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PREPARATION AND PROPERTIES OF ALDONIC ACIDS AND THEIR LACTONES AND BASIC CALCIUM SALTS

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ABSTRACT

Directions are given for the preparation of the crystalline acids and lactones derived from gluconic, mannonic, galactonic, xylonic, arabonic, and rhamnonic acids. The compositions of the basic calcium salts of mannonic, galactonic, xylonic, arabonic, rhamnonic, lactobionic, and maltobionic acids are reported and their use for the purification of sugar acids is described. The utility of dioxane as a solvent in preparing the free sugar acids and their lactones is emphasized. The method for preparing gluconic acid by crystallization from water is suitable for the manufacture of that substance in large quantity.

CONTENTS

I. INTRODUCTION

The electrolytic method [1] for the oxidation of aldoses has simplified the production of aldonic acids so that the acids derived from the more abundant naturally-occurring sugars are available in large quantities

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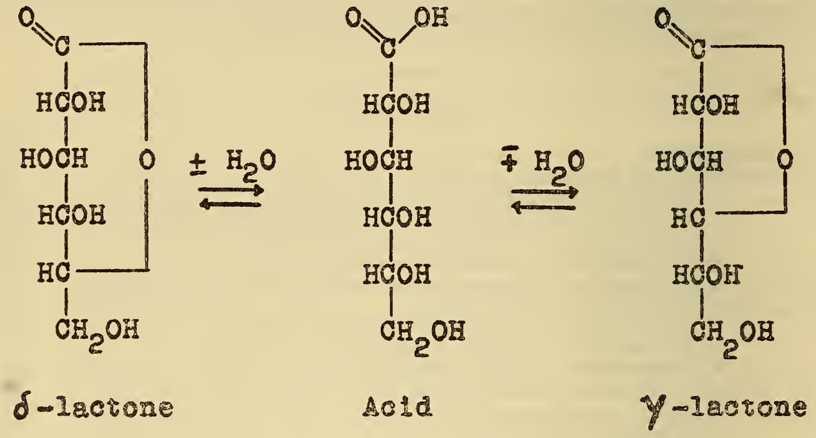
[1] Isbell and Frush, B.S. Jour. Research, vol. 6 (RP328), p. 1145, 1931; and Isbell, Frush, and Bates, B.S. Jour. Research, vol. 8 (RP436), p. 571, 1932.

for investigation. Although many of the sugar acids have been known for a long time and some of the methods for separating the crystalline products are satisfactory, other methods are too difficult to follow and are of little practical importance. Consequently, one desiring to prepare the substances is required to do considerable experimental work in order to select a suitable procedure. The purpose of this publication is to report methods which have been found most satisfactory for preparing the sugar acids and for obtaining the crystalline lactones, with the object of making the lactones and acids more readily available so as to stimulate their industrial application. Since there are large undeveloped sources of dextrose, mannose, galactose, xylose, arabinose, and rhamnose, the acids and lactones derived from these sugars are of particular interest, even though the commercial application of the sugar acids, other than gluconic, must await cheaper methods for the preparation of the parent sugars or their solutions.

In aqueous solution the aldonic acids form γ- and δ- lactones spontaneously and reversibly.[2] This is illustrated in the case of gluconic acid by the following formulas:



Equilibrium solutions of certain acids such as gluconic and lactobionic give relatively strongly acid reactions, showing the presence of considerable free acid; others, as for example, gulonic and α-glucoheptonic, give nearly neutral solutions. The relative amounts of the γ- and δ-lactones in the equilibrium solutions are also subject to wide variation. The pH of the solution, the availability of the acid and the ease of separation of the products are dependent on the position of the equilibrium.

The acids are ordinarily obtained by evaporating at a low temperature freshly prepared aqueous solutions, such as may be derived by treating the calcium salts with oxalic acid. Since the δ-lactones are formed rapidly they are obtained in like manner by dehydrating the acid solution at a low temperature, in the absence of mineral acid and with proper seeding. The γ-lactones are prepared by dehydration of the acids at higher temperatures and in the presence of mineral acids. Because of the equilibrium existing between the free acid and its lactones, a solution will sometimes yield nearly quantitatively

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[2] Levene and Simms, J. Biol. Chem., vol. 65, p. 31, 1925. Charlton, Haworth, and Peat. J. Chem. Soc. p. 89, 1926. Haworth and Nicholson, J. Chem. Soc., p. 1899, 1926.

either the free acid or the lactone, depending upon the substance which happens to crystallize first. For example, an 80 percent aqueous solution of gluconic acid, on seeding with gluconic δ-lactone, gives exclusively crystalline gluconic δ-lactone. On the other hand, we have found that in the absence of δ-lactone seed and in the presence of gluconic acid seed, it gives exclusively crystalline gluconic acid. It is emphasized that concentrated aqueous solutions of the sugar acids are frequently supersaturated with respect to more than one constituent and may yield more than one product. It is frequently difficult to obtain the crystalline acid or lactone for the first time, particularly when the equilibrium is not favorable for its separation, but after crystals are available for seeding no further difficulty is experienced.[3]

In this paper emphasis is placed on the use of several solvents which have recently become commercially available. The use of dioxane in particular is noteworthy. Since this solvent boils at 100° C. and does not combine chemically with either the free acids or their lactones, it may be used to advantage in dehydrating aqueous solutions by distillation. The water and dioxane distill simultaneously, leaving a solution from which either the δ-lactone or acid crystallizes readily. The use of dioxane in separating the sugar acids on a large scale is limited by the cost, but since it can be recovered this solvent may find some application in working up products which are not sufficiently pure to crystallize from water alone.

If the electrolytic process is employed for converting the sugar into the acid, the crude sirups obtained by the hydrolysis of polysaccharides may be used. Thus, many of the sugar acids or their salts can be prepared as cheaply and readily as the corresponding crystalline sugars. In the event that the lactone of the sugar acid crystallizes easily it may be prepared directly from the electrolyzed solution. In such case the calcium or barium is removed by precipitation as oxalate or sulphate; on evaporating the resulting solution the crystalline lactone is obtained. This method is particularly useful for preparing rhamnonic lactone, but it is not suitable for preparing lactones which are difficult to crystallize.

A new method for preparing aldonic acids and their δ-lactones from their sodium salts was recently reported by Brackenbury and Upson.[4] The sodium salt is treated with glacial acetic acid and the δ-lactone or acid is crystallized from the mixture of acetic acid and sodium acetate.

II. GLUCONIC ACID AND ITS DERIVATIVES

In 1884 Kiliani and Kleemann [5] showed that the residue obtained by evaporating an aqueous gluconic acid solution contained a large quantity of lactone. Somewhat later Fischer [6] succeeded in isolating pure crystalline gluconic γ-lactone. In 1914 Nef [7] reported a second lactone now called gluconic δ-lactone, while in 1928 Rehorst [8] prepared crystalline gluconic acid. The equilibrium between gluconic

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[3] The polarimetry section of the Bureau of Standards will furnish seed of any of the products described in this paper upon request as long as the present supply is available.
[4] Brackenbury and Upson, J. Amer. Chem. Soc., vol. 55, p. 2512, 1933.
[5] Kiliani and Kleemann, Ber., vol. 17, p. 17, 1884.
[6] Fischer, Ber., vol. 23, pp. 804 and 2625, 1890.
[7] Nef, Ann., vol. 403, p. 323, 1914. This substance was reported as gluconic β-lactone.
[8] Rehorst, Ber., vol. 61, p. 163, 1928.

acid and its lactones in aqueous solution was investigated first by Nef, who found that the δ-lactone is formed more rapidly than the γ-lactone and that if one evaporated a gluconic acid solution at a low temperature the δ-lactone was obtained in predominating amount, but if the solution were dehydrated at a high temperature in the presence of mineral acids the γ-lactone predominated. Until recently the γ-lactone was obtained by evaporation of aqueous gluconic acid to dryness, extraction of the residue with alcohol, and crystallization from the alcoholic extract. This method is not satisfactory because the alcohol used as a solvent esterifies a part of the sugar acid, forming ethyl gluconate, which interferes with the crystallization of the desired product. Condensation products are also formed which are troublesome to remove and reduce the yield of lactone.

As pointed out by Nef, these amorphous condensation products probably contain glucono-gluconic acid and similar substances, which have not been studied extensively. A better method for the preparation of gluconic γ-lactone was devised by Pasternack and Cragwall.[9] It consists in dehydrating aqueous gluconic acid by distillation in the presence of butyl alcohol. Although the γ-lactone crystallizes readily from the butyl alcoholic solution, considerable difficulty is usually encountered in eliminating the butyl gluconate formed during the dehydration. This objectionable feature is overcome by the new method reported on page 655, which consists in heating gluconic δ-lactone in glacial acetic acid. Inasmuch as the δ-lactone is commercially available at low cost, this simple process makes the γ-lactone available for instruction and research in carbohydrate chemistry.

Gluconic δ-lactone is prepared commercially by the method of Pasternack and Giles,[10] which consists in crystallizing the lactone from concentrated aqueous solutions containing gluconic δ-lactone seed. Small amounts of the lactone can be made without seed by the dioxane method reported in this paper. Gluconic δ-lactone crystallizes in large compact crystals which are easily separated from even thick sirups.

As reported by Rehorst,[11] an aqueous gluconic acid solution on evaporation at 100° C. gives a thick sirup which solidifies under appropriate conditions yielding crystalline gluconic acid. In our experience the acid is obtained only in the absence of δ-lactone seed and by seeding with crystalline gluconic acid. The product obtained by this method is a hard solid which contains all the impurities of the original material. We have found that crystalline gluconic acid can be prepared in nearly pure filterable crystals by evaporating a solution of gluconic acid to about 65 percent, seeding and continuing the evaporation while crystallization of the free acid takes place. It is noted that gluconic acid will crystallize from an aqueous solution in equilibrium with its lactones. Consequently one can prepare the acid easily from gluconic γ- or δ-lactone. In the event that the γ-lactone is used as a raw material it is advantageous to add some acetic acid, as this aids in establishing the equilibrium and also decreases the solubility of the acid. Although crystallization of gluconic acid from aqueous solution is probably the cheapest method, the others are suitable for preparing small quantities and for instruction of students in carbohydrate chemistry.

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[9] Pasternack and Cragwall, U.S. Patent No. 1830618, Nov. 3, 1931.

[10] Pasternack and Giles, U.S. Patent No. 1862511, June 7, 1932.

[11] Rehorst, Ber., vol. 63B, p. 2279, 1930.

1. DIOXANE METHOD FOR PREPARING CRYSTALLINE GLUCONIC ACID

One hundred grams of dry calcium gluconate and 27 g of crystalline oxalic acid are mixed with 100 ml of water in a 300 ml Erlenmeyer flask. The flask is shaken vigorously for 5 minutes; the precipitated calcium oxalate is removed by passing the solution through a Büchner funnel fitted with a filter paper covered with a thin layer of kieselguhr. The filtrate is cooled in an ice bath. After the residue on the filter has been washed successively with 10 ml of water and 10 ml of dioxane, the filtrate is transferred to a 2-liter distillation flask and 200 ml of dioxane added. The resulting solution is concentrated rapidly under reduced pressure at about 30° C. Although crystallization of the acid frequently occurs spontaneously as evaporation takes place, it is desirable to seed the product because otherwise the δ-lactone may crystallize first. Crystals of the acid for seeding can be obtained by evaporating a few ml of the freshly prepared solution containing dioxane on a watch crystal. When the material becomes thick with the crystalline product, distillation is stopped and the crystals are transferred to a filter with about 25 ml of dioxane. The first crop of crystals is usually nearly pure gluconic acid and weighs about 30 g. If the temperature of the distillation is too high the product may be contaminated with δ-lactone. By evaporating the mother liquor additional crystals are obtained but these usually contain more or less δ-lactone.

2. CRYSTALLIZATION OF GLUCONIC ACID FROM AQUEOUS ALCOHOL

A dry mixture containing equimolecular proportions of finely powdered crystalline calcium gluconate (50 g) and oxalic acid (13.5 g) is added in small portions with vigorous mechanical stirring to a small quantity of ice water (25 ml). When stirred several minutes all the calcium gluconate is converted into gluconic acid and calcium oxalate. The mixture is diluted with 50 ml of isopropyl alcohol and poured on a 4-inch Büchner funnel fitted with a filter paper covered with kieselguhr. The residue is washed with a few ml of water and the filtrate is diluted with isopropyl alcohol until the volume is about 250 ml. Crystallization of gluconic acid may be induced by seeding or scratching the walls of the flask. After standing 18 hours in the refrigerator the resulting crystalline gluconic acid is separated, washed with isopropyl alcohol, and dried. About 7 g is obtained. Absolute ethyl alcohol may be used in place of the isopropyl alcohol, in which case the yield is slightly lower. This is the simplest method for preparing small quantities of gluconic acid.

3. CRYSTALLIZATION OF GLUCONIC ACID FROM WATER

About 500 ml of a 40-percent solution of aqueous gluconic acid, which may be prepared from calcium gluconate and oxalic acid, is placed in a 2-liter Claissen distillation flask containing an inlet tube for introducing additional liquid. The solution is evaporated rapidly under reduced pressure at about 40° C. When the material in the flask becomes sirupy (about 65 percent dry substance), gluconic acid seed is added and evaporation is continued while simultaneously freshly prepared gluconic acid solution is introduced dropwise into the flask at such a rate that the crop of crystals continues to increase. The freshly prepared gluconic acid solution is made from time to time during the distillation from 100 g of calcium gluconate, 27 g of oxalic acid and 200 ml of water, or from crude crystalline gluconic acid which it is desired to purify. Addition of gluconic acid and simultaneous evaporation are continued until the desired quantity of crystals is present in the flask. The material is transferred to a filter and the crystals are separated from the mother liquor. In small-scale preparations, a small amount of acetic acid or ethyl alcohol may be used in removing the crystals from the flask. The product is washed with ethyl alcohol followed by dry ether or dioxane. Nearly quantitative yields may be obtained by reworking the mother liquors. The product may be purified by repeating the process, or if desired it may be recrystallized by dissolving it in cold water and adding glacial acetic acid according to the method of Brackenbury and Upson (footnote 4).

Crystalline gluconic acid may also be made from gluconic δ- or γ-lactone. One part of δ-lactone is dissolved in one part of boiling water; after filtration the solution is cooled in ice water and seeded with crystalline gluconic acid. After standing several hours at 0° C. a crop of crystalline acid may be separated. The mother liquors can be concentrated and additional acid obtained. Use of an organic solvent such as acetic acid or dioxane is advantageous in washing the crystals and working up the mother liquors. Crystalline gluconic acid melts at 120°–131° C., the temperature varying with the rate of heating. In 5 percent aqueous solution $[\alpha]^{20}_{D} = -6.9°$, 2 minutes after solution, $+7.3°$ after 24 hours. Rehorst (footnote 11) reports the melting point as 130°–132° C. (not sharp, sintering at 112°) and the specific rotation as $-6.7°$, initially, $+7°$ after 24 hours.

4. PREPARATION OF GLUCONIC δ-LACTONE

A dry mixture of 150 g of calcium gluconate and 40.6 g of crystalline oxalic acid is added in portions, with shaking, to 150 ml of warm water in a 500 ml Erlenmeyer flask. After the calcium gluconate is decomposed, 150 ml of dioxane is added, and the resulting solution is passed through a Büchner funnel fitted with a filter paper covered with kieselguhr. The filtrate is cooled to room temperature and evaporated rapidly at about 35° C. in vacuo by using a good water pump. When the volume of the solution is reduced to about 100 ml, 150 ml of dioxane and some crystalline gluconic δ-lactone are added. Distillation at 30° to 35° C. is continued until the mixture becomes thick with the crystalline product. The crystals are separated by filtration, washed with dioxane, and dried. The mother liquor is returned to the distillation flask and concentrated until additional crystals are obtained. In a typical experiment the first crop weighed 69 g and gave $[\alpha]^{20}_{D} = +64.9°$ initially, $+10°$ after 24 hours; the second crop weighed 32 g and gave $[\alpha]^{20}_{D} = +63.5°$ initially, $+17.0°$ after 24 hours; the third crop weighed 11.5 g and gave $[\alpha]^{20}_{D} = +58.2°$ initially, $+13.5°$ after 24 hours. If the solution is heated too long or at too high a temperature, the last fractions will contain gluconic γ-lactone, while if the solution of gluconic acid is evaporated too rapidly at a low temperature gluconic acid will crystallize rather than the lactone. Any fraction which contains crystalline gluconic acid and the δ-lactone may be converted into δ-lactone by digesting a short time with dioxane. The presence of the acid in the lactone can be detected by its sour taste or by its appearance. It crystallizes in small irregularly shaped clusters of slender pointed crystals, while the δ-lactone separates in large chunky prisms. The lactone melts at 150° to 152° C. In 5 percent aqueous solution, $[\alpha]^{20}_{D} = +66.2°$ initially, $+8.8°$ after 24 hours. These rotations are in approximate agreement with those recently reported by Brackenbury and Upson (footnote 4).

5. PREPARATION OF GLUCONIC γ-LACTONE FROM GLUCONIC δ-LACTONE

About 100 ml of glacial acetic acid and 50 g of gluconic δ-lactone are placed in a round-bottomed pyrex flask fitted with a reflux condenser. The mixture in the flask is heated to boiling and refluxed about 1 hour. On cooling the resulting solution and seeding with γ-lactone, crystalline gluconic γ-lactone separates slowly. After standing overnight in the refrigerator the product is collected on a filter, washed successively with glacial acetic acid, alcohol, and ether. About 23 g of crude γ-lactone of about 90 percent purity is obtained. If desired, additional δ-lactone may be added to the acetic acid mother liquor to replace that converted to γ-lactone and the process repeated. The crude γ-lactone can be recrystallized by dissolving it in 3 parts of hot acetic acid or about 12 parts of hot dioxane. The lactone separates on cooling the solution. The mother liquors may be used for recrystallizing additional material.

The γ-lactone melts at 133° to 135° C. In 5 percent aqueous solution, $[\alpha]^{20}_{D} = +68.0°$ initially, $+67.2°$ after 24 hours. Fischer (footnote 6) previously reported the melting point as 130° to 135° C., and $[\alpha]^{20}_{D} = +68.2°$.

Gluconic γ-lactone seed may be made in the following manner: An aqueous gluconic acid solution is evaporated in vacuo to a thick sirup. After being heated for several hours in order to convert the acid to the lactone the residue is extracted with hot normal butyl alcohol. Upon cooling the alcoholic extract, crystallization occurs spontaneously, particularly when the solution is allowed to evaporate in an open dish.

III. XYLONIC ACID AND ITS DERIVATIVES

Xylonic acid and strontium xylonate were prepared first by Allen and Tollens [12] by the oxidation of xylose with bromine water. Subsequently characteristic cadmium-cadmium bromide [13] and cadmium chloride salts have been reported. These salts are useful for identification purposes but they are not suitable for preparing xylonic acid and its derivatives. The free acid can be prepared from calcium xylonate [14] or from lead xylonate.[15] We have previously reported (RP328) a simple method for preparing amorphous calcium xylonate by the electrolytic oxidation of xylose, and separation of the product as a basic calcium xylonate. Although crystalline xylonic acid is at present unknown, the γ-lactone crystallizes fairly readily. Heretofore this lactone has been prepared only in small quantities, but with

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[12] Allen and Tollens, Ann., vol. 260, p. 306, 1890.
[13] Bertrand, Bull. soc. chim. (3), vol. 5, p. 556, 1891.
[14] Nef, Ann., vol. 403, p. 252, 1914.
[15] Hasenfratz, Compt. rend., vol. 196, p. 350, 1933.

the development of cheaper methods for the production of xylose[16] and calcium xylonate it is now possible to prepare it in any desired quantity. Previous investigators usually crystallized the lactone from acetone.[17] This solvent is not satisfactory for preparing the crude lactone because a large part of the material is left in the form of an amorphous residue. We have found that this difficulty may be overcome by crystallization from a concentrated aqueous solution without the use of solvents other than water. Since the lactone is very soluble in water it is difficult to obtain high yields in small-scale preparations, but in large quantities the heavy massecuite may be treated more economically. In small-scale laboratory preparations crystallization from dioxane is advantageous. Xylonic γ-lactone is miscible with dioxane at the boiling point (760 mm), while at 20° C. 100 g of dioxane dissolves about 7 g of lactone.

1. PREPARATION OF CALCIUM XYLONATE FROM XYLOSE

A solution containing 300 g of xylose and 30 g of potassium bromide in 1 liter of water is placed with 100 g of calcium carbonate in a 2-liter three-necked flask cooled by running water, and equipped with a mechanical stirrer and two graphite electrodes about 2.2 cm in diameter and immersed to a depth of 6.7 cm. A current of 1 ampere at 15 volts is passed through the solution. Under these conditions the current density is about 2 amperes per dm ^{2} of anode surface. After 110 ampere-hours about 98 percent of the sugar is oxidized with an anode efficiency of about 95 percent. When a Fehling's test on the electrolyte is nearly negative, electrolysis is stopped. The electrolyzed solution is filtered and the xylonic acid is precipitated as basic calcium xylonate by the addition of 200 g of hydrated lime in 2 liters of water (150 g theoretically required). In order to obtain a product which filters readily it is necessary to precipitate the basic salt slowly. This is done by allowing the hot milk of lime and hot calcium xylonate solution to flow slowly and simultaneously, with stirring, into a precipitating jar. After the solutions are mixed, the product is heated to about 80° C. in order to complete the precipitation.

The basic salt is collected on a large Büchner funnel and washed with lime water until free from bromide. It is then suspended in water and carbon dioxide introduced with agitation until the solution gives a neutral reaction to phenolphthalein. An excess of CO_2 should be avoided as it results in a residue which is difficult to separate. The insoluble calcium carbonate is removed by filtration and the filtrate concentrated to 650 ml. The resulting solution usually contains about 225 g of calcium xylonate which corresponds to a 60 percent yield. The mother liquor from the basic salt may be used in a subsequent oxidation in order to reclaim the calcium xylonate and potassium bromide contained therein. Or if desired a second crop of basic salt may be obtained by concentrating the solution and adding more lime, followed by heating.

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[16] Schreiber, Geib, Wingfield, and Acree, Ind. Eng. Chem., vol. 22, p. 497, 1930.
[17] Clowes and Tollens, Ann., vol. 310, p. 175, 1899.

2. PREPARATION OF XYLONIC γ-LACTONE

A hot solution containing 215 g of calcium xylonate in 650 ml of water is mixed with 73.25 g of crystalline oxalic acid. The precipitated calcium oxalate is separated and after the addition of a few ml of concentrated hydrochloric acid the solution is evaporated in vacuo. When the sirup becomes thick it is cooled to 30° C. and seeded with crystalline xylonic γ-lactone, after which boiling is continued. The lactone crystallizes slowly from the solution and the material finally becomes solid. It is necessary to conduct the evaporation slowly and to heat the residue in vacuo for several hours in order to convert the acid into the lactone. The residue is extracted with about 150 ml of hot dioxane. Most of the material goes into solution, leaving a small quantity of residue (8 g). The composition of this residue and the conditions under which it is formed are not known. The amount varies in an unknown manner, possibly depending on the method of dehydration. In order to obtain the crystalline lactone without seed it is usually necessary to extract the thick aqueous sirup containing the crude lactone with acetone. The acetone extract on evaporation in an open dish gives crystalline xylonic lactone.

The dioxane solution is concentrated in a distillation flask under reduced pressure to about 100 ml. The resulting sirup is seeded with xylonic γ-lactone crystals and benzene is added until the solution becomes slightly turbid. On standing, about 110 g of lactone crystallizes from the solution. After separation of the crystals the mother liquor is concentrated and additional lactone obtained. The total yield of crude lactone is about 160 g or approximately 90 percent.

The crude xylonic lactone may be recrystallized by dissolving it in boiling dioxane, using 2 ml of dioxane for each gram of lactone. A small quantity of amorphous residue remains which is separated by filtration. On cooling, nearly pure xylonic γ-lactone crystallizes from the resulting solution. When in fairly pure state, xylonic lactone may be recrystallized easily from numerous solvents, such as acetone, isopropyl, butyl, isoamyl alcohol, or glacial acetic acid. The lactone melts at 98° to 101° C. In 5 percent aqueous solution, $[\alpha]^{20}_{D} = +91.8°$ 2 minutes after solution, $+86.7°$ after 24 hours. The melting point agrees with that of other investigators but the initial rotation is slightly higher than the values of Nef, $+89.56°$ (footnote 14) or Hasenfratz, $+85.5°$ (footnote 15).

3. PREPARATION OF BASIC CALCIUM SALTS FOR ANALYSIS

A solution containing 20 g of calcium xylonate in 600 ml of ice water is mixed with 400 ml of cold milk of lime containing 7 g of calcium oxide. The solution is filtered quickly and the clear filtrate evaporated in vacuo to about 100 ml. The mixture is then warmed on the steam bath in order to complete the precipitation of the basic salt, which is subsequently collected on a filter, washed with lime water, and dried at 60° C. in vacuo. All operations are conducted rapidly in order to avoid contamination with carbon dioxide. The calcium content, reported in table 2, was determined by precipitation as calcium oxalate. The CaO content was determined by dissolving in 0.5 *N* HCl, followed by the addition of an excess of standard alkali and back titration with 0.1 *N* HCl by using phenolphthalein as an indicator.

IV. MANNONIC ACID AND ITS DERIVATIVES

Mannose occurs in many vegetable products in the form of polysaccharides which give the sugar by hydrolysis. A convenient source is shavings from ivory nuts, which can be purchased from button factories. The liquors obtained from the hydrolysis of ivory nuts can be used directly for preparing calcium mannonate.[18]

The methods for separating calcium mannonate from the byproducts of the older bromine or chlorine oxidation of mannose required considerable work, and the use of expensive chemicals. Our electrolytic process of oxidation, (RP328), eliminates these expensive steps because the desired product crystallizes from the electrolyte without prior purification. The salt separates in well defined crystals provided that the solution is not too concentrated, is seeded and kept in motion during crystallization; if these precautions are not observed a gelatinous product results.

Bernhauer and Irgang,[19] in applying the electrolytic method to crude mannose derived from the hydrolysis of ivory nuts observed a lower efficiency than we did with pure mannose. It has been our experience that crude glucose, mannose, xylose, and arabinose solutions frequently give low and variable current efficiencies, while high current efficiencies are nearly always obtained with fairly pure materials. The lower efficiencies are probably due to reactions brought about by the impurities. This subject requires further investigation.

As shown by Nef,[20] mannonic δ-lactone is obtained by evaporating a freshly prepared solution of mannonic acid. The equilibrium solution of mannonic acid contains a large quantity of γ-lactone and very little δ-lactone and free acid. Hence it is necessary to work rapidly and to take precaution to avoid the formation of γ-lactone. Mannonic δ-lactone crystallizes very readily but it will not crystallize from an equilibrium mannonic acid solution, in which respect it differs from gluconic δ-lactone. Crystalline mannonic acid is at present unknown.

1. CALCIUM MANNONATE

A 300 g sample of 20-mesh ivory nut meal is treated according to the method of Nelson and Cretcher (footnote 18) with 3 liters of boiling 1 percent sodium hydroxide and the mixture allowed to stand 30 minutes, with occasional stirring. It is then filtered through muslin, washed thoroughly, and dried in air. The purified meal (207 g) is mixed with an equal weight of 75 percent sulphuric acid and allowed to stand 22 hours at room temperature. Then the thick mass is mixed with 2.3 liters of water and heated to boiling. After 6 hours at about 100° C. the mixture is filtered and the hot filtrate neutralized with calcium carbonate.

The solution containing calcium sulphate in suspension is decolorized by the addition of about 10 g of activated carbon followed by filtration. If sufficient decolorizing carbon is not used the solution foams during distillation and must be treated again with carbon. It is then concentrated by distillation under reduced pressure to about 1.5 liters and the amount of sugar in solution is determined by

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[18] Nelson and Cretcher, J.Amer.Chem.Soc., vol. 52, p. 404, 1930.

[19] Bernhauer and Irgang, Biochem.Z., vol. 249, p. 216, 1932.

[20] Nef, Ann., vol. 403, p. 306, 1914; also Hedenberg, J.Am.Chem.Soc., vol. 37, p. 345, 1915. This substance was originally called a "β"-lactone. See Goodyear and Haworth, J.Chem.Soc., p. 3136, 3144, 1927; also Haworth and Nicholson, J.Chem.Soc., p. 1899, 1926.

Fehling's method. The solution usually contains about 150 g of sugar calculated as mannose. After the addition of about 45 g of calcium carbonate and 20 g of calcium bromide the solution is electrolyzed, using an anode current density of 2 amperes/dm.^{2} When nearly all of the sugar has been oxidized electrolysis is stopped. The oxidation of crude mannose solution usually requires about 1.5 times the theoretical amount of current. Theoretically 1 mole of sugar requires 53.6 ampere hours. The electrolyzed solution is filtered, concentrated to about 400 ml, and refiltered hot. The resulting solution, seeded with calcium mannonate, and stirred during crystallization, will yield about 100 g. The crystalline salt, $Ca(C_6H_{11}O_7)_2.2H_2O$ is separated by filtration. The calcium mannonate which remains in solution is precipitated as the basic salt by adding an excess of hydrated lime (60 g) to the mother liquor, essentially as described on page 656. After carbonation of the resulting basic salt additional calcium mannonate is obtained. The total yield is usually about 65 percent of that theoretically possible from the mannose in the solution.

2. MANNONIC δ-LACTONE

A dry mixture of 250 g of calcium mannonate and 67.6 g of crystalline oxalic acid is added in portions, with mechanical mixing, to 250 ml of water; the resulting solution is filtered and quickly cooled in an ice bath. An equal volume of dioxane is added and the solution is evaporated rapidly at 35° C. in vacuo. Crystalline mannonic δ-lactone separates as the solution is concentrated. It is advisable to separate the product in several fractions, as the first crystals are pure δ-lactone, while the later fractions are largely γ-lactone. The crystals are separated and washed with dioxane. About 115 g of pure δ-lactone (m.p. 158° to 160° C.) and 55 g of γ-lactone are usually obtained from the amounts stated. The δ-lactone can be obtained without the use of dioxane but the yield is less. The lactone can be recrystallized by dissolving it in the smallest possible amount of water at room temperature, filtering the resulting solution, adding an equal volume of dioxane, and evaporating rapidly in vacuo until a crop of crystals is obtained. The δ-lactone is difficult to purify as it is easily converted into γ-lactone. It is necessary to work rapidly and to avoid high temperatures. In 5 percent aqueous solution $[\alpha]_D^{20} = 114.8°$, 2 minutes after solution, $+30.3°$ after 24 hours. These values agree approximately with the results obtained by Goodyear and Haworth (footnote 20).

3. MANNONIC γ-LACTONE

The calcium is removed from 100 g of calcium mannonate in 100 ml of hot water by treatment with 27 g of oxalic acid. After filtration the aqueous mannonic acid is evaporated in vacuo in the presence of a few drops of hydrochloric acid. When the solution becomes concentrated, crystallization occurs. Evaporation is continued, however, until the residue is nearly solid and the hydrogen chloride is largely evaporated. The resulting crude lactone is recrystallized from water. By carefully working up the mother liquors a nearly quantitative yield is obtained. The product melts at 151° to 152° C. and gives $[\alpha]_D^{20} = +51.5°$, 2 minutes after solution. Fischer and Hirschberger[21] originally reported the melting point as 149°–153° and $[\alpha]_D^{20} = +53.81°$, while Nef (footnote 20) found $[\alpha]_D^{20} = +51.8°$.

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[21] Fischer and Hirschberger, Ber., vol. 22, p. 3218, 1889.

V. GALACTONIC ACID AND ITS DERIVATIVES

As early as 1862 Barth and Hlasiwetz[22] prepared galactonic acid and its γ-lactone. The constitution of these substances was uncertain until studied by Schnelle and Tollens[23] and by Nef.[24]

Galactonic acid differs from the other sugar acids in that it is easily crystallized from water. It then contains 2 molecules of water for each molecule of acid,[25] but if crystallized from hot aqueous alcohol the product contains only 1 molecule of water for each 2 molecules of acid. Nef designated the latter product ortho-bis-galaconic acid. The more highly hydrated product gives up water easily and is difficult to prepare in pure state. The ortho-bis-galactonic acid is more stable but even this on treatment with dehydrating agents yields the anhydrous form. The γ-lactone also forms a hydrate which on careful drying becomes anhydrous.

Galactonic δ-lactone has not been prepared in the crystalline state although a solution of it may be obtained by the oxidation of galactose with bromine water in the presence of a buffer.[26] The δ-lactone exhibits a dextro rotation and is hydrolyzed fairly rapidly. In this respect it differs widely from the γ-lactone which is hydrolyzed very slowly and only to a small degree.

The most convenient source of galactonic acid is calcium galactonate prepared by the electrolytic oxidation of galactose in the presence of a bromide and calcium carbonate (RP328). Calcium galactonate is more difficultly soluble than most salts of the sugar acids and crystallizes very easily. Hence it may be prepared by the electrolytic method from the hydrolytic products derived from lactose,[27] or better, from galactan-bearing plants such as agar-agar or the Western larch.[28]

1. GALACTONIC ACID

Equivalent quantities of finely powdered oxalic acid and calcium galactonate are added, with stirring, to 1 g of hot water for each g of calcium galactonate. The mixture is shaken about 3 minutes and the resulting aqueous galactonic acid is filtered and cooled. Crystalline galactonic acid hydrate separates at once. A second crop may be obtained by the addition of about four volumes of absolute ethyl alcohol. One experiment, using 100 g of calcium galactonate, yielded a first crop of 16 g (m.p. 120° to 124° C.), and a second crop (from alcohol) of 21 g (m.p. 125° to 127° C.). The dihydrate first described by Kiliani gives up water easily even by air-drying at room temperature, thereby forming the hydrate $(C_6H_{12}O_7)_2 \cdot H_2O$, termed ortho-bis-galactonic acid by Nef (footnote 24). The crude product is usually a mixture of the two forms melting at any point between 120° and 140° C. The dihydrate melts at 118° to 120° C., and the ortho-bis acid at 140° to 142° C. The latter may be conveniently prepared by crystallization from aqueous alcohol as described by Nef.

The anhydrous acid has recently been obtained by Brackenbery and Upson (footnote 4) by treating sodium galactonate with acetic acid.

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[22] Barth and Hlasiwetz, Ann., vol. 122, p. 96, 1862.

[23] Schnelle and Tollens, Ann., vol. 271, p. 81, 1892.

[24] Nef, Ann., vol. 403, p. 273, 1914.

[25] Kiliani, Ber., vol. 55, p. 95, 1922.

[26] Isbell and Hudson, B.S.Jour. Research, vol. 8, (RP418), p. 332, 1932.

[27] Kiliani, Ber., vol. 65B, p. 1269, 1932.

[28] A process for preparing a galactose solution from Western larch is described by Acree in U.S. Patent No. 1816136, dated July 28, 1931.

We have found that the anhydrous form may be prepared by digesting the crude hydrate for 5 or 10 minutes with boiling dioxane. The product so obtained melts at 148° C. $[\alpha]_D^{20} = -13.6°$ (2 minutes).

2. GALACTONIC γ-LACTONE

An aqueous solution of galactonic acid is prepared from equivalent quantities of calcium galactonate (100 g) and oxalic acid (24.2 g) by using about 2 parts of hot water for each part of calcium galactonate. After filtration a few drops of concentrated hydrochloric acid are added and the solution concentrated to about 100 ml, after which an equal volume of secondary amyl alcohol is added and the mixture concentrated to a thick sirup. A second addition of secondary amyl alcohol and further boiling in vacuo results in a sirup which on seeding or scratching gives crystalline anhydrous galactonic γ-lactone. After being cooled and allowed to stand for about 1 hour the crystals are separated by filtration and washed with a 50 percent mixture of absolute ethyl and secondary amyl alcohols. A second crop of crude anhydrous γ-lactone crystals is obtained by evaporating the mother liquors. The total yield is nearly quantitative.

The crude anhydrous galactonic γ-lactone may be purified by recrystallization from hot absolute ethyl alcohol.

Galactonic γ-lactone melts at 110° to 112° C. and gives $[\alpha]_D^{20} = -77.4°$.

These values agree with the results of Nef (footnote 24). The anhydrous lactone is easily converted into the hydrated form, which melts at 66° C. A 100 g sample of galactonic γ-lactone is dissolved in 35 ml of boiling water. After the addition of 150 ml of isopropyl alcohol, cooling, and seeding, crystallization occurs. About 85 g of nearly pure galactonic γ-lactone hydrate (m.p. 66° C.) is obtained. Additional product may be reclaimed from the mother liquors. The lactone hydrate may be prepared also by crystallization from aqueous acetic acid as described by Levene and Meyer.[29]

VI. ARABONIC ACID AND ITS DERIVATIVES

Arabinose occurs in many gums and other plant products, frequently in combination with galactose and other sugars. The best sources are mesquite and cherry gums. The gum is hydrolyzed with acid, the excess acid neutralized with calcium carbonate, the solution concentrated, and the arabinose extracted with methyl alcohol. After evaporating the alcohol the resulting crude arabinose may be used directly for preparing calcium arabonate by means of the electrolytic method (RP328). The acid is easily separated from the impurities as the crystalline calcium salt [30] or the basic calcium salt reported in table 2.

Crystalline arabonic acid was prepared first by Hauers and Tollens [31] and again by Böddener and Tollens,[32] but it was not correctly identified until recently.[33] It crystallizes well, resembling galactonic acid in many respects. The crystalline acid can be separated easily from

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[29] Levene and Meyer, J.Biol.Chem., vol. 46, p. 307, 1921.
[30] Kiliani, Ber., vol. 19, p. 3029, 1886.
[31] Hauers and Tollens, Ber., vol. 36, p. 3321, 1903.
[32] Böddener and Tollens, Ber., vol. 43, p. 1645, 1910.
[33] Rehorst, Ber., vol. 63B, p. 2279, 1930.

a concentrated aqueous solution, particularly by using dioxane as outlined below. Arabonic δ-lactone is unknown at present, but as shown by Isbell and Hudson (footnote [26]), it is dextrorotatory and is hydrolyzed comparatively rapidly. Arabonic γ-lactone [34] [35] was prepared first by evaporation of aqueous arabonic acid. This method requires considerable time and is not as satisfactory as dehydration of the aqueous acid by distillation with butyl alcohol in a manner analogous to the preparation of gluconic γ-lactone (footnote 9).

1. ARABONIC ACID

Equivalent quantities of calcium arabonate (75 g of $Ca(C_5H_9O_6)_2 . 5H_2O$) and oxalic acid (20.6 g), are mixed dry and added in portions to a small volume of water (100 ml). The resulting mixture is shaken continuously for 5 minutes and passed through a filter paper covered with kieselguhr into a flask kept in ice water. The residue is washed, first with a little water and then with dioxane. After addition of 125 ml of dioxane the solution is evaporated in vacuo at about 35° C. to approximately 50 ml, when 50 ml of dioxane is added and evaporation continued. Crystalline arabonic acid forms as the solution becomes quite concentrated. When the material in the flask has largely crystallized, evaporation is interrupted and 100 ml of dioxane is added. After standing for several hours the crystalline arabonic acid is separated, washed with dioxane, and air-dried at room temperature. The mother liquors on concentration at a low temperature yield a second crop of arabonic acid. The total yield is about 40 g or nearly 75 percent. The residues may be reclaimed as γ-lactone. The crude arabonic acid is recrystallized by dissolving it in an equal weight of water and repeating the above process. The product melts at 111° to 116° C. and gives $[\alpha]^{20}_{D} = -9.8°$, 2 minutes after solution.

2. ARABONIC γ-LACTONE

Equivalent amounts of oxalic acid (34 g) and calcium arabonate (124 g of $Ca(C_5H_9O_6)_2 . 5H_2O$) are mixed in 500 ml of hot water. The mixture is filtered and after the addition of a few drops of hydrochloric acid the solution is concentrated in vacuo to about 100 ml, when 150 ml of normal butanol is added. The solution is boiled down again to about 100 ml and 100 ml of butanol added. After a third concentration the solution is removed from the flask with the aid of 50 ml of butanol, and seeded. Crystallization begins at once, and a large crop of crystals forms in the course of half an hour. The crystalline product (46 g) is collected on a filter, washed with butanol, and dried at 30° C. in vacuo.

The mother liquors give additional lactone by concentrating in a current of air. The crude product can be purified by recrystallization from hot dioxane.

The lactone melts at 95° to 98° C. and gives $[\alpha]^{20}_{D} = -71.6°$, 2 minutes after solution.

Fischer and Piloty reported $[\alpha]^{20}_{D} = -73.9°$ and Böddener and Tollens $-70.8°$.

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[34] Bauer, J.prakt.Chem., vol. 30, p. 380, 1884.

[35] Fischer and Piloty, Ber., vol. 24, p. 4218, 1891.

VII. RHAMNONIC ACID AND ITS DERIVATIVES

Rhamnonic acid forms two crystalline lactones[36] [37] [38] which crystallize readily and are easily prepared from an aqueous solution of rhamnonic acid. The crystalline acid is not known but aqueous solutions may be prepared from calcium or barium rhamnonate. The latter substances are obtained readily by the electrolytic oxidation of rhamnose in the presence of a bromide and calcium or barium carbonate (RP328). Calcium rhamnonate crystallizes in slender needles, which occlude the mother liquor, forming gelatinous masses from which it is difficult to separate the pure salt. A solution of calcium rhamnonate suitable for preparing the lactones can be easily obtained by means of the basic calcium salt reported in table 2. The method of preparation is analogous to that described for the preparation of calcium xylonate on page 656.

1. RHAMNONIC δ-LACTONE

A solution of 75 g of anhydrous calcium rhamnonate in 300 ml of water is treated at 60° C. with 23.7 g crystalline oxalic acid. After filtration the solution is evaporated at about 35° C. in vacuo and brought to crystallization. The product (45 g) is collected on a filter, washed with ethyl alcohol, and dried in vacuo.

A small second crop of δ-lactone is obtained by concentrating the mother liquor in vacuo. The residue, on evaporation in air, gives rhamnonic γ-lactone. Virtually all the product crystallizes. Rhamnonic δ-lactone melts at 172° to 182° C. and gives $[\alpha]_D^{20} = -100.4°$, 2 minutes after solution. Jackson and Hudson found the same melting point and $[\alpha]_D^{20} = -98.4°$, 7 minutes after solution.

2. RHAMNONIC γ-LACTONE

A solution containing 45.5 g of rhamnose, 15 g of barium bromide ($BaBr_2.2H_2O$), and 1 liter of water is placed with 25 g of barium carbonate in an electrolytic cell, and a current of 0.5 ampere is passed through the solution. After 15.5 ampere-hours about 96 percent of the sugar is oxidized with an anode efficiency of about 80 percent. When a Fehling's test is nearly negative electrolysis is stopped and the electrolyzed solution filtered.

The barium is removed by the addition of the required amount of sulphuric acid. The bromide ion is precipitated by the addition of the equivalent amount of silver carbonate after which a few drops of concentrated hydrochloric acid are added to make the solution strongly acid. The barium sulphate and silver bromide are separated by filtration and the aqueous rhamnonic acid is evaporated in vacuo to a thick sirup from which crystalline rhamnonic γ-lactone separates. The crystals are collected on a filter, washed with ethyl alcohol, and dried. The mother liquor gives additional material on evaporation, and the total yield is about 80 percent of the theoretical. The lactone is easily recrystallized from ethyl alcohol. It melts at 149° to 151° C. and gives $[\alpha]_D^{20} = -39.2°$, 2 minutes after solution. These values agree with those reported by other investigators.

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[36] Will and Peters, Ber., vol. 21, p. 1813, 1888.
[37] Jackson and Hudson, J.Am.Chem.Soc., vol. 52, p. 1270, 1930.
[38] Votoček and Beneš, Bull. Soc. chim., vol. 43, p. 1330, 1928.

VIII. SUMMARY

Directions are given for the preparation of the more common aldonic acids and their lactones. The melting points and specific rotations of the acids and lactones are given in table 1.

The composition of the basic calcium salts reported in the paper are given in table 2. The samples were prepared by the method outlined in detail for the basic salt of xylonic acid (p. 657). Small variations in composition are inherent in the method of preparation, and since the products cannot be recrystallized because of hydrolysis, the analyses are on relatively crude salts.

TABLE 1.—*Specific rotations and melting points*

Substance	Formula	Melting point	Specific rotation[1]			
			2 minutes	5 minutes	10 minutes	24 hours
		°C.				
Gluconic acid	$C_6H_{12}O_7$	120–131	−6.9	−5.2	−4.2	+7.3
Gluconic γ-lactone	$C_6H_{10}O_6$	133–135	+68.0	+68.0	+68.0	+62.7
Gluconic δ-lactone	$C_6H_{10}O_6$	150–152	+66.2	+65.6	+63.9	+8.8
Galactonic acid	$C_6H_{12}O_7$	148	−13.6	−13.2	−13.0	−17.0
Galactonic γ-lactone hydrate	$C_6H_{10}O_6.H_2O$	65–66	−71.0	−71.0	−71.0	−70.6
Galactonic γ-lactone anhydrous	$C_6H_{10}O_6$	110–112	−77.4	−77.4	−77.4	−76.4
Mannonic γ-lactone	$C_6H_{10}O_6$	151–152	+51.5	+51.5	+51.5	+51.5
Mannonic δ-lactone	$C_6H_{10}O_6$	158–160	+114.8	+114.5	+114.2	+30.3
Rhamnonic γ-lactone	$C_6H_{10}O_5$	149–151	−39.2	−39.2	−39.2	−39.1
Rhamnonic δ-lactone	$C_6H_{10}O_5$	172–182	−100.4	−100.1	−99.6	−35.1
Xylonic γ-lactone	$C_5H_8O_5$	98–101	+91.8	+91.8	+91.8	+86.7
Arabonic acid	$C_5H_{10}O_6$	111–116	−9.8	−9.5	−9.3	−11.8
Arabonic γ-lactone	$C_5H_8O_5$	95–98	−71.6	−71.6	−71.6	−71.6

[1] Aqueous solutions containing 1.25 g of substance dissolved in 25 ml read at 20° C. in a 2 dm tube on a Bates saccharimeter. The factor 0.3462 was used to convert the ° S. to angular degrees.

TABLE 2.—*Basic calcium salts of aldonic acids*

Basic calcium salt of	Formula	Analysis			
		Found		Calculated	
		Total Ca	CaO	Total Ca	CaO
d-Gluconic[1]	$Ca(C_6H_{11}O_7)_2.2CaO$	22.10	20.30	22.12	21.65
d-Mannonic	$Ca(C_6H_{11}O_7)_2.2CaO$	23.51	22.45	22.12	21.65
d-Galactonic	$Ca(C_6H_{11}O_7)_2.2CaO$	24.08	23.02	22.12	21.65
d-Xylonic	$Ca(C_5H_9O_6)_2.2CaO$	25.05	22.74	24.90	23.24
l-Arabonic	$Ca(C_5H_9O_6)_2.2CaO$	24.85	22.89	24.90	23.24
l-Rhamnonic	$CaC_6H_{11}O_6)_2.2CaO$	24.32	22.05	23.50	21.93
Maltobionic	$Ca(C_{12}H_{21}O_{12})_2.4CaO$	20.52	22.93	20.45	22.90
Lactobionic[2]	$Ca(C_{12}H_{21}O_{12})_2.4CaO$	20.65	23.00	20.45	22.90

[1] Fischer, Ber., vol. 23, p. 2615, 1890.

[2] Hudson and Isbell, B.S.Jour. Research, vol. 3 (RP82), p. 60, 1929.

WASHINGTON, August 30, 1933.