Thermal Degradation of 1,6-Anhydro-β-D-glucopyranose

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Received May 24, 1971

Thermal degradation of 1,6-anhydro-β-D-glucopyranose-*1*-^{14}C, -*2*-^{14}C, and -*6*-^{14}C gave carbon dioxide, carbon monoxide, and a variety of carbonyl compounds that were isolated and traced to the labeled positions. Variations of the yields and radiochemical patterns of the products on addition of sodium hydroxide or zinc chloride indicated the nature of the complex consecutive and concurrent reactions involved.

A variety of mechanisms have been suggested for the thermal degradation of cellulose and related model compounds to low-molecular-weight products.[2-5] These mechanisms are generally based on the isolation and identification of the products assuming the formation of either free-radical[3] or carbonium ion intermediates[4] without sufficient evidence. Furthermore, they entail a single pathway for the formation of each product, which is very unlikely under the pyrolytic conditions, when the molecule is physically torn into pieces.[2] To gain further insight into the complex nature of the pyrolytic transformations, a systematic approach has been adapted in this laboratory involving combinations of thermal analysis methods (differential thermal analysis, thermogravimetric analysis, and derivatography) and parallel chemical studies.[6-10] These investigations have shown that heating of carbohydrates results in transition of the crystalline structure, anomerization of free sugar, cleavage of the glycosidic bond, condensation of the glycosyl group, and ultimately degradation of the molecule through acid- and alkali-catalyzed reactions. Analysis of the products obtained from the pyrolysis of 1,6-anhydro-β-D-glucopyranose (levoglucosan), before and after treatment with zinc chloride or sodium hydroxide, is shown in Table I.[7] This table shows that addition of zinc chloride promotes the formation of char, water, and 2-furaldehyde, which are the expected products of an acid-catalyzed dehydration reaction,[11,12] whereas the addition of alkali promotes the production of low-molecular-weight carbonyl compounds, which may be accounted for by base-catalyzed rearrangement and fragmentation reactions of carbohydrates.[13]

These data have been combined with the radiochemical patterns obtained by tracing the individual products to different segments of specifically labeled samples of 1,6-anhydro-β-D-glucopyranose-^{14}C to obtain more precise information about mechanisms of pyrolytic reactions.

TABLE I

PYROLYSIS PRODUCTS OF 1,6-ANHYDRO-β-D-GLUCOPYRANOSE

Product	Yield, % Neat	$+ZnCl_2$	$+NaOH$
Acetaldehyde	1.1	0.3	7.3
Furan	1.0	1.3	1.6
Acrolein	1.7	<0.1	2.6
Methanol	0.3	0.4	0.7
2,3-Butanedione	0.5	0.8	1.6
2-Butenal	0.7	0.2	2.2
1-Hydroxy-2-propanone	0.8	<0.1	1.1
Glyoxal	1.4	<0.1	4.9
Acetic acid	1.7	0.7	1.5
2-Furaldehyde	0.9	3.0	0.4
5-Methyl-2-furaldehyde	0.1	0.3	
Carbon dioxide	2.9	6.8	5.7
Water	8.7	20.1	14.1
Char	3.9	29.0	16.0
Balance (tar)	74.3	36.8	40.3

Results and Discussion

Samples of 1,6-anhydro-β-D-glucopyranose-*1*-^{14}C, -*2*-^{14}C, and -*6*-^{14}C were prepared from the corresponding labeled D-glucose by the standard method[14] involving alkaline hydrolysis of phenyl β-D-glucopyranosides. The products were pyrolyzed without any additive and in the presence of 5% sodium hydroxide or 5% zinc chloride. In one experiment 1,6-anhydro-β-D-glucopyranose was prepolymerized in the presence of 5% zinc chloride before pyrolysis. These experiments gave several samples of carbon dioxide, carbon monoxide, and aqueous pyrolysate containing a variety of carbonyl compounds. Samples of carbon dioxide were converted to barium carbonate. The carbon monoxide samples were oxidized to carbon dioxide by iodine pentoxide[15] and also recovered as barium carbonate. The carbonyl compounds present in the pyrolysates were converted to the 2,4-dinitrophenylhydrazone (DNPH) derivatives, by treatment with acidic 2,4-dinitrophenylhydrazine, and the resulting DNPH mixtures were separated by thin layer chromatography.[16] The separation gave sufficient quantities of the DNPH derivatives of 2-furaldehyde, 2,3-butanedione, pyruvaldehyde, acetaldehyde, and glyoxal for radioanalysis. All these carbonyl compounds and some of the related hydroxy

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[1] Established through a grant from the Hoerner-Waldorf Corporation of Montana.

[2] F. Shafizadeh, *Advan. Carbohyd. Chem.*, **23**, 419 (1968).

[3] J. B. Berkowitz-Mattuck and T. Noguchi, *J. Appl. Polym. Sci.*, **7** 709 (1963)

[4] G. A. Byrne, D. Gardiner, and F. H. Holmes, *J. Appl. Chem.*, **16**, 81 (1966).

[5] K. Kato and H. Komorita, *Agr. Biol. Chem.*, **32** (6), 715 (1968); K. Kato, T. Doihara, F. Sakai, and N. Takahashi, *Sci. Pap. Cent. Res. Inst., Jap. Monop. Corp.*, **108**, 361 (1966).

[6] F. Shafizadeh, G. D. McGinnis, R. A. Susott, and H. W. Tatton, *J. Org. Chem.*, **36**, 2813 (1971).

[7] F. Shafizadeh, C. W. Philpot, and N. Ostojic, *Carbohyd. Res.*, **16**, 279 (1971).

[8] F. Shafizadeh and G. D. McGinnis, *ibid.*, **16**, 273 (1971).

[9] F. Shafizadeh, G. D. McGinnis, R. A. Susott, and C. W. Philpot, *ibid.*, **15**, 165 (1970).

[10] G. W. Smith and F. Shafizadeh, *J. Chem. Soc. B*, 908 (1971).

[11] F. H. Newth, *Advan. Carbohyd. Chem.*, **6**, 83 (1951).

[12] E. F. L. J. Anet, *ibid.*, **19**, 181 (1964).

[13] R. L. Whistler and J. N. BeMiller, *ibid.*, **13**, 326 (1958).

[14] G. H. Coleman, *Methods Carbohyd. Chem.*, **2**, 397 (1963).

[15] M. Steinberg, A. Glasner, and E. Levy, *Anal. Chem.*, **34**, 1629 (1962).

[16] G. A. Byrne, *J. Chromatogr.*, **20**, 528 (1965).

TABLE II

SPECIFIC RADIOACTIVITIES OF 1,6-ANHYDRO-β-D-GLUCOPYRANOSE-*1*-^{14}C, -*2*-^{14}C, AND -*6*-^{14}C, AND THEIR PYROLYSIS PRODUCTS IN 10^2 μCi/MOL

	Neat			5% NaOH			5% $ZnCl_2$			5% $ZnCl_2$, polymerized		
Compd	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C
1,6-Anhydro-β-D-glucopyranose	6.08	3.53	5.42	6.08	3.53	5.42	6.08	3.53	5.42	6.08	3.53	5.42
2-Furaldehyde	3.70	3.65	1.94	1.84	3.55	3.97	5.20	3.36	0.90	4.88	3.42	1.17
2,3-Butanedione	1.51	1.93	1.70	1.00	1.09	3.07	3.90	2.02	1.46			
Pyruvaldehyde	1.66	0.93	1.04	1.41	1.48	1.67	3.01	1.65	1.61	2.40	1.63	2.06
Acetaldehyde	0.61	1.08	1.96	0.038	1.03	3.00	0.026	0.026	1.62			
Glyoxal	0.93	0.68	0.038	1.54	1.70	1.96	1.78	1.00	1.54	2.17	1.18	1.44
Carbon dioxide	2.08	0.87	0.034	1.90	0.62	0.048	2.65	1.17	0.052	3.03	1.00	0.53
Carbon monoxide	1.28	0.67	0.92	2.32	0.63	0.07	2.22	0.97	0.62	2.88	0.88	0.66

TABLE III

PERCENTAGE OF THE PYROLYSIS PRODUCTS TRACED TO THE LABELED CARBONS OF 1,6-ANHYDRO-β-D-GLUCOPYRANOSE

	Neat			5% NaOH			5% $ZnCl_2$			5% $ZnCl_2$, polymerized		
Compd	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C	*1*-^{14}C	*2*-^{14}C	*6*-^{14}C
2-Furaldehyde	60.8	103.4	35.8	30.2	100.7	73.0	86.0	95.8	16.6	80.3	96.9	21.6
2,3-Butanedione	24.8	54.6	31.3	16.5	31.0	56.5	64.8	57.7	26.9			
Pyruvaldehyde	27.3	26.3	19.1	23.3	42.0	30.7	49.7	46.7	29.6	39.5	46.2	38.0
Acetaldehyde	10.1	30.5	36.0	6.3	29.2	55.1	4.4	7.3	29.8			
Glyoxal	15.4	19.2	6.9	25.5	48.3	36.0	29.5	28.2	28.2	35.7	33.4	26.6
Carbon dioxide	34.3	24.5	6.3	31.2	17.7	8.9	43.7	33.3	9.5	49.8	28.3	9.8
Carbon monoxide	21.1	18.9	16.8	38.4	18.0	13.7	36.7	27.6	11.4	47.4	24.9	12.2

derivatives have been previously identified among the pyrolysis products of cellulose and levoglucosan.[2,7,17,18] The last three bis-DNPH derivatives could have also been obtained from the corresponding α-hydroxycarbonyl compounds under the employed experimental conditions.[16] Thus, the 2,3-butanedione bis-DNPH may have been also derived from 2-hydroxy-3-butanone, the pyruvaldehyde bis-DNPH from 1-hydroxy-2-propanone or 2-hydroxypropanal, and the glyoxal bis-DNPH from glycolaldehyde. Quantitative analysis of these products carried out by gas chromatography is shown in Table I.

The specific radioactivity of the labeled levoglucosan samples and the isolated products given in Table II were determined by the standard liquid scintillation and gel-suspension counting techniques.[19] The radiochemical data gave the percentage of each product originating from C-1, C-2, and C-6 positions of the sugar molecule (see Table III). The resulting data on radiochemical patterns and the yields of each product, on pyrolysis of the anhydro sugar under the acidic, neat, alkaline, and prepolymerized conditions, were used for unravelling the nature of the reactions involved.

2-Furaldehyde.—Furan compounds are generally formed from the acid-catalyzed dehydration of carbohydrates[11,12] and are not very likely to involve recombinations of the sugar fragments. On this basis 2-furaldehyde could have been derived either from the first or the last five carbons of the anhydro sugar. The formation of 2-furaldehyde from these fragments is confirmed by the radiochemical data which, within ±4% experimental error, show that in all cases it contains 100% of C-2. However, under the acidic condition this product originates about 86% from the first five carbons and 17% from the last. Under the alkaline condition the situation is reversed and about 30% originates from the first five carbons and 73% from the last. In the absence of any additives the results obtained are more similar to the acid-catalyzed rather than the base-catalyzed condition.

Formation of 2-furaldehyde from C-1 to C-5 is consistent with the acid-catalyzed degradation pathway of carbohydrates involving the conversion of the enolic forms of intermediate 3-deoxyglycosuloses to furan compounds.[12] It is also consistent with the observations of Kato and coworkers,[5] who have identified 3-deoxyglycosuloses among the pyrolysis products of cellulose, D-glucose, D-fructose, and D-xylose and have shown that 5-(hydroxymethyl)-2-furaldehyde could form 2-furaldehyde. The 3-deoxyglycosuloses are produced by a general acid- and alkali-catalyzed reaction of carbohydrates.[12,13,20] They are even formed during the processing and storage of food.[21–23]

Considering that cleavage of the glycosidic bond, reversible polymerization, and opening of the ring structure could readily take place under the pyrolytic conditions,[6] the reactions in Scheme I account for the formation of 2-furaldehyde from C-1 to C-5 of 1,6-anhydro-β-D-glucopyranose. According to this scheme, 2-furaldehyde may be formed either directly from 1,6-anhydro-β-D-glucopyranose or from its polymerization product. Since it is known that the anhydro sugar is readily polymerized on heating in presence of an acidic catalyst[7,24,25] and the polymeric material provides the same product and isotopic pattern as the anhydro sugar, the latter possibility cannot be ignored.

The competing pathway which is the main source of 2-furaldehyde under the alkaline condition is related

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[17] R. G. Schwenker, Jr., and L. R. Beck, Jr., *J. Polym. Sci., Part C*, (**2**), 331 (1963).

[18] S. Glassner and A. R. Pierce, III, *Anal. Chem.*, **37**, 525 (1965).

[19] J. C. Turner, *Int. J. Appl. Radiat. Isotopes*, **20**, 761 (1969).

[20] J. C. Speck, Jr., *Advan. Carbohyd. Chem.*, **13**, 63 (1958).

[21] A. A. El-Dash and J. E. Hodge, *Carbohyd. Res.*, **18**, 259 (1971).

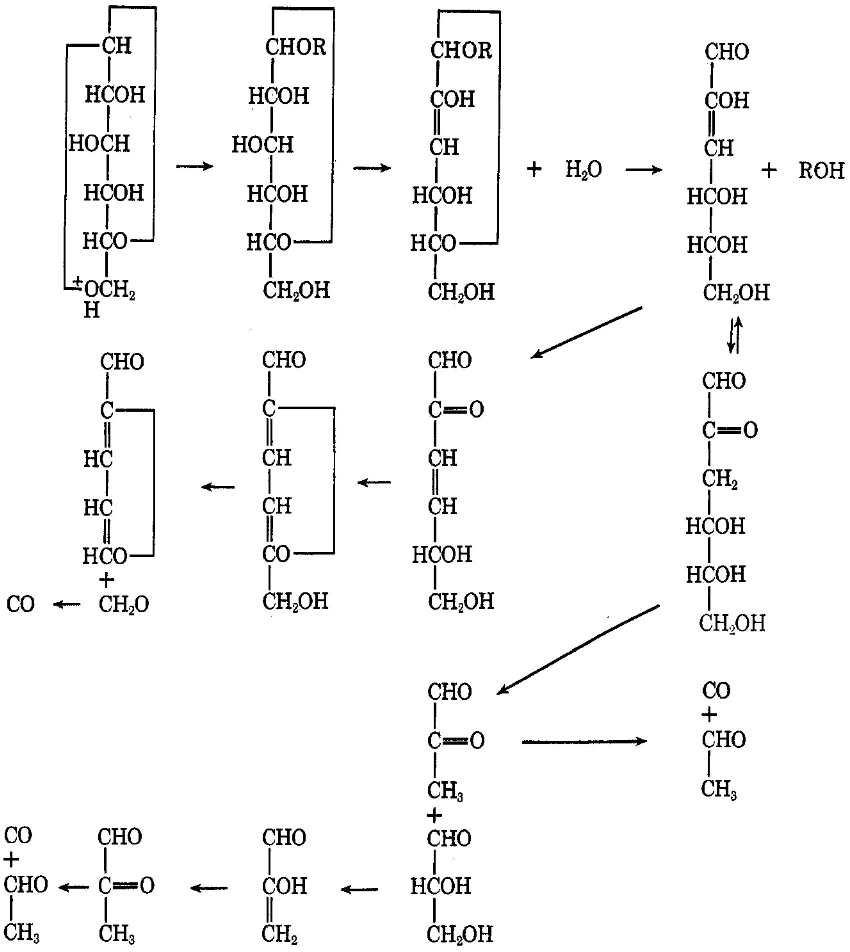
[22] H. Kato and Y. Sakurai, *Nippon Nogeikagaku Kaishi*, **38**, 536 (1964); *Agr. Biol. Chem.*, **28**, No. 12, A46 (1964).

[23] S. Oka, *Agr. Biol. Chem.*, **33**, 554 (1969).

[24] M. L. Wolfrom, A. Thompson, and R. B. Ward, *J. Amer. Chem. Soc.*, **81**, 4623 (1959).

[25] J. da S. Carvalho, W. Prins, and C. Schuerch, *ibid.*, **81**, 4054 (1959).

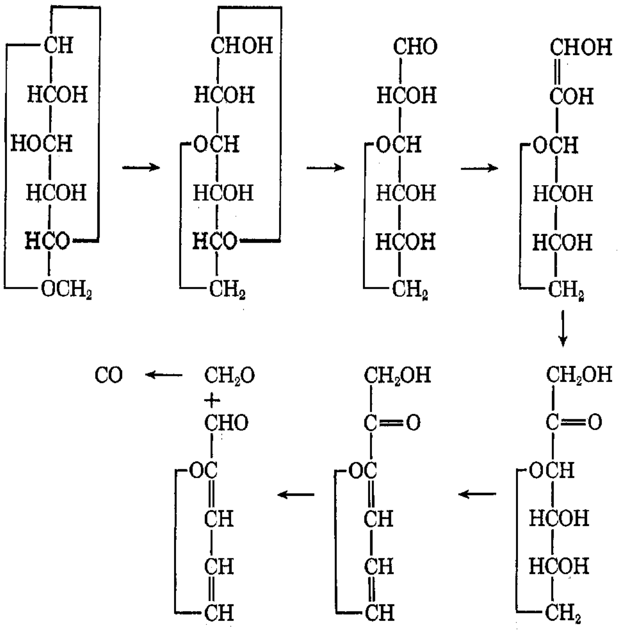
SCHEME I[a]



[a] R = H or D-glucose units.

to the observation of Gardiner,[26] who has isolated 2-furyl hydroxymethyl ketone from the pyrolysis of cellulose, 3,6-anhydro-D-glucose, and other sugars. In this pathway (Scheme II), formation of the 3,6-anhydro ring leads to production of 2-furyl hydroxymethyl ketone, which on degradation gives 2-furaldehyde from the C-2–C-6 fraction of the original compound.

SCHEME II



Since, as shown in Table I, the yield of 2-furaldehyde is substantially increased in the presence of zinc chloride and reduced with addition of sodium hydroxide, it could be surmised that the former pathway is enhanced by acidic conditions, whereas the latter pathway is not promoted by alkali and merely becomes dominant because the competing mechanism is hindered.

The two competing pathways even apply to aqueous reactions, because minor quantities of 2-furyl hydroxymethyl ketone have been obtained from the treatment of sucrose and D-fructose with aqueous acid, which results mainly in the formation of 5-(hydroxymethyl)-2-furaldehyde.[27] Although the formation of furan derivatives is catalyzed by acids, the 3-deoxy precursor, as noted before, is produced under both acidic and alkaline conditions.[12,20] Under the alkaline condition, however, it more readily undergoes dealdolization and other degradation reactions.

2,3-Butanedione.—This compound also follows the pattern shown by 2-furaldehyde. Under the acidic condition it is derived mainly from C-1 (65%) and under the alkaline condition mainly from the C-6 (56%). However, in contrast to 2-furaldehyde the radiochemical data indicate some fragment recombination which in this case is quite feasible and may be

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[26] D. Gardiner, *J. Chem. Soc. C*, 1473 (1966).

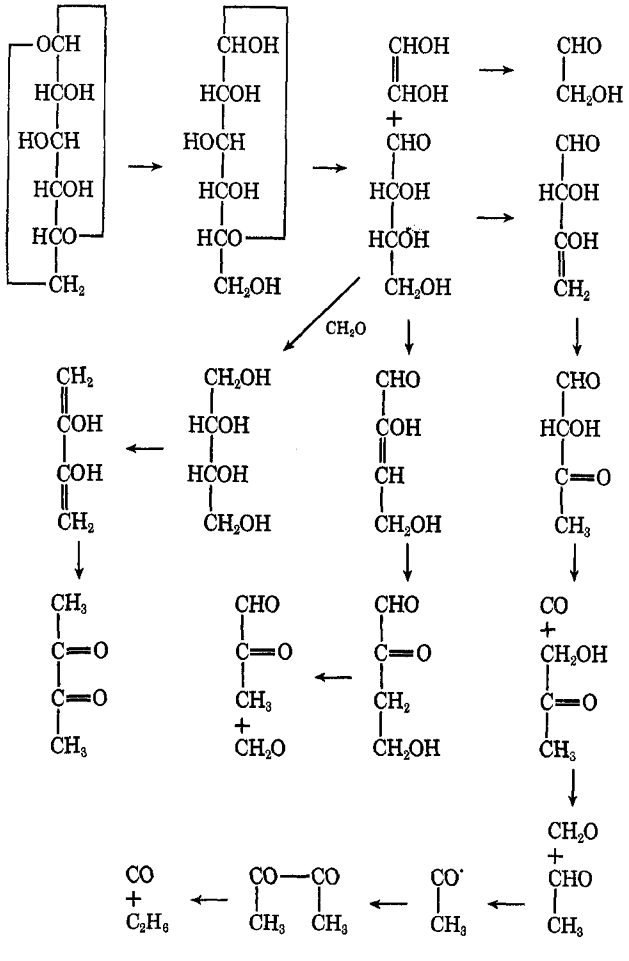
[27] R. E. Miller and S. M. Cantor, *J. Amer. Chem. Soc.*, **74** 5236 (1952).

due to aldol condensation or combination of $CH_3CO\cdot$ free radicals.[2,3] If no fragment recombination had occurred, all of the product should have been derived from C-1–C-4 (25%), C-2–C-5 (30%), and C-3–C-6 (31%). This leads to a discrepancy of about 14%, which must be accounted for by the recombination of nonradioactive carbon fragments from C-3 to C-5.

The same logic also applies to the radiochemical data obtained under the alkaline and acidic conditions. Recombination of free radical fragments is also supported by detection of esr signals and investigations of Heyns and Klier,[28] who have shown that pyrolysis of glyceraldehyde gives acetaldehyde and 2,3-butanedione as the first and second largest products. However, these authors also have shown that 2,3-butanedione is the major pyrolysis product of D-erythrose and the radiochemical patterns show a high degree of specificity rather than randomness expected from extensive fragment recombination. Therefore, it seems very likely that the observed patterns result from the breakdown of the sugar moiety into a four-carbon fragment.

Scheme III shows cleavage of the sugar molecule under alkaline condition to D-erythrose that is converted directly or indirectly into 2,3-butanedione and other products. The direct conversion should involve intermolecular disproportionation of the aldotetrose. According to Scheme III the anhydro ring is first opened and the resulting D-glucose moiety then breaks down to the enolic form of glycolaldehyde and D-erythrose. Opening of the anhydro ring conforms with the established alkaline hydrolysis of the glycosidic bond at high temperatures,[29] but under pyrolytic conditions it is accompanied by degradation of the molecule. Further transformation of glycolaldehyde and D-erythrose gives glyoxal from C-1–C-2 and 2,3-butanedione, 1-hydroxy-2-propanone (isolated as pyruvaldehyde), and acetaldehyde carrying the C-6 label.

SCHEME III

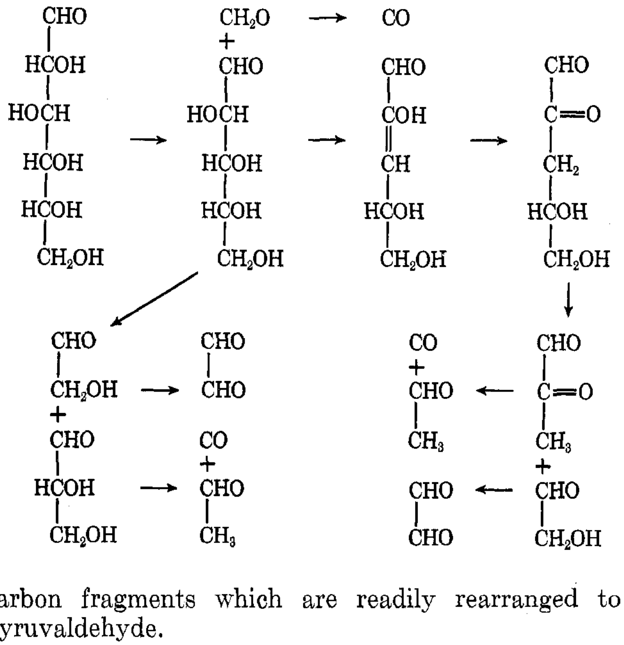


Pyruvaldehyde.—Assuming that pyruvaldehyde and 1-hydroxy-2-propanone are formed only through primary fragmentation of the sugar, the results given in Table III could be further analyzed to show the contribution of the central fragment C-3–C-5 which amounts to 54% for uncatalyzed, 28% for the alkali, and 20% for the acid-catalyzed conditions. This indicates that pyruvaldehyde is derived mainly from C-3–C-5 under the uncatalyzed condition and from C-1–C-3 and C-4–C-6 under the acidic condition. Under the alkali condition it is formed in a more random fashion from C-1–C-3, C-2–C-4, C-3–C-5, and C-4–C-6.

The radiochemical pattern of pyruvaldehyde under the acidic condition may be attributed to the degradation of 3-deoxy-D-*erythro*-hexosulose. As seen in Scheme I, this compound could break to pyruvaldehyde and glyceraldehyde, which is further pyrolyzed to pyruvaldehyde, acetaldehyde, and 2,3-butanedione carrying the C-6 label.

The same reactions could take place under the alkaline condition. However, in these cases the D-glucose moiety formed after opening of the anhydro ring could also break down through Scheme III or more randomly through dealdolization (Scheme IV), to give three carbon fragments which are readily rearranged to pyruvaldehyde.

SCHEME IV



Acetaldehyde.—This compound is formed heavily from C-6 and lightly from C-1. Under the alkaline condition more than half (55%) of the acetaldehyde contains the terminal carbon atom of the anhydro sugar. The high radiochemical yields of acetaldehyde from C-6 under acidic, neat, and alkaline conditions

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[28] K. Heyns and M. Klier, *Carbohyd. Res.*, **6**, 436 (1968).

[29] Elizabeth Dryselius, B. Lindberg, and O. Theander, *Acta Chem. Scand.*, **12**, 340 (1958); A. Dyfverman and B. Lindberg, *ibid.*, **4**, 879 (1950).

and its partial formation from C-2 under the last two conditions are consistent with the thermal degradation pathways presented by Schemes I and III.

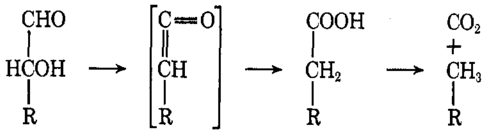
Glyoxal.—The radiochemical data indicate that glyoxal and its precursor, glycolaldehyde, are derived from all the positions, although under the alkaline and acidic conditions the terminal carbons are more favored. Schemes I, III and IV show how these compounds could be formed from the different fragments.

It should be noted that the yields of acetaldehyde, glyoxal, and 1-hydroxy-2-propanone are substantially increased with the addition of sodium hydroxide.[7] The increased formation of these carbonyl compounds under alkaline condition strongly confirms the proposed dealdolization mechanisms, which have many counterparts among the normal alkaline degradation reaction of carbohydrates.[13,20]

Carbon Dioxide.—This compound originates mainly from C-1 and C-2 positions in all cases. However, the specificity is highest under acid condition and lowest under alkali condition.

Formation of carbon dioxide from C-1 may be attributed to the benzylic acid rearrangement of the 3-deoxy-D-*erythro*-hexosulose and pyruvaldehyde to 3-deoxy-D-hexonic acids (metasaccharinic acid) and lactic acid followed by decarboxylation. However, since relatively large quantities of carbon dioxide are formed from both acid and alkaline conditions and the reaction takes place even under mild pyrolytic conditions with cellulose[2] and other carbohydrates, including D-glucose,[30] it seems that a more direct pathway should be involved. A dehydration and rehydration rearrangement at C-1 and C-2 of the original sugar and decarboxylation of the product according to Scheme V could readily account for the carbon dioxide formation. This arrangement proceeds through a ketene intermediate to form a carboxylic acid that is decarboxylated under the pyrolytic conditions. Although to our knowledge there is no known precedent for this reaction with carbohydrates, the formation of ketenes from carbonyl compounds under the pyrolytic conditions is well known.[31]

Scheme V



Various aldehydes, particularly acetaldehyde, could also form carboxylic acids and primary alcohols through intermolecular disproportionation involving a hydride shift. Formation of methanol and acetic acid shown in Table I and erythritol postulated in Scheme III confirm this hypothesis.

Carbon Monoxide.—The radiochemical patterns obtained for carbon monoxide are very similar to those of carbon dioxide. The relatively heavier formation of this compound from C-1 and C-2 is consistent with the proposed schemes in which carbon monoxide is derived by decarbonylation of the various aldehydes[32,33] and decomposition of formaldehyde, under the pyrolytic conditions.[34]

The carboxylic acids derived from the rearrangement or disproportionation reactions could also provide carbon monoxide through decarbonylation.[35,36] Formation of both carbon dioxide and carbon monoxide from C-1 of the aldehydes is further confirmed by the closely similar isotopic patterns that have been obtained for the two compounds.

The above schemes by no means represent all the reactions which take place on pyrolysis of carbohydrates. However, they clearly indicate the nature and mechanism of the reactions involved and lead to the following general conclusions.

Pyrolysis of carbohydrates not only provides a variety of products but also leads to the formation of the individual products from different positions of the sugar molecule, through competing pathways which fall short of complete randomization.

Within each pathway the individual transformations are remarkably similar to known aqueous reactions, especially the acid- and alkali-catalyzed degradation of carbohydrates, but the products formed by the combination of these reactions are further randomized by decarboxylation, decarbonylation, disproportionation, and other molecular rearrangements which are more prevalent at high temperatures.

In the absence of solvent the compelling forces for molecular rearrangements are provided more by an overabundance of energy than by the normal intermolecular and ionic interactions. Thermal anomerization of α-D-xylose, which proceeds as the crystalline material is melted,[6] clearly shows this point. A variety of ionic and solvent interactions have been proposed for cleavage of the cyclic structure in solution which are not applicable to the molten state.[37]

The competing pathways are controlled by ionic species present and the statistical possibility for the first point of attack or cleavage within the glycosyl unit. Under the acidic conditions the molecule or its polymerization product degrades by eliminations of various bonds and hydroxyl groups yielding substantial amounts of water and char (see Table I). Under the alkaline conditions cleavage of the anhydro ring and breakdown of the sugar mainly through reverse aldolization gives a variety of carbonyl compounds. There is no clear line of demarcation between the two types of pathways and both of them take place in the absence of additives. The pattern for pyrolysis of the neat substrate, however, is closer to the acidic conditions, presumably due to the formation of carboxylic acids.

Since the above reactions are of a general nature, the pyrolysis products of homologous carbohydrate compounds like cellulose, starch, 1,6-anhydro-β-D-glucose, D-glucosides, and D-glucose should be similar. There is a considerable amount of experimental support for this conclusion,[2,7,18] although it does not jibe with

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(30) Y. Houminer and S. Patai, *Tetrahedron Lett.*, 1297 (1967).

(31) J. W. Williams and C. D. Hurd, *J. Org. Chem.*, **5**, 122 (1940).

(32) H. Burkett, W. M. Schubert, F. Schultz, R. B. Murphy, and R. Talbott, *J. Amer. Chem. Soc.*, **81**, 3923 (1959).

(33) J. F. Bunnett, J. H. Miles, and K. V. Nahabedian, *ibid.*, **83**, 2512 (1961).

(34) A. R. Hall, J. C. McCoubrey, and H. G. Wolfhard, *Combust. Flame*, **1**, 53 (1957).

(35) E. O. Wilg, *J. Amer. Chem. Soc.*, **52**, 4729 (1930).

(36) G. A. Ropp, *ibid.*, **82**, 842 (1960).

(37) H. S. Isbell and W. Pigman, *Advan. Carbohyd. Chem.*, **24**, 13 (1969).

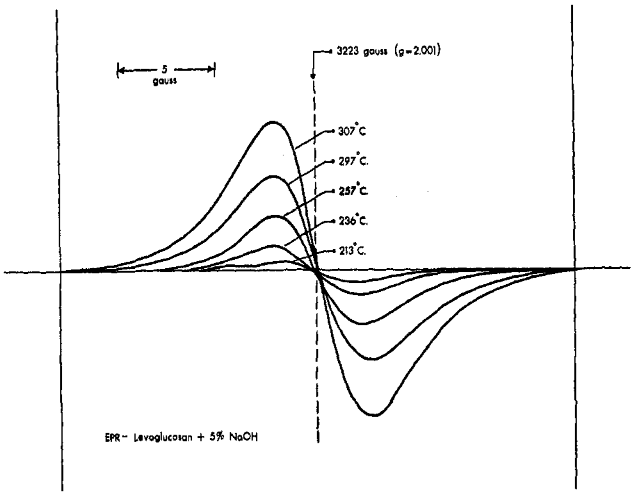

Figure 1.—The esr signals of 1,6-anhydro-β-D-glucopyranose treated with 5% sodium hydroxide and pyrolyzed at different temperatures.

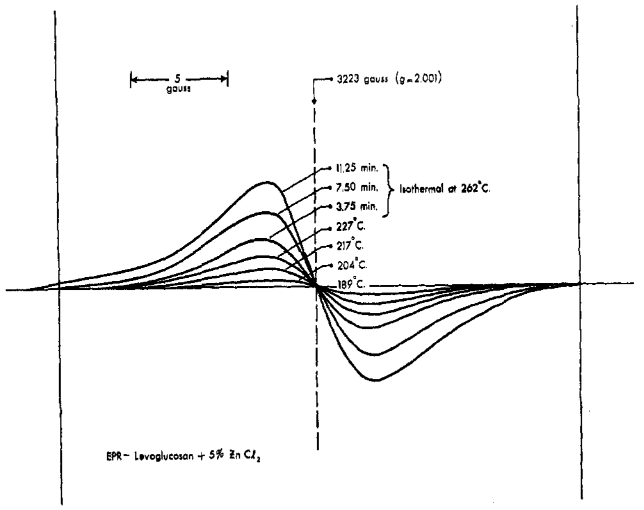

Figure 2.—The esr signals of 1,6-anhydro-β-D-glucopyranose treated with 5% zinc chloride and pyrolyzed under different conditions.

all the data reported by Heyns and Klier.[28] Development of almost the same products[7] and isotopic patterns from the anhydro sugar and its polymerization product in the presence of zinc chloride also confirms this conclusion.

Other than the ionic reactions, homolytic cleavage also plays a significant role in the pyrolytic degradation. However, the signals detected by esr spectroscopy (see Figures 1 and 2) are mainly associated with the stable carbonacious residue, rather than transient free radical intermediates. Some aspects of the free radical formations and the kinetics of the pyrolytic reactions will be discussed in a following report. The subjects that are currently under investigation should also shed some light on the significance of levoglucosan as a model compound for thermal degradation of starch and cellulose and provide a comparison between the pyrolytic reactions and field ionization which takes place on mass spectroscopy.

Experimental Section

Preparation of Samples.—1,6-Anhydro-β-D-glucopyranose labeled at positions 1, 2, or 6 was prepared by standard methods[14] from commercially available D-glucose-*1*-^{14}C, -*2*-^{14}C, and -*6*-^{14}C diluted with nonradioactive materials (5 g). Small portions of the product were dissolved in methanol and mixed with calculated amounts of a solution of sodium hydroxide in methanol or zinc chloride in tetrahydrofuran. The solvents were then removed under vacuum at 50° to give samples of 1,6-anhydro-β-D-glucopyranose containing 5% sodium hydroxide or zinc chloride. The dried materials were kept under anhydrous condition.

Polymerization of the anhydro sugars containing 5% zinc chloride was carried out by heating 100-mg portions at 150° for 30 min in ampoules sealed under a nitrogen atmosphere.[25] Examination of nonradioactive samples showed the presence of 30% of a polymer which precipitated from 85% ethyl alcohol and the absence of any monomeric material that could be detected by tlc. The entire sample of the polymerized radioactive material in each ampoule was used for the pyrolysis experiments.

Pyrolysis.—Samples of treated and untreated ^{14}C-labeled 1,6-anhydro-β-D-glucopyranose (100 mg) were placed in small vials which were introduced into the pyrolysis apparatus consisting of a modified Sargent microcombustion unit attached to a series of receptacles. The system was thoroughly flushed with nitrogen and the sample was pyrolyzed by heating for 8 min at 600°. The pyrolysis products were swept through the system for 3 hr with a gentle stream of nitrogen. The pyrolysate containing carbonyl compounds was condensed in a small flask cooled in a Dry Ice–acetone bath. The carbon dioxide was recovered as barium carbonate in traps containing barium hydroxide solution.[38] The carbon monoxide remaining in the stream was dried by passing through drying tubes ($CaCl_2$ and P_2O_5), oxidized with iodine pentoxide,[15] and collected as barium carbonate in the last traps.

Isolation of the Pyrolysis Products.—The pyrolysate condensed in the cooled flask was combined with washings from the adjoining tubes (5 ml) and treated at room temperature with 10 ml of a saturated solution of 2,4-dinitrophenylhydrazine in 2 *N* HCl[39] for 18 hr. The precipitate of mixed DNPH derivatives was filtered, washed with water, and dried, yield 12 mg. The mixture was dissolved in 6 ml of chloroform, and 1-ml portions of the solution were placed as a line on Baker-flex silica gel IB-F tlc sheet and were developed in three stages with benzene. This gave six major zones in addition to the original strip. The top five zones from chromatograms were collected and extracted with chloroform. The extract was concentrated and rechromatographed, and the developed zones were processed to provide the compounds listed in Table IV.[40-42]

Radiochemical Assay.—The samples were counted with the Tri-Carb liquid scintillation spectrometer model 314 E operated at 6° using a scintillation mixture (Permablend I consisting of 91% PPO and 9% Dimethyl POPOP) produced by Packard Instrument Co. Before counting, the samples were stored in the counter for 20 min to eliminate the effect of light. Duplicate samples were counted five times, each time for 10 min to reduce the random counting error to less than 2%.

Samples of ^{14}C-labeled 1,6-anhydro-β-D-glucopyranose (1 mg) were weighed in a scintillation vial and dissolved in 10 ml of a toluene–methanol mixture (8:2) containing 0.4% of Permablend I. The solutions were counted using toluene-^{14}C as a reference, with the counting efficiency of 61.76%.

The gel-suspension technique[19] was employed for counting the samples of barium carbonate and DNPH derivatives absorbed on tlc silica gel. The barium carbonate sample (~0.5 mg) and 400 mg of Cab-O-Sil gel forming reagent were weighed in a scintillation vial. The mixture was shaken with 10 ml of toluene containing 1% of Permablend I to form a gel. The gel was counted using standard ^{14}C-labeled barium carbonate as a reference, with the counting efficiency of 40.2%.

The DNPH derivatives absorbed on silica were collected from tlc zones and homogenized. A sample of the homogenized material (0.2 g) was extracted with 25 ml of chloroform and the solution was used for determining the concentration of the DNPH derivative by uv spectroscopy. Another sample of the silica powder containing a DNPH derivative (300 mg) was used for preparation of the gel suspension as before. The DNPH deriva-

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(38) F. Eisenberg, Jr., *J. Amer. Chem. Soc.*, **76**, 5152 (1954).

(39) H. A. Iddles and C. E. Jackson, *Anal. Chem.*, **6**, 454 (1934).

(40) L. A. Jones, J. C. Holmes, and R. B. Seligman, *ibid.*, **28**, 191 (1956).

(41) H. H. Strain, *J. Amer. Chem. Soc.*, **57**, 758 (1935).

(42) M. L. Wolfrom and G. P. Arsenault, *Anal. Chem.*, **32**, 693 (1960).

TABLE IV

PHYSICAL PROPERTIES OF 2,4-DINITROPHENYLHYDRAZONE DERIVATIVES ISOLATED FROM THE PYROLYSIS PRODUCTS

	Found				Literature			
			Uv (CHCl$_3$)			Uv (CHCl$_3$)		
	R_f[b]	Mp, °C	λ_{max}, mμ[a]	ε × 10^{-4}	Mp, °C	λ_{max}, mμ	ε × 10^{-4}	Ref
2-Furaldehyde	0.32	222–224	388*	2.90	225	386	2.65	40
2,3-Butanedione (bis)	0.16	312–314	394*, 442	2.92	314–315			41
Pyruvaldehyde (bis)	0.11	298–301	394*, 444	3.81	299–300			41
					304–305			42
Acetaldehyde	0.30	164–165	354*	2.22	167	354	2.22	40
Glyoxal (bis)	0.07	330–333	390, 445*	2.42	326–328			41
					336–338			42

[a] Starred wavelengths denote major maxima. [b] In benzene.

tives produced a strong quenching effect on scintillation that was measured by using ^{14}C-labeled toluene as an internal standard. The counting efficiency varied within the range of 10–30% according to the sample and concentration.

Esr Spectroscopy.—Samples of the anhydro sugar (1 part) were mixed with ground glass (9 parts) and ground together thoroughly to ensure uniform mixing. The ground samples (4–7 mg) were accurately weighed into a 2-mm capillary tube. The tube was placed into the cavity of a Varian E-3 esr spectrometer heated with a specially designed variable-temperature accessory.

Registry No.—1,6-Anhydro-β-D-glucopyranose, 498-07-7.

Acknowledgments.—The authors are pleased to acknowledge the assistance of Drs. S. L. Jindal and R. A. Susott in preparation of the ^{14}C-labeled 1,6-anhydro-β-D-glucose samples and esr studies, respectively. They also thank the U. S. Public Health Service, Food and Drug Administration for Grant No. FD-00036 in support of this work.

Studies on the Vilsmeier–Haack Reaction. IV.[1] Convenient Synthesis of 2,2′-Anhydro-1-β-D-arabinofuranosylcytosine (2,2′-Cyclocytidine) and Its Derivatives[2]

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Received July 6, 1971

A carcinostatic nucleoside, 2,2′-anhydro-1-β-D-arabinofuranosylcytosine (2,2′-cyclocytidine) (**5**), was prepared in a yield of 55% by treatment of cytidine (**4**) with Vilsmeier–Haack reagent **1** or **2**. 5′-Chloro-5′-deoxy-2,2′-anhydro-1-β-D-arabinofuranosylcytosine (**6**) and 2′,5′-dichloro-2′,5′-dideoxycytidine (**7**) were also prepared by prolonged treatment of **4** with **1**. Treatment of **5** and **6** with mild alkali gave 1-β-D-arabinofuranosylcytosine (**9**) and 5′-chloro-5′-deoxy-1-β-D-arabinofuranosylcytosine (**10**), respectively, whereas treatment of either of **6** and **7** with strong alkali gave 2′,5′-anhydro-1-β-D-arabinofuranosylcytosine (**11**).

2,2′-Anhydro-1-β-D-arabinofuranosylcytosine (2,2′-cyclocytidine) (**5**) has been shown to be an intermediate[3-5] for the synthesis of a carcinostatic nucleoside, 1-β-D-arabinofuranosylcytosine (**9**),[6] and by itself a potent carcinostatic agent.[7] 1-β-D-Arabinofuranosylcytosine (**9**) has been synthesized by several procedures, such as (a) from cytidine *via* 2,2′-anhydro intermediates,[3,8,9] (b) from 1-β-D-arabinofuranosyluracil,[10] or (c) from the appropriate sugars,[11-13] but most of these involve tedious steps. Recently, **5** and **9** were successfully synthesized[14] directly from **4** by use of a partially hydrolyzed phosphorus oxychloride.[15] We wish to report an improved method to prepare **5**, **9**, and their derivatives.

N,N′-Dimethylformamide (DMF) combines with inorganic acid halides to form active reagents (Vilsmeier–Haack reagents),[16-18] which are useful as formylating, halogenating, and dehydroxylating agents.[19] Thus, phosphorus oxychloride and thionyl chloride react with DMF to form the complex **1**[18] and the complex **2**,[17] respectively (Scheme I). The latter may be converted into the crystalline complex **3** by removal of sulfur dioxide,[17] and **3** re-forms **2** on addition of sulfur dioxide.[20] The reaction of nucleosides with the com-

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(1) Part III: K. Kikugawa, M. Ichino, and T. Kawashima, *Chem. Pharm. Bull.*, **19**, 2466 (1971).

(2) (a) A preliminary account of part of this work has appeared in K. Kikugawa and M. Ichino, *Tetrahedron Lett.*, 867 (1970). (b) This paper was presented at the 91th Annual Meeting of the Pharmaceutical Society of Japan, April 8, 1971, Fukuoka, Japan. (c) The commonly applied term "2,2′-cyclocytidine" has been retained in the title for the convenience of "key word index" users.

(3) E. R. Walwick, W. K. Roberts, and C. A. Dekker, *Proc. Chem. Soc.*, 84 (1959).

(4) I. L. Doerr and J. J. Fox, *J. Org. Chem.*, **32**, 1465 (1967).

(5) W. V. Ruyle and T. Y. Shen, *J. Med. Chem.*, **10**, 331 (1967).

(6) S. S. Cohen, "Progress in Nucleic Acid Research and Molecular Biology," Vol. V, J. N. Davidson and W. E. Cohn, Ed., Academic Press, New York, N. Y., 1966, p 1.

(7) A. Hoshi, F. Kanzawa, K. Kuretani, M. Saneyoshi, and Y. Arai, *Gann*, **62**, 145 (1971).

(8) W. K. Roberts and C. A. Dekker, *J. Org. Chem.*, **32**, 816 (1967).

(9) H. P. M. Fromageot and C. B. Reese, *Tetrahedron Lett.*, 3499 (1966).

(10) J. S. Evans, E. A. Musser, G. D. Mengel, K. R. Forsblad, and J. H. Hunter, *Proc. Soc. Exp. Biol. Med.*, **106**, 350 (1961).

(11) T. Y. Shen, H. M. Lewis, and W. V. Ruyle, *J. Org. Chem.*, **30**, 835 (1965).

(12) B. Shimizu and F. Shimizu, *Chem. Pharm. Bull*, **18**, 1060 (1970).

(13) L. E. Orgel and R. A. Sanchez (*Chem. Eng. News*, Oct 6, 1969) synthesized **9** with a plausible prebiosynthetic route.

(14) T. Kanai, T. Kojima, O. Maruyama, and M. Ichino, *Chem. Pharm. Bull.*, **18**, 2569 (1970).

(15) Y. Furukawa and M. Honjo, *ibid.*, **16**, 2286 (1968).

(16) A. Vilsmeier and A. Haack, *Chem. Ber.*, **60**, 119 (1927).

(17) H. H. Bosshard, R. Mory, M. Schmid, and H. Zollinger, *Helv. Chim. Acta*, **42**, 1653 (1959).

(18) H. H. Bosshard and H. Zollinger, *ibid.*, **42**, 1659 (1959).

(19) L. F. Fieser and M. Fieser, "Reagents for Organic Synthesis," Wiley, New York, N. Y., 1967, p 284.

(20) K. Kikugawa and T. Kawashima, *Chem. Pharm. Bull.*, **19**, 2627 (1971).