, 3 and 4); (b) or it may yield one molecule of glycol aldehyde, one molecule of erythrose and one molecule of glucose (Equations 5 and 4). From a study of these possibilities it may be shown that only one-half of the maltose molecule yields lactic acid and that that portion of the disaccharide molecule is the glucosido section.[11a]

It has been established recently that the yields of lactic acid obtained from alkaline solutions of glucose are a function of both the alkali concentration and the temperature of the reacting system. An examination of

[11a] *Cf.* H. Wolff, *Biochem. Z.*, **210**, 459 (1929).

Fig. 1 will show that the general characteristics of lactic acid formation in alkaline solutions of maltose are also practically the same as those obtained from glucose except that the yield is less in the case of the disaccharide, *i. e.*, it tends to be somewhat less than one-half at both temperatures.

Nef reached the conclusion that the tetroses in alkaline solutions did not give rise to the formation of a 1,2-enediol, *i. e.*, $CH_2OHCHOHC(OH)=CHOH$. The formation of this enediol would be essential because on fission it would yield glyceric aldehyde, a necessary intermediate in the production of lactic acid in alkaline solutions of the sugars. It is quite possible that the speed of rearrangement of the 1,2-enediol to the corresponding saccharinic acid is too rapid when compared to that of glyceric aldehyde formation. Nef came to his point of view because he found that *glycol aldehyde*[12] *did not yield the least trace of lactic acid when it was treated with 8 N potassium hydroxide solution.* However, he did obtain significant amounts of four carbon atom saccharinic acid (1,3-dihydroxybutyric acid), a fact which shows that a tetrose sugar had formed under the conditions of his experiments.[13] These general findings were confirmed in this Laboratory, although alkaline solutions were employed of lesser normality than those used by Nef.

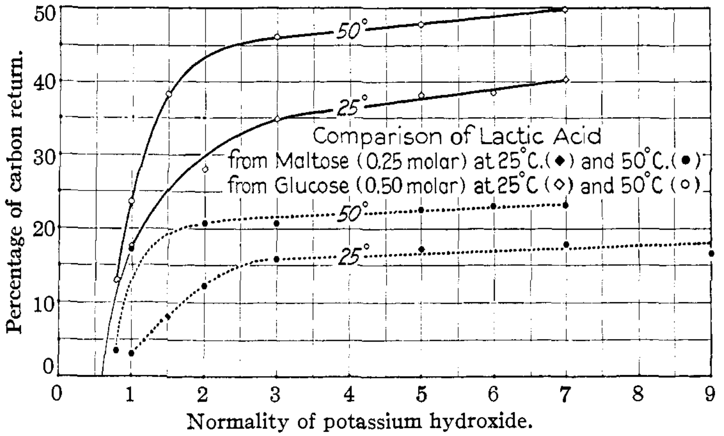


Fig. 1.

By reason of the facts pointed out in the two preceding paragraphs, namely, that glucose in alkaline solution will yield lactic acid and that erythrose under the same conditions will not do so, it is clear that on the basis of the above-outlined decomposition of maltose the conclusion must be reached that the source of the lactic acid in this reaction is the glucosido section of the disaccharide molecule and not the glucose section.

[12] Nef, *Ann.*, **376**, 40 (1910).

[13] *Cf.* Fischer and Landsteiner, *Ber.*, **25**, 2553 (1892).

Since 0.25 molar maltose (equivalent to 0.50 molar glucose) and 0.50 molar glucose solutions were used, it is now evident that the lactic acid yields from maltose should tend to be one-half those which could be obtained from glucose. On the basis that the lactose molecule is 4-galactosido-glucose, it may be predicted safely that this well-known sugar will not yield much lactic acid when treated with aqueous solutions of potassium hydroxide under the same conditions that were used in our maltose study. This arises from the fact that the hexosido section of the milk sugar molecule[3] does not give rise to much lactic acid in alkaline solutions, while the hexose seticon of this sugar gives rise to a tetrose which, as is pointed out above, does not give the 1,2-enediol which is necessary for lactic acid formation. Mr. Robert C. Hockett of this Laboratory is at present studying the behavior of the disaccharides toward aqueous solutions of potassium hydroxide.

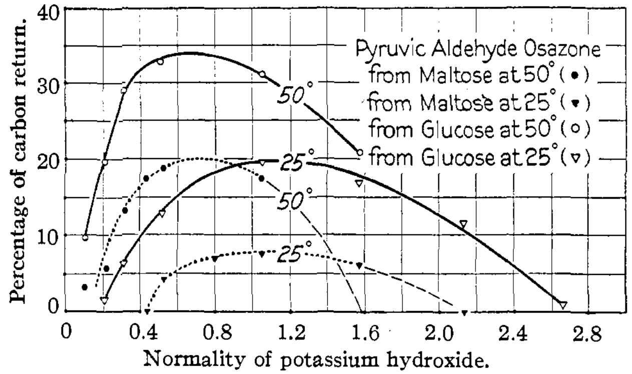
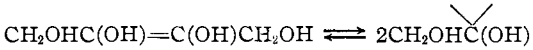


Fig. 2.

Should 3-glucosido-arabinose hydrolyze to any great degree, the yields of lactic acid should tend to be greater than those which we found, because Mr. Rollin F. Conaway of this Laboratory has shown recently that arabinose in alkaline solutions will yield pyruvic aldehyde, which changes under these conditions to lactic acid.

Pyruvic Aldehyde Osazone.—If the above reactions of maltose take place, then the disaccharide in alkaline solution should give rise to yields of pyruvic aldehyde osazone in amounts which approximate one-half those obtained from equivalent solutions of glucose. By our experimental methods this was shown in general to be true (Fig. 2). Pyruvic aldehyde is the forerunner of lactic acid. It is derived from the glyceric aldehyde obtained through a splitting of the 3,4-enediol of the glucosido section of the maltose molecule. Alkaline solutions of glycol aldehyde do not give rise to pyruvic aldehyde osazone when the experiment is carried out under the same conditions as are used with sugars that do so react. Hence, any tetrose formed from glycol aldehyde will not give a 1,2-enediol, a reaction necessary for the production of glyceric aldehyde, which, in turn, is the forerunner of pyruvic aldehyde.

Formic Acid.—It has been pointed out previously that formic acid may arise from the action of alkalies on hydroxymethylene. In the above equations it is seen that the glucose section of the maltose molecule yields at least two molecules of hydroxymethylene. Since erythrose forms a 2,3-enediol in preference to a 1,2-enediol, then two molecules of glycol aldehyde (active form) will form according to the equation



$$CH_2OHC(OH){=}C(OH)CH_2OH \rightleftharpoons 2CH_2OH\overset{\diagdown\diagup}{C}(OH) \qquad (6)$$

It has been shown in this Laboratory that alkaline solutions of glycol aldehyde will yield formic acid. Since the glucosido section of the maltose molecule will yield formic acid also, then it becomes clear from all of these facts that alkaline solutions of maltose should produce more formic acid than do equivalent alkaline solutions of glucose. From the data in Fig. 3 this comparison is found to be true.

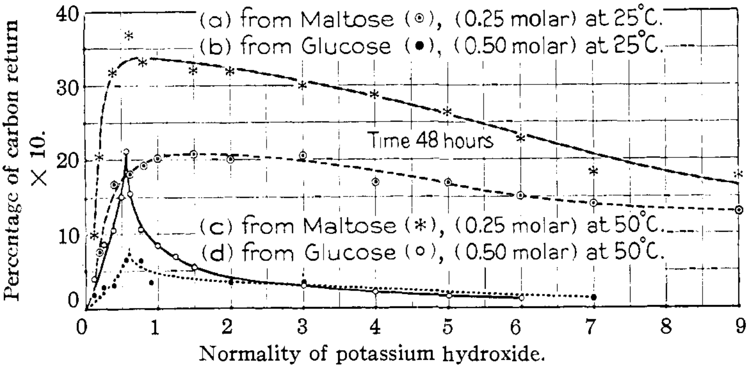


Fig. 3.—Comparison of formic acid.

Acetic Acid.—Our data, Fig. 4, with reference to acetic acid production in alkaline solutions of maltose show that this acid is found in smaller amounts than it is in alkaline solutions of glucose. The possible sources of acetic acid arising from the alkaline decomposition of maltose as given by the above equations may be these: (a) pyruvic aldehyde formed in the alkaline decomposition of the glucosido section of the maltose molecule; (b) the glycol aldehyde obtained in Equation 5 and also that arising from the splitting of the tetrose-2,3-enediol in Equations 4 and 5. Since not over 1% of glycol aldehyde carbon is returned as acetic acid under the conditions of these experiments, then from our data it may be fairly concluded that the principal source of acetic acid in these maltose reactions must be the glucosido section of the disaccharide molecule. From these statements it is clear that the amounts of acetic acid obtained from maltose must be less than those arising from equivalent solutions of glucose. Whether 2,3-dihydroxybutyric acid formed from the action of potassium hydroxide on the tetrose present will yield acetic acid has not yet been established.

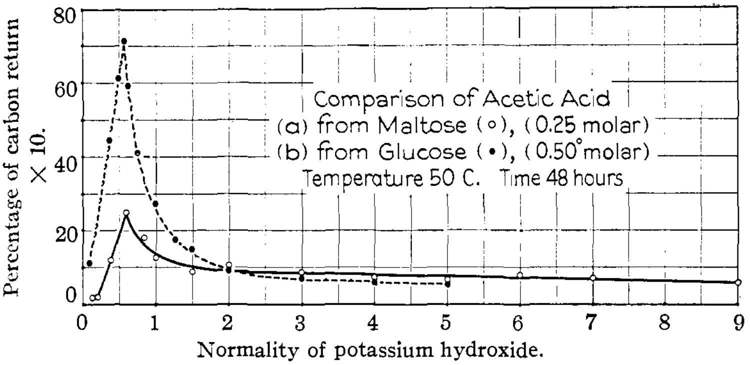


Fig. 4.

Production Relation of Reaction Products.—From Fig. 5 it is seen that the data obtained for pyruvic aldehyde osazone, lactic, acetic and formic acids from alkaline solutions of maltose are of the same general character as those which have been obtained previously in this Laboratory from glucose, mannose and fructose, a fact which would seem to support the points of view set forth in the above explanation of the mechanism involved in the alkaline decomposition of maltose.

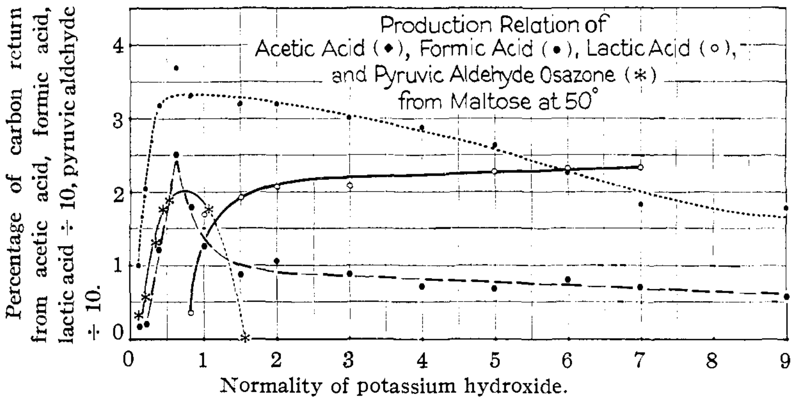


Fig. 5.

Effect of Temperature and Alkali Concentration.—Our data on the alkaline decomposition of maltose show that the effects of alkali concentration and increased temperature are of the same general type as those obtained with the hexose sugars.

The Hydrolysis of Maltose. (a) Mannose Hydrazone.—De Bruyn and van Ekenstein[14] obtained *d*-mannose phenylhydrazone from maltose after boiling it for three hours in a 0.166 *N* potassium hydroxide solution. Kolb[15] also obtained it at room temperature after the sugar had stood with a dilute barium hydroxide solution for a long period. It is clear that the mannose in these two cases might have arisen from the following sources. (1) Glucose arising from the possible hydrolysis of maltose would be converted into the well-known series of hexose sugars, one of which is mannose. (2) The theoretically possible 4-glucosido-mannose might be hydrolyzed into its possible monosaccharide units. The possibility of the existence of the 4-glucosido-mannose in an alkaline solution seems to be borne out by the fact that Lewis[16] obtained mannonic acid lactone from the hydrolysis of the glucosido products resulting from the oxidation of maltose with alkaline copper hydroxide. *To obtain mannose as its hydrazone from an alkaline solution of maltose would therefore seem to be proof that an hydrolysis of a 4-glucosido-hexose compound had occurred.* The results of our experiments are shown in Fig. 6. The first formation of a noticeable precipitate of mannose phenylhydrazone from maltose in 0.105 *N* potassium hydroxide solution occurred after eleven days at 25°. A corresponding sample of *d*-glucose under similar conditions showed some precipitate in five days, thus making it appear that the conversion of maltose into its monosaccharide

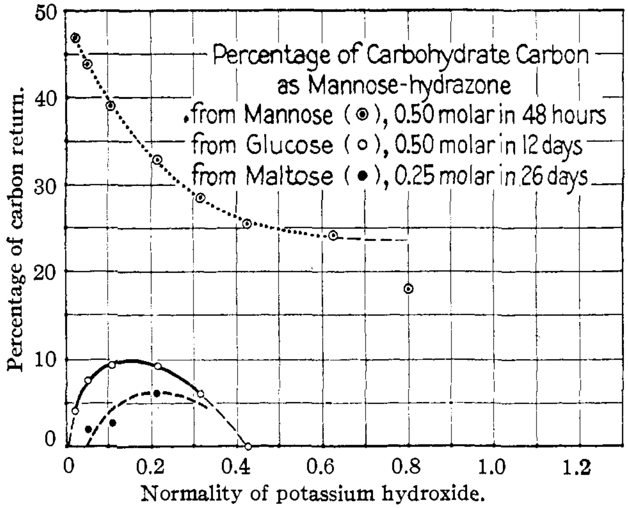


Fig. 6.

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[14] Lobry de Bruyn and Alberda van Ekenstein, *Rec. trav. chim.*, **18**, 147–149 (1899); *cf.* **15**, 92 (1896).

[15] Kolb, *Biochem. Z.*, **63**, 1–57 (1914).

[16] Lewis, *Am. Chem. J.*, **42**, 301–314 (1909).

units by either Mechanism 1 or 2 required about the same length of time as the formation of the mannose phenylhydrazone from glucose. The formation of the hydrazone directly from mannose is very rapid since precipitation began within two hours. In the long periods during which these sugars were exposed to the action of alkaline solutions, longer for maltose than for *d*-glucose, the mannose hydrazone was no doubt being decomposed, so that sharp comparisons between the curves cannot be made nor far-reaching conclusions drawn, but some interesting suggestions are contained in the data. (1) A comparison of corresponding points on the two curves shows that the maltose values are approaching those for *d*-glucose, and also that the hydrolysis of maltose or its glucosido isomers is a function of the alkalinity.[17] (2) The rise in the two curves shows that the de Bruyn and van Ekenstein equilibrium is disturbed by changes in the alkali normality. (3) The downward trend of these curves is thought to be due to the fact that the hexose enediols are beginning to undergo splitting at their double bonds, a reaction which would result in lowering the available concentration of the hexose sugars. It is a noteworthy fact in this connection that the maxima are between 0.15–0.20 N alkali, a value slightly above that (0.05 N) which Nef[18] indicated as the point where enediolic splitting first begins for the hexoses. The only significance of the mannose curve is that it marks off the regional limits in which we might hope to find the hydrazone in the case of the other sugars. This curve is taken as a measure of the decrease in the concentration of the aldehydo form of this sugar in the presence of an increase in the alkali normality. This seems to be in harmony with the point of view expressed by Wohl and Neuberg[19] and Fischer with reference to the non-formation of the hydrazones from the salts of the enediols.

(b) **Pyruvic Aldehyde Osazone.**—A comparison of the mannose phenylhydrazone curves from maltose and glucose with those of pyruvic aldehyde osazone obtained from these sugars seems to indicate very strongly that an increase in the concentration of the alkali causes a change in the equilibrium present which favors a splitting of the maltose enediols. Such a change should result in a decrease in the yields of derivatives obtained from 4-glucosido-hexose sugars and an increase in the products arising from the fission of glucosido sugars containing less than 12 carbon atoms. Attention is directed to the fact that the mannose hydrazone formation from both glucose and maltose ceases at approximately the same alkalinity (0.42 N) as that at which pyruvic aldehyde formation begins at 25°.

[17] Nef, *Ann.*, **403**, 207 (1914).

[18] Nef, *ibid.*, **403**, 206 (1914).

[19] Wohl and Neuberg, *Ber.*, **33**, 3099 (1900); Fischer, *ibid.*, **28**, 1149 (1895); *cf.*, Zemplén and Kunz, *ibid.*, **56**, 1705 (1923).

Summary

1. Objectives of these experiments were as follows: (a) to determine whether maltose in alkaline solutions reacts in accordance with the views that postulate the presence of hexosido-hexose-enediols or whether it undergoes hydrolysis at the glucosido union; (b) to ascertain whether temperature and the concentration of the alkali used influence the character of the reaction products.

2. (a) Maltose was dissolved in aqueous solutions of potassium hydroxide of various normalities (0.1–9.0), the sugar concentration being 0.25 molar. After forty-eight hours of agitation at 25 and 50°, the reaction mixtures were quantitatively examined for acetic, lactic and formic acid. (b) Similar aqueous-alcoholic solutions of maltose, phenylhydrazine and potassium hydroxide were examined for pyruvic aldehyde osazone.

3. Maltose yields larger amounts of formic acid than glucose does under exactly the same conditions, while smaller amounts of pyruvic aldehyde osazone, lactic and acetic acids are obtained than from the hexose sugar.

4. These quantitative differences between maltose and glucose have been explained on the assumption that the disaccharide forms two enediols, namely, maltose-1,2-enediol (4-glucosido-glucose-1,2-enediol) and maltose-2,3-enediol (4-glucosido-glucose-2,3-enediol)). Since the glucosido linkage is on the fourth carbon atom, it is further assumed that the 4-glucosido-glucose-3,4-enediol does not rupture, if formed.

5. Maltose-1,2-enediol (4-glucosido-glucose-1,2-enediol) may split into hydroxymethylene and 3-glucosido-arabinose, and this latter product in turn may yield 2-glucosido-erythrose and hydroxymethylene. Maltose-2,3-enediol (4-glucosido-glucose-2,3-enediol) may split into 2-glucosido-erythrose and glycol aldehyde. The 2-glucosido-erythrose formed in both cases may undergo hydrolysis into erythrose and glucose.

6. In its alkaline decomposition by the above mechanism the maltose molecule tends to form one molecule of glucose (*i. e.*, one-half of the maltose molecule), one molecule of erythrose and two molecules of hydroxymethylene. Under these experimental conditions glucose is the only one of these products which will form lactic acid in alkaline solution. As shown experimentally the yields of this acid should tend toward a value one-half that of an equivalent solution of glucose.

7. Pyruvic aldehyde is derived from glyceric aldehyde, which must be derived from glucose or any possibly formed arabinose. Since erythrose and hydroxymethylene do not yield pyruvic aldehyde, the experimental yields of pyruvic aldehyde osazone may be less than those of equivalent solutions of glucose, *i. e.*, should tend toward one-half the values obtained from the hexose. This was found to be the case.

8. Formic acid may be obtained from the two molecules of hydroxymethylene, erythrose (the alkaline glycol aldehyde of these experiments)

and glucose. These combined sources should yield more formic acid than an equivalent solution of glucose. This was found to be true.

9. Acetic acid is obtained in much smaller amounts from glucose than it is from erythrose. On the basis of the above-outlined decomposition of maltose, less acetic acid should be obtained from it than from glucose. Our data are in harmony with this point of view.

10. An experiment was made with maltose in the presence of phenylhydrazine at the lower alkalinities, seeking mannose hydrazone as an index of hydrolysis, a control being run with glucose. Mannose hydrazone was first obtained from maltose in eleven days, at 25°; after nineteen days the production reached a constant value. The hydrazone was obtained between 0.05–0.42 *N* potassium hydroxide. The presence of mannose is thought to be due to its formation from the glucose obtained by the hydrolysis of maltose or of 4-glucosido·mannose at the lower alkalinities.

11. The assumption of the presence of glucosido-hexose enediols and their degradation products in alkaline solutions of maltose seems to be in harmony not only with our experimental data, but also with those of Lewis and Buckborough,[1] and Glattfeld and Hanke.

12. Our data seem to be in harmony with the view that the two kinds of decomposition open to the maltose molecule—enediolic splitting and hydrolysis—go on simultaneously in alkaline solution, but whereas the former is a rapid function of the time, the latter is a very slow one.

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ACETYL CHOLINE CHLORIDE

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Choline and its esters have achieved considerable importance in recent years due to their property of producing a marked vasodilation[1] and also because it has been shown that they are normally present in the serous coat of the intestine and probably are the normal peristaltic hormones.[2] The acetyl derivative is especially interesting since it has been found that it possesses a depressor effect 100,000 times as great as that produced by choline itself and yet it is only three times as toxic as this substance.[3]

Salts of acetyl choline have been found in certain natural products.[4] Baeyer and later Nothnagel synthesized the chloride by acetylating

[1] Fraenkel, "Arzneimittel-Synthese," Julius Springer, Berlin, **1927**, p. 336.

[2] Le Heux, *Arch. ges. Physiol. (Pflüger's)*, **173**, 8 (1918).

[3] Hunt and Taveau, *U. S. Hyg. Lab. Bull.*, No. 73 (1911); Hunt, *Pharmacol.*, **7**, 301 (1915).

[4] Ewins, *Biochem. J.*, **8**, 44 (1914); Boruttau and Cappenburg, *Arch. Pharm.*, **259**, 33 (1921); *C. A.*, **15**, 2692 (1921).