1.45 g. of 1-benzoyl-2-(*p*-dimethylaminophenyl)-1,2,3,4-tetrahydroquinoline (IX) and 5.0 g. of sodium hydroxide in 40 cc. of 70% ethanol was refluxed for 15 hours, most of the suspended material going into solution. An equal volume of water was added, and the ethanol distilled. The mixture was extracted with ether and the ether solution dried over anhydrous potassium carbonate. After distillation of the ether, the residue was crystallized from absolute ethanol, colorless rosettes of 2-(*p*-dimethylaminophenyl)-1,2,3,4-tetrahydroquinoline (X) being obtained, 0.83 g. (76%), m.p. 100–103°. Completely purified material melted at 107.5–108.5°, also in admixture with an authentic sample of the compound (see below).

Anal. Calcd. for $C_{17}H_{20}ON_2$: C, 80.91; H, 7.99; N, 11.10. Found: C, 80.99; H, 8.09; N, 11.52.

Acidification of the basic layer, followed by ether extraction, afforded 0.46 g. (92%) of benzoic acid.

Catalytic Reduction of 2-(*p*-Dimethylaminophenyl)-quinoline (VII).—A suspension of 1.00 g. of 2-(*p*-dimethylaminophenyl)-quinoline (VII) and 0.10 g. of platinum oxide catalyst in 65 cc. of absolute ethanol containing 10 cc. of glacial acetic acid was hydrogenated at atmospheric pressure until the theoretical volume of hydrogen corresponding to the reduction of two double bonds had been absorbed. The suspended material went into solution during the reduction. The solution was filtered, made alkaline with sodium hydroxide solution, and the ethanol was distilled *in vacuo*. The residue was taken up in ether and dried over Drierite. On evaporation of the ether, 0.80 g. of solid remained, m.p. 100–103°. On crystallization from absolute ethanol, 0.54 g. (53%) of 2-(*p*-dimethylaminophenyl)-1,2,3,4-tetrahydroquinoline (X), colorless needles, m.p. 103–105°, was obtained. An additional recrystallization gave material of m.p. 107–108°.

Acid Hydrolysis of 1-Benzoyl-2-(*p*-dimethylaminophenyl)-1,2-dihydroquinoline (VI).—A solution of 5.00 g. of VI in 150 cc. of 9 *M* sulfuric acid was steam distilled. Benzoic acid began to appear in the distillate almost immediately. Altogether, 1.60 g. (93%) of benzoic acid was collected. There was no evidence of benzaldehyde.

The acid solution remaining in the distilling flask was made basic by the addition of sodium hydroxide solution. A red solid precipitated, 4.40 g. It resisted all attempts at purification.

LAWRENCE, KANSAS

[CONTRIBUTION FROM THE DEPARTMENT OF AGRICULTURAL CHEMISTRY, PURDUE UNIVERSITY]

Isolation and Properties of a Series of Crystalline Oligosaccharides from Xylan[1,2]

BY ROY L. WHISTLER AND CHEN-CHUAN TU

RECEIVED JANUARY 7, 1952

A polymer homologous series of crystalline oligosaccharides composed only of D-xylose units has been isolated and characterized. The series extends from the dimer to the hexamer inclusive. Members of the series have been obtained through partial acid hydrolysis of the xylan from corncob holocellulose. Evidence suggests that these oligosaccharides are linear chains composed of β-D-xylopyranoside units linked 1 → 4.

A homologous series of oligosaccharides identified and described as xylobiose, xylotriose, xylotetraose, xylopentaose and xylohexaose has now been isolated in crystalline form. These represent the first obtained crystalline oligosaccharides which are composed only of pentose sugar units. They also represent the second crystalline homologous series of oligosaccharides to be isolated. The first oligosaccharide series examined in detail was that obtained by the graded hydrolysis or acetolysis of cellulose.[3]

Conclusive evidence is submitted that the ring structure and configuration of the members of the new homologous series are the same as those of the cellulose series. The latter differ quantitatively by having a projecting $-CH_2OH$ group on each ring. A comparison of the properties of the two homologous series might yield significant data on the influence of the primary alcohol group on the molecular behavior of polysaccharides.

Preparation of the oligosaccharides in the present homologous series is accomplished by applying the Whistler and Durso charcoal chromatographic procedure[4] to a mixture of oligosaccharides obtained by the partial hydrolysis of xylan derived from corncob holocellulose. Kuhn[5] has calculated the yields of various fragments to be expected on the partial depolymerization of a uniform polymer. Using these data as rough approximations in the case of xylan it might be presumed that a maximum yield of disaccharide would be obtained by stopping the hydrolysis at two-thirds completion, and a maximum yield of trisaccharide could be expected by stopping the hydrolysis at one-half completion. When this is done, good yields of xylobiose and xylotriose are obtained.[6] Besides these two sugars there are present in the hydrolysis mixture other fragments of the xylan molecule and these are also separable by the charcoal chromatographic procedure. The entire series of oligosaccharides up to and including the hexasaccharide are thus prepared and are found to crystallize rather easily.

Each of the xyloöligosaccharides possesses a sharp melting point and produces a crystalline pattern on examination with X-rays. Comparison of the X-ray patterns, however, suggests that a slight decrease in the degree of crystallinity may occur with increase in molecular weight of the oligosaccharide. Each successive higher member of the series produces a pattern with fewer lines. Thus, while xylobiose produces twenty easily recognized lines on a flat film 5 cm. distant, xylohexaose produces but six lines. Although none of the patterns examined here have a detectable amorphous halo, it might be expected that at some higher molecular weight amorphous material would be present in sufficient extent to be evident by a

(1) Journal Paper No. 593 of the Purdue Agricultural Experiment Station.

(2) Paper presented before the XIIth International Congress of Pure and Applied Chemistry, New York, September, 1951.

(3) L. Zechmeister and G. Tóth, *Ber.*, **64B**, 854 (1931); R. Willstätter and L. Zechmeister, *ibid.*, **46**, 2401 (1913); E. E. Dickey and M. L. Wolfrom, THIS JOURNAL, **71**, 825 (1949).

(4) R. L. Whistler and D. F. Durso, THIS JOURNAL, **72**, 677 (1950).

(5) W. Kuhn, *Ber.*, **63B**, 1503 (1930).

(6) R. L. Whistler and C. C. Tu, THIS JOURNAL, **73**, 1389 (1951).

halo on the film much as is observed when xylan is analyzed by X-rays.

The structure of the crystalline xylobiose is demonstrated by establishing that its hexaacetate is identical with the crystalline 4-(β-D-xylopyranosyl)-β-D-xylose hexaacetate reported by Whistler, Bachrach and Tu.[7] In aqueous solution the free sugar mutarotates in the positive direction. Thus the structure proposed for the disaccharide is 4-(β-D-xylopyranosyl)-, -D-xylopyranose.

The degree of polymerization of the other isolated oligosaccharides is indicated by iodometric titration of their reducing groups and by iodometric titration after hydrolysis which yields the value expected from the assumption of complete hydrolysis to D-xylose. The structure of xylotriose and the higher homologs is shown to be that of 1→4-linked D-xylopyranose units, probably in the β-configuration. This evidence is based on hydrolysis data, on results of periodate oxidation, and on previous proof of the structure of xylobiose. When the oligosaccharides are slowly hydrolyzed, periodic samples taken during the hydrolysis show on paper chromatograms, only lower members of the oligosaccharide series. As hydrolysis proceeds, all oligosaccharide spots gradually disappear in inverse order of their D. P., finally leaving only a D-xylose spot on the chromatograms. For example, xylotriose on hydrolysis shows xylobiose and D-xylose. Eventually the xylotriose spot disappears followed by disappearance of the xylobiose spot.

The periodate oxidation data are consistent with a 1→4-glycosidic link between pentose units, if the links in the higher oligosaccharides are uniform. Since the pyranose ring form has been proven to exist in xylobiose[7] it may be assumed in the higher homologs. Methylation work on xylan[8] has also demonstrated the existence of the pyranose ring in the polymer.

Uniformity of linkage has been shown[9] in the series of acetates derived from these oligosaccharides by direct acetylationa nd hence must be assumed to exist in the unacetylated oligosaccharides also. In addition, when the oligosaccharides are hydrolyzed and periodic examination of hydrolysate is made, only one disaccharide spot has ever been found by paper chromatography. If the linkages of xylohexaose, for example, were not uniform, there would be formed on hydrolysis at least one other disaccharide in more than trace amounts.

The present work also demonstrates the ease with which an homologous oligosaccharide series may be obtained from the parent polymer using the methods of large-scale chromatography.

Experimental

Partial Hydrolysis of Xylan.—The xylan used in these experiments was the reprecipitated A-fraction obtained from corncob holocellulose.[10]

Thirty grams was hydrolyzed at a time. The xylan was dissolved in 1.5 l. of fuming hydrochloric acid (d^{16}_4 1.21) at −16°, 30 min. sufficing. The hydrolysis conditions were similar to those used by Willstätter and Zechmeister[3,11] for cellulose. The hydrolysis was allowed to proceed at 0° and was stopped either at two-thirds completion or at one-half completion, the end-point being determined by observation of the optical rotation. The longer hydrolysis period was employed for isolation of xylobiose, and the shorter hydrolysis period employed where higher oligosaccharides were the prime objects for isolation. To stop the hydrolysis, the solutions were neutralized with sodium bicarbonate by stirring in the presence of ice. About 2 ml. of *n*-octyl alcohol was added to prevent frothing. To facilitate removal of salts the neutral solution (about 4 l.) was stored at 5° for about 3 days. The supernatant solution was then decanted and filtered. Three similarly prepared filtrates were then combined for chromatographic separation of the sugars.

Chromatographic Separation of Oligosaccharides.—The above filtrates (12 l.) were each chromatographically separated on charcoal–Celite (2:3) columns (75 × 850 mm.) following the method of Whistler and Durso[4] and using the automatic apparatus of Durso, Schall and Whistler.[12]

A typical procedure follows: After a hydrolysate was passed through a column, the column was washed with 60 l. of water to remove salts and D-xylose. Then the column was washed successively with five 8-l. portions of 5% ethanol, each portion being kept separate. Next the column was washed successively 5 times with 8-l. portions of 15% ethanol and finally with two 8-l. portions of 50% ethanol. All effluent wash solutions were separately concentrated to a dry powder and the number of sugars in each roughly determined by chromatography on paper strips. The strips were developed downward for 22–25 hours with a mixture of water–pyridine–butanol-1 in the ratio 3:4:6, except for the higher oligosaccharides which required a 3:5:5 ratio and longer times (*ca.* 90 hours). The locations of the sugars were determined by spraying the strip with Tollens reagent, drying, and developing the color by heating to 100° for 2 minutes in an oven. The yield of sugar mixture in each portion of effluent from the charcoal column and the various sugars present as indicated by paper chromatography are shown in Table I.

TABLE I

SUGAR YIELDS AND COMPOSITIONS OF ELUATES FROM A CHARCOAL COLUMN OF XYLAN HYDROLYSATE

Eluate in 8-l. portions	Dry sugar yield, %	Sugars present
5% ethanol 1	6.1	X_1, X_2
2	0.5	X_2
3	1.8	X_2, X_3
4	2.0	X_2, X_3
5	2.4	X_2, X_3
15% ethanol 1	9.1	X_2, X_3, X_4
2	5.0	X_4, X_5
3	1.8	X_3, X_4, X_5
4	1.1	X_4, X_5
5	0.6	X_4, X_5
50% ethanol 1	7.2	X_4, X_5
2	6.1	X_5, X_6

Here X_1, X_2, X_3, etc., represent D-xylose, xylobiose, xylotriose and higher oligosaccharides composed of D-xylose units. At times traces of D-xylose may occur in subsequently removed oligosaccharide fractions. It was noted that if instead of five 8-l. portions of 5% ethanol, the column was washed with but three portions and then with 15% ethanol, the first 8-l. portion of 15% ethanol contained only xylotriose in approximately 10% yield.

Chromatographic Purification.—The oligosaccharide fractions shown in Table I were further purified by rechromatography from 10% aqueous solution on 49 × 650 mm. or 75 × 850 mm. charcoal–Celite (1:1) columns.

Xylobiose.—Dry powder from suitable (*e.g.*, 1st and 2nd) 5% ethanol effluents was placed on the charcoal column by

(7) R. L. Whistler, J. Bachrach and C. C. Tu, THIS JOURNAL, **74**, 3059 (1952).

(8) W. N. Haworth and E. G. V. Percival, *J. Chem. Soc.*, 2850 (1931).

(9) R. L. Whistler and C. C. Tu, THIS JOURNAL, **74**, 3059 (1952).

(10) R. L. Whistler, D. R. Bowman and J. Bachrach, *Arch. Biochem.*, **19**, 25 (1948).

(11) R. Willstätter and L. Zechmeister, *Ber.*, **62**, 722 (1929).

(12) D. F. Durso, E. D. Schall and R. L. Whistler, *Anal. Chem.*, **23**, 425 (1951).

TABLE II

CHARACTERISTICS OF XYLOSE AND SOME OLIGOSACCHARIDES FROM XYLAN

Sugar	$[\alpha]^{25}D$ in water	M.p., °C.	Iodometric equivalent weight: Before hydrolysis Found	Before hydrolysis Calcd.	After hydrolysis Found	After hydrolysis Calcd.	Yield, % based on xylan
Xylose	18.8[a]	145[a]		75			
Xylobiose	−32.0 → −25.5	185–186	146	141	72	75	5
Xylotriose	−39.4 → −47.0	205–206	204	207	72	75	8
Xylotetraose	−48.80 → −60.0	219–220	270	273	73	75	4
Xylopentaose·$^1/_2H_2O$	66.0[b]	231–232	345	339	74	75	3
Xylohexaose·$2H_2O$	72.8[b]	236–237	403	405	74	75	2

[a] Indicates accepted values. [b] Indicates no mutarotation observed.

TABLE III

DATA OBTAINED FROM PERIODATE OXIDATION

	Quantity used, g.	Formic acid formed: Moles per mole of sugar: 46	Found after 166 hours	348	Calcd.	Periodate consumed: Moles per mole of sugar: 46	Found after 166 hours	348	Calcd.
Xylobiose	0.0223	2.1	2.4	2.8	3	3.2	3.6	4.1	4
Xylotriose	.0296	2.1	2.5	2.9	3	3.9	4.7	4.9	5
Xylotetraose	.0321	2.2	2.4	2.8	3	4.5	5.4	5.9	6
Xylopentaose	.0477	2.2	2.4	2.8	3	4.6	5.9	6.8	7
Xylohexaose	.0578	2.1	2.4	2.9	3	5.0	6.8	7.8	8

absorption from 10% aqueous solution. After being washed with 15 l. of water, the column was washed with 8 l. of 5% ethanol to remove the xylobiose. On evaporation of the solution the residual sirup gave only one spot on paper chromatography.

Xylotriose.—This sugar was generally isolated in pure form from the 15% ethanol effluent described in the third paragraph above, but was also obtained from the mixture of Table I by rechromatography of the combined 3rd, 4th and 5th fractions of 5% ethanol effluent and the 1st fraction of the 15th ethanol effluent. The mixture on the column was washed with 15 l. of water and with 18 l. of 2% ethanol, all of which were discarded. The column was then washed with 36 l. of 9% ethanol, the last two-thirds of which contained only xylotriose.

Xylotetraose.—Dry powder from fraction 2 (Table I) of the 15% ethanol effluent placed on a charcoal column from water as described was washed with 15 l. of water, 16 l. of 2% ethanol and 8 l. of 15% ethanol and the effluents discarded. The next 15 l. of 15% ethanol removed only xylotetraose, as indicated by paper chromatography of the sirup obtained by evaporation.

For isolation of **xylopentaose and xylohexaose**, fractions 1 and 2 of the 50% ethanol effluent of Table I were combined, dissolved in water and sorbed on a 49 × 650 mm. charcoal–Celite (1:1) column as usual. The column was washed with 4 l. of water, 8 l. of 15% ethanol and 2 l. of 20% ethanol and the effluents discarded. The next 6 l. of 20% ethanol removed only xylopentaose. Subsequently 2 l. of 30% ethanol removed a mixture of penta- and hexasaccharide but the next 4 l. of 30% ethanol removed only xylohexaose.

Crystallization of Oligosaccharides.—Chromatographically pure dry powders from the above separations were the source materials for crystallization. Characteristics of the crystalline preparations are listed in Table II.

Xylobiose.—Two grams of dry powder was dissolved in 1 to 2 ml. of water, 50 ml. of methanol at 60° added and the mixture was allowed to stand for about 5 minutes until there precipitated a small amount of flocculent inorganic matter which seems to arise from the Celite. The solution was filtered warm, then held at 5° overnight. The crystals formed were filtered and recrystallized from methanol in the same manner.

Anal. Calcd.: C, 42.53; H, 6.42. Found: C, 42.5; H, 6.4.

Xylotriose.—Eight grams of dry powder was dissolved in 28 ml. of water, 35 ml. of absolute ethanol added and the solution warmed for 5 minutes, filtered to remove the flocculent precipitate, and the ethanol concentration of the hot solution was brought to 85%. Crystals formed on cooling and were separated and purified by recrystallization from hot 85% ethanol.

Anal. Calcd.: C, 43.47; H, 6.30. Found: C, 43.4; H, 6.4.

Xylotetraose.—One gram of dry powder was dissolved in 5 ml. of water and methanol added to a final concentration of 85%. The solution was warmed and filtered, and 23 ml. of hot butanol-1 added. Crystallization took place from the hot solution within 2 to 5 minutes. The separated crystals were recrystallized in the same way.

Anal. Calcd.: C, 43.95; H, 6.27. Found: C, 43.8; H, 6.3.

Xylopentaose.—Two grams of dry powder was dissolved in 50 ml. of water and 20 ml. of absolute ethanol added. The mixture was warmed to 60°, filtered and hot absolute ethanol added to a concentration of 80%. On very gradual cooling for 5 minutes crystallization occurred. Recrystallization was effected in the same way.

Anal. Calcd. for $C_{25}H_{42}O_{21}\cdot{}^1/_2H_2O$: C, 43.80; H, 6.2. Found: C, 43.8; H, 6.2.

Xylohexaose.—About 1.5 g. of dry powder was dissolved in 10 ml. of water and 22 ml. of absolute ethanol added. After warming to 60° and filtering, 18 ml. of absolute ethanol was added, and crystallization occurred on cooling gradually to room temperature overnight. Recrystallization was done in the same way.

Anal. Calcd. for $C_{30}H_{50}O_{25}\cdot 2H_2O$: C, 42.62; H, 6.38; H_2O, 4.25. Found: C, 42.7; H, 6.5; H_2O, 4.1.

Equivalent Weight of Crystalline Oligosaccharides.—The sample (0.22–0.25 milliequivalent weight of each sugar) was weighed into a 250-ml. erlenmeyer flask and dissolved in 1 ml. of water. Seven and one-half ml. of iodine solution (0.0500 *N*) and 25 ml. of buffer solution (*p*H 11.2, made by dissolving 1.2 g. of sodium hydroxide and 7.1 g. of disodium hydrogen phosphate in 850 ml. of water) were added dropwise simultaneously into the flask while the solution was swirled. The solution was allowed to stand for 20 minutes. Fifty ml. of 3% sulfuric acid was then added, and the resulting solution was titrated immediately with standard thiosulfate solution (0.0100 *N*) with 2 ml. of 1% starch solution as indicator. A blank titration was made under the conditions of each sugar determination. The results are shown in Table II.

Hydrolysis of Oligosaccharides.—Each oligosaccharide (0.3000 g.) was dissolved in 0.05 *N* HCl in a 10-ml. volumetric flask to make a 3% solution. A portion of the solution was hydrolyzed at 99 ± 1° and the change in rotation noted periodically.

At intervals 0.1-ml. (3 mg.) portions of the solution were neutralized with sodium bicarbonate and placed on a paper strip for chromatographic development with water–pyridine–butanol-1 (3:4:6 ratio). In each case the developed chromatograms showed the presence of only lower oligosac-

charides and D-xylose with the eventual disappearance of one after another of the oligosaccharides until the presence of only D-xylose was indicated. The final optical rotation of the solution also was that calculated for D-xylose alone.

Another aliquot of each oligosaccharide solution was hydrolyzed completely, and the solution was then neutralized with 0.05 *N* sodium hydroxide and titrated iodometrically as described above. The equivalent weights of the hydrolysates are reported in Table II.

Periodate Oxidation.—A sample sufficient to produce about 10 mg. of formic acid was weighed into a 500-ml. stoppered bottle and dissolved in distilled water. Following addition by pipet of 100 ml. of 5% potassium chloride solution and 20 ml. of 0.3 *M* sodium periodate, the reaction was allowed to proceed in the dark at 15° with shaking. A pair of 10-ml. aliquots were withdrawn at intervals and filtered through coarse sintered glass. The first 10-ml. portion was used to determine the formic acid produced and to determine the total amount of iodine in the form of iodate.[13] The second portion was used for the determination of the excess periodate.[14] The amount of periodate consumed by the sugar was found by subtracting the periodate remaining from the iodate produced.

(13) F. Brown, S. Dunstan, T. G. Halsall, E. L. Hirst and J. K. N. Jones, *Nature*, **156**, 285 (1945); T. G. Halsall, E. L. Hirst and J. K. N. Jones, *J. Chem. Soc.*, 1427 (1947).

(14) Fleury and Lange, *J. pharm. chim.*, **17**, 107, 196 (1933).

The periodate oxidation data are shown in Table III. The calculated values are based on pyranose rings linked 1,4'.

Osazones. Phenylxylobiosazone.—Five-tenths gram of the crystalline xylobiose, 1 g. of phenylhydrazine hydrochloride, 1.5 g. of sodium acetate pentahydrate and 12 ml. of distilled water were mixed in a 50-ml. erlenmeyer flask. The flask was loosely stoppered and placed in boiling water. After 20 minutes with occasional shaking, to avoid supersaturation, 0.25 g. of yellow needle-like crystals formed. They were twice recrystallized from either 60% ethanol or a mixture of 1,4-dioxane and petroleum ether (b.p. 65–75°), m.p. 195–196° dec., $[\alpha]^{22}D$ −22.5 → −77.0 (33 hr., *c* 0.65 in a 7:3 mixture of pyridine and absolute ethanol).

Anal. Calcd. for $C_{22}H_{28}O_7N_4 \cdot H_2O$: N, 11.71. Found: N, 11.5.

Phenylxylotriosazone.—Xylotriose treated in the same way for osazone formation gave yellow crystalline needles after 25 minutes of heating and on slight cooling of the solution, yield 0.23 g.

The material was twice recrystallized from 95% ethanol, m.p., 214–215° dec., $[\alpha]^{25}D$ → −53.5 (12 hr., *c* 0.41 in a 7:3 mixture of pyridine and absolute ethanol).

Anal. Calcd. for $C_{27}H_{36}O_{11}N_4 \cdot H_2O$: N, 9.17. Found: N, 9.2.

LAFAYETTE, INDIANA

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE OHIO STATE UNIVERSITY]

The Structure of Maltotriose[1]

BY A. THOMPSON[2] AND M. L. WOLFROM

RECEIVED FEBRUARY 18, 1952

An improved method for the isolation of maltotriose as its hendecaacetate is presented. The method for proving the structure of a trisaccharide, first established with panose, has now been employed to confirm the structure of maltotriose as assigned by Sugihara and Wolfrom by other methods. The acetylated partial hydrolyzate of the alditol was resolved by silicate column chromatography and the components β-D-glucose pentaacetate, D-glucitol (sorbitol) hexaacetate, β-maltose octaacetate and maltitol nonaacetate were adequately identified in the crystalline state. This definitely confirms the structure 4-α-maltopyranosyl-D-glucose (I) for maltotriose.

Maltotriose[3] merits the interest of chemists in the carbohydrate and allied fields because it is one of the hydrolytic products of starch. It is therefore highly desirable to find better methods for preparing this sugar in a pure condition. We wish to describe herein a method for separating maltotriose from complex carbohydrate mixtures in which it may occur. The method used is in part that of Whistler and Durso[4] for separating sugars by means of a carbon column. The maltotriose in a commercial hydrolyzate of starch can be concentrated considerably by this means. When the aqueous solution of the complex carbohydrate mixture is placed on a carbon column, the monosaccharides are not adsorbed. Oligosaccharides are then removed by developing with successively higher concentrations of ethanol in water. Pure crystalline β-maltotriose hendecaacetate can then be prepared by means of silicate column chromatography of the acetylated concentrate.

Previously, Wolfrom, Thompson and Galkowski[5] used a method for the determination of the structure of trisaccharides which involved marking the monosaccharide unit containing the reducing group by hydrogenation to the corresponding alcohol (alditol), followed by partial hydrolysis with separation and identification of the acetylated fragments in the hydrolyzate by means of silicate column chromatography. This method in principle was also employed by French[6] in a structural study of panose. We wish to report herein the extension of this technique in confirming the structure of maltotriose which was first established, in a not entirely unequivocal manner, by Sugihara and Wolfrom,[7] through the employment of methylation procedures and enzymic partial hydrolysis.

It is advantageous to determine the conditions of hydrolysis which will produce the maximum quantities of disaccharide material. In the hydrolysis of panitol, in which two kinds of linkages exist, the reaction time *t* to bring about hydrolysis to maximum quantities of disaccharide material was calculated[5] to be

$$t = \frac{\log[(k_1 + k_2)/k_2]}{k_1} \qquad (1)$$

when the reaction was carried out with a sugar concentration of 2% in a 0.05 *N* sulfuric acid solution

(1) Reported in *Abstracts Papers Am. Chem. Soc.*, **121**, 5P (1952).

(2) Corn Industries Research Foundation Associate of The Ohio State University Research Foundation (Project 203).

(3) M. L. Wolfrom, L. W. Georges, A. Thompson and I. L. Miller, THIS JOURNAL, **71**, 2873 (1949); L. W. Georges, I. L. Miller and M. L. Wolfrom, *ibid.*, **69**, 473 (1947).

(4) R. L. Whistler and D. F. Durso, *ibid.*, **72**, 677 (1950).

(5) M. L. Wolfrom, A. Thompson and T. T. Galkowski, *ibid.*, **73**, 4093 (1951).

(6) D. French, *Science*, **113**, 352 (1951).

(7) J. M. Sugihara and M. L. Wolfrom, *ibid.*, **71**, 3357 (1949).