CXCIV.—*The Constitution of Polysaccharides. Part IX. The Degradation of Cellulose to an Anhydrotrisaccharide.*

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In former papers from this laboratory it was shown that cotton cellulose is composed of 1 : 5-anhydroglucose residues * and the question was discussed as to the number of such residues present in the unpolymerised molecule of the polysaccharide. It was

* See Appendix for discussion and revision of numerical nomenclature as applied throughout this paper.

pointed out (Irvine and Hirst, J., 1923, **123**, 518) that the simplest view which satisfies the primary conditions is that cellulose is a polymerised di(1 : 5-anhydroglucose) (*loc. cit.*, p. 524, Formula IV) but that a formula of this type is at variance with the highest authentic yield of cellobiose octa-acetate so far obtained from a normal cellulose. In order to accommodate this result and yet preserve the greatest simplicity of structure, it was suggested that cellulose may be regarded as a polymeride of tri-1 : 5-anhydroglucose. The suggestion was tentative, as shown by the following significant extract from the argument :—"*in the meantime it is prudent to select a formula for cellulose which will give a yield of cellobiose approximating to this figure* (50—60%) *rather than to the higher value* 105·5." The same reasonable caution has been exercised in our other references to this question, but nevertheless the views expressed have been generally invested with a rigidity and finality for which no claim was made. The present position is that the exact number of anhydroglucose residues which form the cellulose molecule remains unknown and speculation regarding this factor must be guided from time to time as knowledge accumulates. Recently, *X*-ray spectrographic methods have been brought to bear on the problem and the results are interpreted as indicating that the C_6 residues are marshalled in even numbers. This conclusion is opposed to the idea that the molecular unit is an anhydro-trisaccharide, but the results of standard chemical methods of attacking constitutional questions must equally be taken into account, even if, for the time being, they conflict with data obtained by purely physical processes. Obviously, the anhydro-trisaccharide formula need no longer be considered if convincing chemical evidence is forthcoming that the cellulose molecule is of a single type and definitely contains an even number of C_6 residues. But it will be generally agreed that the acceptance of any particular molecular formula must rest ultimately on the depolymerisation of cellulose to the simplest non-reducing compound possessing the empirical formula $C_6H_{10}O_5$ and retaining the 1 : 5-anhydro-linking. Further, such a compound should be capable of undergoing the reverse change of polymerisation without disturbance of the characteristic 1 : 5-anhydride ring and without forming complex glucosides of the type produced by the polymerisation of glucosan (Irvine and Oldham, J., 1925, **127**, 2903). This prospect is still remote, but we now submit experimental evidence, bearing on the problem, which has been obtained by studying the graded acetolysis of cotton cellulose.

A review of the scattered literature and more particularly of papers which have appeared in the past few years reveals that acetolysis is an extremely complex process and is not confined to a

succession of reactions conducted on cellulose triacetate. Much recent work rightly emphasises the esterifying effect of the sulphuric acid employed in acetolysis, but nevertheless the earlier researches of Klein and of Schliemann reveal the essential nature of the changes involved. These workers showed that even when the yield of cellulose octa-acetate is as much as 60% of the cellulose used, degradation products persist which may be regarded as simple dextrins and possess a greater complexity than a disaccharide. These dextrin acetates are dextrorotatory and appear to be the immediate precursors of cellobiose octa-acetate, so that acetolysis may be regarded as a series of ill-defined steps as represented below. The scheme does not include the formation of aceto-sulphates as, from the point of view of the present investigation, these products arise from secondary reactions, and as cellulose has been completely converted into 2 : 3 : 6-trimethyl glucose, it is also unnecessary to make separate provision for *iso*cellobiose acetate.

Cellulose → Acetylated cellulose → Acetylated cellulose dextrins → [X] → Cellobiose octa-acetate → Glucose penta-acetate.

Acetylation and depolymerisation are the essential features of the earlier reactions, and the opening of anhydro-rings by hydrolysis is specially characteristic of the two final stages, but there is no sharp line of demarkation between these different types of change. The hypothetical stage indexed as [X] therefore represents the formation of compounds in which the maximum depolymerisation of cellulose has taken place, whilst the specific hydrolytic action responsible for the formation of cellobiose octa-acetate is a minimum. The present investigation deals with the exploration of this group of acetolysis products.

Several workers, notably Bertrand, Hess, and Pringsheim, have occupied themselves with what is essentially the same subject of research and have obtained results of great interest. It is, however, a difficult matter to define exactly the experimental conditions under which acetolysis can be controlled so as to give consistent results. The physical condition of the cellulose used and the treatment to which it has been subjected, minute variations in the purity of the reagents employed, the temperature and other factors still unrecognised, combine to affect the series of reactions profoundly. In consequence, through no fault of the observers, the statements in the literature are occasionally conflicting. We find, however, that the progress of acetolysis can be ascertained by systematic physical examination of samples of the product and that it is possible to arrest the reactions at a stage immediately before cellobiose octa-acetate is formed and when insoluble dextrins are

present in minimum amount. This result was secured by working between two limiting conditions, the first being that no trace of crystalline structure should be observable in the solid product when examined by ordinary microscopic methods. Control experiments showed that as small a proportion as 3% of cellobiose octa-acetate can be identified with certainty in this way. The second limiting condition was that a solution of the product in chloroform should be dextrorotatory and should show only a faint cone of light when viewed in the ultramicroscope. This secured that insoluble dextrins constituted less than 7% of the total. The product obtained when these conditions were preserved was a white amorphous powder and the composition was that of a cellulose triacetate, but its properties presented a sharp contrast with those of a normal acetate of the polysaccharide. In particular, it displayed a much wider range of true solubility, possessed a distinct, although indefinite, melting point, and was dextrorotatory. The change of sign from the lævorotation characteristic of cellulose triacetate is in itself proof that extensive depolymerisation had taken place. Despite the uniformity in properties and composition displayed by different preparations, the product was a mixture and contained at least two constituents which reduced Fehling's solution, the ratio of the cuprous oxide formed before and after acid hydrolysis being 1 : 2·2. The greatest caution must be exercised in interpreting all analytical data in the case of acetolysis products and consequently the nature of the degraded acetate was ascertained by submitting it to (1) deacetylation and (2) methylation. Treatment with aqueous dimethylamine eliminated the acetyl groups and enabled dextrins to be removed. The main product thus obtained was freely soluble in water but, owing to secondary reactions between reducing groups and the alkaline reagent (Irvine, Thomson, and Garrett, J., 1913, **103**, 238), the material was contaminated with alkylamino-derivatives and could not be completely separated into its constituents. This section of our work is being continued, but it may be stated that we have isolated from the above mixture an amorphous disaccharide which was present to the extent of about 20%. The sugar melted with decomposition at 180°, and had $[\alpha]_D + 14°$ in water, but until this extension of the research is complete it is impossible to characterise the compound definitely as an *iso*cellobiose.

These experimental difficulties were overcome by submitting the mixture of degradation compounds to the methylation process, which was adjusted so as to eliminate both alkylated disaccharides and dextrins. Contrasted with the behaviour of cellulose under parallel conditions, the methylation proceeded with remarkable smoothness and no difficulty was experienced in obtaining the fully

substituted derivative, a result which supplies additional evidence that extensive depolymerisation of the polysaccharide had been accomplished. As only the material insoluble in sodium hydroxide was collected, any disaccharide or monosaccharide constituents were retained in the alkaline liquors and were thus eliminated. Methylated dextrins, amounting to 6% of the total weight, were left undissolved on treatment with ether, so that, so far as molecular complexity is concerned, the product finally isolated was intermediate between lower dextrins and disaccharides. In this way, a 50% yield of a white, amorphous powder was obtained which was readily soluble in organic solvents, and possessed the properties of a glucoside. Molecular-weight determinations by the cryoscopic method in benzene solution, and confirmed by Rast's process, gave the value 656, showing that the substance was derived from a trisaccharide. The analytical composition also corresponded exactly with that required for $[C_6H_7O_2(OMe)_3]_3$ and the combined results might well have been accepted at this stage as conclusive. The behaviour on hydrolysis, however, warrants the conclusion that only 70% of the material consisted of tri(trimethyl anhydroglucose), the remainder being the corresponding methylated trisaccharide. This evidence was obtained by heating with acid methyl alcohol, which effected hydrolysis and the condensation of the liberated sugars with the solvent. The product of this reaction was isolated by vacuum distillation and consisted of a mixture of trimethyl methylglucoside (90%) with tetramethyl methylglucoside (10%). This opinion was verified in two ways. The mixture of glucosides was hydrolysed and the sugars thus obtained consisted exclusively of 2 : 3 : 6-trimethyl glucose and 2 : 3 : 5 : 6-tetramethyl glucose, both of which were isolated in crystalline form in yields which correspond with the above composition. Further, precisely the same result was obtained when the original methylated product was hydrolysed by means of aqueous acid to give the corresponding sugars directly without the intermediate formation of the glucosides. The combined evidence leads to the opinion that, under the conditions specified, cellulose can be degraded to a mixture of acetates derived from the following compounds in the proportions stated :—

1. Dextrins	6%	3. Triglucose	15%
2. Anhydro-triglucose	35%	4. Diglucose	20%

Although the results clearly favour the idea that anhydro-triglucose forms part of the cellulose aggregate, the possibility remains that anhydro-diglucose molecules may also be concerned in the polymerisation which leads to the polysaccharide. This possibility has been foreshadowed by the work of Irvine and Oldham (*loc. cit.*), who showed that the polymerisation of β-glucosan

involves both odd and even numbers of the monomeric unit, and is emphasised by consideration of the yields of the various products obtained in the present research. Although our work was carried out under conditions as nearly as possible quantitative, only 76% of the degraded acetates was accounted for in the form of pure derivatives. Further, during the original acetolysis, about 30% of the cellulose was converted into soluble or volatile products. Making allowance for these factors, it follows that the yield of trisaccharide derivatives actually obtained by us is of the order 35% when referred to the weight of cellulose originally employed. This is a minimum value and there seems no reason for doubt that at least one-third of the polysaccharide is based upon the triglucose unit. The remaining two-thirds of the polymerised aggregate remains unknown and the research is consequently being continued on the following lines: (1) the constitutional study of the tri- and di-saccharide products of graded acetolysis, (2) the polymerisation of these products and (3) the quantitative relationship between the tri- and di-saccharides.

Discussion of Results.

The views expressed in the present paper depend on the conversion of cellulose into simple methylated compounds, one of which is derived from triglucose and one from the corresponding anhydro-triglucose. Although it was possible to separate these two constituents from other compounds, it proved impossible to separate them from each other and thus the conclusions are in part based upon analytical figures. This in itself would be insufficient, but although there is ample supplementary evidence, it is necessary to discuss the results critically and to consider in how far explanations other than that offered are valid. Much depends on the fact that tetramethyl glucose is formed as a scission product when the above methylated mixture is hydrolysed, and the obvious suggestion is that the small amount of this sugar which was isolated may be traced to glucose penta-acetate or to cellobiose octa-acetate present in the original starting material. This possibility is discounted by the fact that the degraded acetates were amorphous, although this observation does not exclude the presence of an *iso*cellobiose acetate. Of greater importance is the technique employed in the methylation which, on the basis of control experiments, was adjusted so as to eliminate both mono- and di-saccharides.

It is therefore evident that the tetramethyl glucose did not originate in glucose or in cellobiose, but in a polyhexose containing more than two C_6 residues. If the methylated material which yielded the tetramethyl glucose consists of a single chemical individual,

it follows that ten C_6 groups are present, but, in such an event, the molecular weight would exceed 2000. If, on the other hand, the material is a mixture, one component must be a fully methylated reducing sugar containing any number of glucose residues from three to nine, the remaining constituent being the corresponding poly(trimethyl anhydroglucose). The number of possible mixtures which satisfy the analytical data is large and in the following table some typical examples are given, all of which possess practically the same composition and would give the results obtained in this investigation.

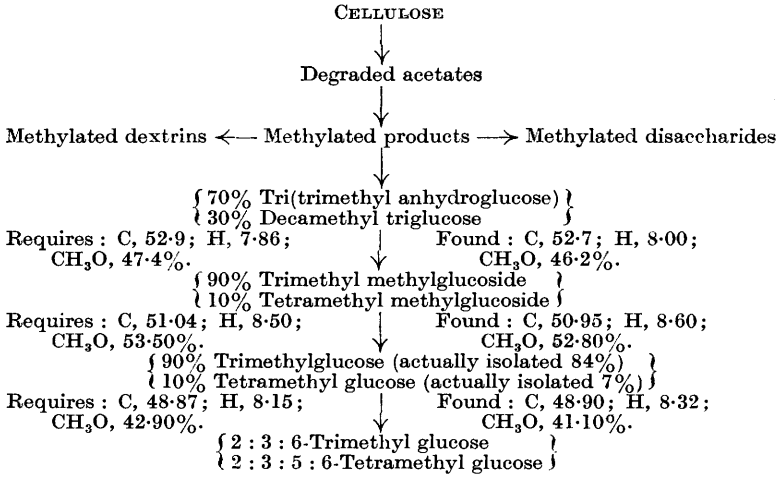
I. A mixture containing 70% of a tri(trimethyl anhydroglucose) and 30% of a methylated trisaccharide. (Molecular weight approximately 625.)

II. A mixture containing 60% of a tetra(trimethyl anhydroglucose) and 40% of a methylated tetrasaccharide. (Molecular weight approximately 834.)

III. A mixture containing 50% of a penta(trimethyl anhydroglucose) and 50% of a methylated pentasaccharide. (Molecular weight approximately 1043.)

IV. A mixture containing 40% of a hexa(trimethyl anhydroglucose) and 60% of a methylated hexasaccharide. (Molecular weight approximately 1251)

The list reveals that the molecular-weight determinations are discriminative and, as the value 656 was obtained, lead to a definite decision on the question of molecular magnitude. The consistency of our results is also displayed by the following scheme, which serves to simplify the series of reactions :



Appendix.

Subsequent to the completion of the paper now communicated, the subject of the ring structure of glucose has been re-opened by Charlton, Haworth, and Peat (this vol., p. 89), who bring forward evidence that the stable form of the sugar is an amylene-oxide. That the oxydic ring in glucose may occupy different positions was first experimentally verified by one of us ten years ago, and the necessity to take all the different possibilities into account has repeatedly been emphasised (Irvine, "Some Constitutional Problems in Carbohydrate Chemistry," J., 1923, **123**, 900). For a considerable time it has been recognised by workers in this field that as the γ-oxydic formula for glucose depended on collateral rather than on direct experimental evidence, it might require modification and that to have the position of the oxygen ring settled experimentally would mark an advance in sugar chemistry. We do not commit ourselves at this stage to unqualified acceptance of the modified formula for glucose—the suggestion offered introduces a number of simplifications and is worthy of extended trial—but we defer our opinion until results obtained by the ultimate oxidation of tetramethyl glucose are available.

The formulæ of a number of methylated sugars may, however, be adjusted, *e.g.*, 2 : 3 : 5-trimethyl glucose would be described as the 2 : 3 : 4-form, but from the point of view of the present research greater importance is attached to the sugar known as 2 : 3 : 6-trimethyl glucose. A review of the constitutional study of this compound does not justify altering the position of the methyl groups, so that by shifting the ring to the neighbouring carbon atom the formula becomes :

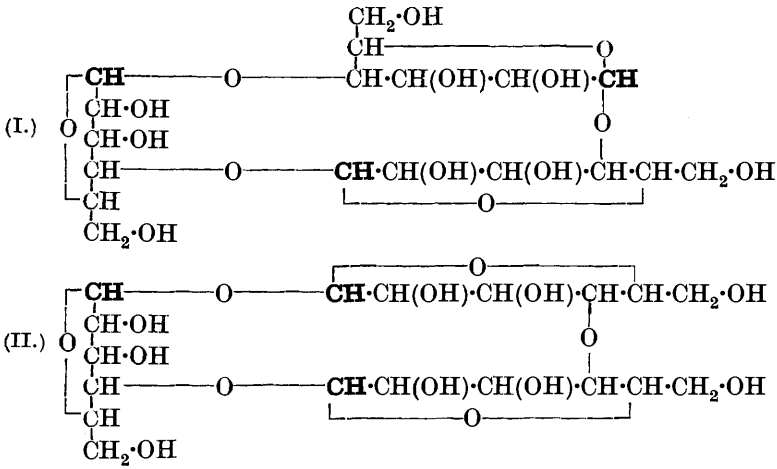
$$\mathrm{OH{\cdot}\overset{\displaystyle\ulcorner\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-\!\!-}{C}H{\cdot}CH(OMe){\cdot}CH(OMe){\cdot}CH(OH){\cdot}CH{\cdot}CH_2{\cdot}OMe.}$$

(with an O ring bridging the first and fifth carbon atoms shown)

The linkage present in cellulose would thus presumably be through positions 1 and 4 in place of 1 and 5 and the polysaccharide would therefore be described as poly-(1 : 4-anhydroglucose). It would, however, be premature to accept this conclusion as final, in view of the fact the particular variety of trimethyl glucose now under consideration can react as a γ-sugar. This possibility has been pointed out by Irvine and Hirst (J., 1922, **121**, 1221) in their study of the condensation of the compound with methyl alcohol, and it follows that the formation of this sugar is not absolutely diagnostic as to the structure of the complex from which it originates. This possibility and its effect on constitutional questions are discussed in a forthcoming paper.

The application of a poly(1 : 4-anhydroglucose) structure to

cellulose may now be considered and the opportunity may be taken to simplify the alternative structural formulæ applicable to this unit, as further consideration has shown that, provided α- and β-configuration is not taken into account, only two possibilities exist.* These as modified are :



The position of the potential reducing groups is indicated in block type.

In view of the results communicated in this paper, and taking into account the increasing importance attached to the formation of *iso*cellobiose from cellulose, it would be inadvisable to exclude formula II entirely. The retention of both types formulated above accounts for the formation of isomeric cellobioses and it is possible that both may function, although unequally, in the polymerisation process which leads to cellulose.

Experimental.

Graded Acetolysis of Cotton Cellulose.—The material employed was provided by Mr. William Rintoul of Messrs. Nobels, Ltd., and was identical with that used in our other investigations (see previous papers). Systematic experiments showed that when the temperature at which acetolysis is conducted is maintained uniformly at 15°, the best results were obtained when the reagent consisted of acetic anhydride and sulphuric acid in the ratio of 5·9 parts to 1 part

* I am indebted to Dr. A. Geake of the Shirley Institute (British Cotton Industry Research Association), who first kindly directed my attention to this simplification.—J. C. I.

by weight. These conditions having been established, the remaining factor, *viz.*, the duration of the reaction, was determined, in the first instance approximately, by microscopic examination of samples of the product. When precipitated by water, dissolved in alcohol, reprecipitated and dried, the material showed a wide range of solubility in organic solvents including benzene. These solutions, which were dextrorotatory, did not give films on evaporation and no trace of crystalline matter was visible in the residue, although control experiments with artificial mixtures showed that 3% of cellobiose octa-acetate can be detected in this way. Further, when dissolved in chloroform, the solution was practically optically clear when viewed in the ultramicroscope, a faint cone of light being, however, visible owing to the persistence of a small proportion of higher dextrins. The control experiments were conducted throughout the entire series of products ranging from a normal cellulose triacetate to cellobiose octa-acetate. A typical preparation is now described, but as the reaction is affected by numerous factors, including the surface condition of the cotton, it is necessary to state that in order to obtain consistent results the acetolysis must be standardised by the physical examination indicated above.

The cellulose (40 g.), dried at 110°, was added gradually to a mixture of 400 c.c. of acetic anhydride and 40 c.c. of sulphuric acid, the pasty mass being kept at 15° and thoroughly stirred for 5 hours. The cellulose was noticeably affected in 90 minutes and dissolved within 24 hours, giving a faintly yellow liquid. During the subsequent 72 hours the colour changed to orange, but the solution remained clear and as at this stage the ultramicroscopic test was satisfactory the liquid was poured into 4 litres of water. The solid thus precipitated showed a tendency to coagulate to a plastic mass which was disintegrated by grinding under water to give a fine flaky powder. After filtration, the product was thoroughly washed with water and dried slowly at 30—40°/10 mm. Thereafter it was dissolved in hot rectified spirit and reprecipitated by the addition of water. Average yield of dry solid = 50 g., when the duration of the acetolysis varied from 95 to 101 hours.

The material was only slightly soluble in hot absolute alcohol, but easily soluble in rectified spirit. It was also readily soluble in cold acetic acid, acetone or chloroform, but less so in benzene. It melted without decomposition, but the temperature of fusion varied between 120° and 160° in different preparations. Moisture (determined at 110°), 1·35%; ash, negligible; sulphur, absent. Found: C, 49·9; H, 5·8; Acetyl, 64·2. Cellulose triacetate requires C, 50·0; H, 5·55; Acetyl, 62·5%. The specific rotation varied

from about + 10° to + 20°, but in the work now described material was selected which gave the following values:

	Solvent.	*c.*	$[\alpha]_D$.
Preparation I.	Chloroform.	1·75	+20·3°
,, II.	,,	2·58	19·4
,, III.	Acetone.	1·76	22·1

The acetate reduced Fehling's solution and as this result is important experimental details are given. A weighed quantity (0·3289 g.) was dusted into an excess of boiling Fehling's solution, the boiling being continued for 4 minutes. The weight of dry cuprous oxide thus obtained was 0·1467 g. In order to determine the effect of hydrolysis on the reducing power, 0·3289 g. of the acetate was dissolved in 60% alcohol containing 6% of hydrogen chloride, and the solution was boiled under a condenser for 4 hours. Thereafter the alcohol was replaced by 8% aqueous hydrochloric acid, and the mixture again boiled for 8 hours. A trace of solid matter remained undissolved and the solution, which was practically colourless, was neutralised and added to boiling Fehling's solution. As the weight of cuprous oxide obtained was 0·3250 g., the ratio of the reducing power before and after hydrolysis is 1 : 2·2. Attempts to eliminate the reducing component of the mixture effected only negligible separation but revealed, as the following experiment shows, that little significance can be attached to melting points in these compounds.

18 G. of the acetate were dissolved in 250 c.c. of hot rectified spirit and, on cooling, a first fraction of 12 g. separated. On adding 270 c.c. of water to the mother-liquor a precipitate formed and after further dilution to 850 c.c. the liquid was set aside over-night. The second fraction thus obtained weighed 5·4 g.

	Original material.	Fraction I.	Fraction II.
M. p.	155°	130—140°	140—148°
C	49·9%	49·6%	49·6%
H	5·8%	5·75%	5·6%
$[\alpha]_D$ in chloroform ...	+18·5°	+20·3°	+19·4°

Molecular weight determinations by Rast's process or by the cryoscopic method in acetic acid gave very variable results, owing apparently to the presence of a small quantity of insoluble dextrin. The presence of this impurity was confirmed by digesting the acetate with 2*N*-sodium hydroxide at 25° for 3 hours. On neutralisation, a flocculent precipitate separated which darkened on washing and drying. This product, which was present only in small amount, had the properties of a dextrin, decomposed at 220°, and gave C, 44·5; H, 6·4% [$(C_6H_{10}O_5)_x$ requires C, 44·4; H, 6·2%].

Methylation of the Depolymerised Acetate.—As the direct replace-

ment of acetyl by methoxyl in one operation is not of general application, the details of a typical experiment are quoted. The acetate (46 g.) was suspended in 225 c.c. of 2*N*-sodium hydroxide and disintegrated by mechanical stirring. After the temperature had been raised to 30°, small equivalent quantities of methyl sulphate and 30% sodium hydroxide were added at short intervals and, when three-fourths of the reagents had been introduced, the temperature was gradually raised to 70° and maintained at this point until the remainder of the methylating mixture had been added. Total reagents used : 110 c.c. of methyl sulphate and 110 g. of sodium hydroxide in 180 c.c. of water.

After completion of the reaction by heating at 100° for 35 minutes, the product remained in solution and was extracted with chloroform. The extract was dried and the solvent removed, when a clear syrup remained which set on cooling to a glass. Yield, 14 g.; OMe, 38·1%. The products of three similar preparations were united and subjected to three further methylations. As the methoxyl content increased, the solubility in alkali diminished so that the methylated product could be removed by filtration and, an important point, the process of chloroform extraction was discontinued so that the disaccharide derivatives remained in the alkaline liquor. Yield, 30 g.; OMe, 45·6%. Five subsequent methylations did not affect the methoxyl content, which agrees exactly with that required for a trimethyl cellulose.

The product was ground to a white solid and extracted repeatedly with boiling ether, which left a small residue (6%) undissolved. This had OMe, 43·7%, and was evidently derived from the cellulose dextrin present in the starting material. On removal of the solvent ether, the main product (94%) was isolated as a clear, colourless glass which could be powdered to a fine white solid softening at about 40°. This proved to be essentially tri(trimethyl 1 : 5-anhydroglucose) mixed with the corresponding methylated trisaccharide. Properties : readily soluble in cold ether, chloroform, benzene, or acetone, less soluble in methyl alcohol, rectified spirit or water; no action on Fehling's solution until after acid hydrolysis. Found : C, 52·7; H, 8·0; OMe, 46·2. A mixture of 70% of tri(trimethyl anhydroglucose) and 30% of a fully methylated trisaccharide requires C, 52·9; H, 7·9; OMe, 47·4%.

Solvent.	*c.*	$[\alpha]_D$.
Chloroform	3·564	+7·0°
Benzene	1·682	10·1
Acetone	3·367	15·7

Molecular Weight.—The mean of two determinations by Rast's method was 662. The mean of three consistent results by the

cryoscopic method with benzene as solvent was 663, and an independent repetition of the same process gave 642. The calculated value for the mixture postulated above is 625.

Simultaneous Hydrolysis and Condensation with Methyl Alcohol.—The mixture of methylated trisaccharide derivatives (4·3552 g.) was dissolved in methyl alcohol containing 1% of hydrogen chloride and heated at 110° for 70 hours. The solution, which was pale yellow and contained no suspended matter, was then neutralised with silver carbonate, filtered, and evaporated to dryness. 4·562 G. of a colourless glucosidic syrup remained, corresponding with a yield of 92% of the theoretical maximum. On distillation, the liquid boiled at 115°/0·3 mm. and 3·87 g. were collected in two fractions to permit of duplicate analyses. On the basis that the original methylated is a mixture of 70% of tri(trimethyl anhydroglucose) and 30% of a fully methylated trisaccharide, the above product of hydrolysis should consist of trimethyl methylglucoside (90%) and tetramethyl methylglucoside (10%). This was verified by analysis. Fraction I : C, 50·9; H, 8·3; OMe, 52·6. Fraction II : C, 50·95; H, 8·6; OMe, 52·8%. Calculated for the mixed glucosides : C, 51·0; H, 8·5; OMe, 53·5%.

Hydrolysis of the Mixed Glucosides.—The syrup (2·67 g.) was dissolved in 110 c.c. of 8% hydrochloric acid, a trace of charcoal added, and the solution boiled under a condenser until the rotation was constant. The subsequent treatment was as usual and as the mixed sugars, isolated initially as a syrup, weighed 2·2 g., the yield was 88%. In order to separate the sugars, the syrup was dissolved in water, and the solution thoroughly extracted with chloroform. The aqueous solution was evaporated to dryness under diminished pressure, the resulting syrup being redissolved in absolute alcohol which was, in turn, evaporated. Finally, the sugar was dissolved in pure dry ether and after filtration the solvent was slowly evaporated. On nucleation with 2 : 3 : 6-trimethyl glucose, the product solidified to a hard mass of crystals (1·85 g.), m. p. 105°; permanent $[\alpha]_D$ in water, for $c = 1{\cdot}058$, + 71·8° (Found : C, 48·5; H, 8·05; OMe, 41·6. Calc. : C, 48·65; H, 8·1; OMe, 41·9%).

The remaining methylated sugar was isolated from the chloroform solution, which was dried and evaporated, as a syrup weighing 0·25 g. This crystallised on keeping and, when purified from light petroleum, gave pure 2 : 3 : 5 : 6-tetramethyl glucose displaying the standard physical constants. Yield of trimethyl glucose : yield of tetramethyl glucose = 84% : 7%. Calc., 90% : 10%.

Direct Hydrolysis of the Methylated Product.—A 5% solution in 8% hydrochloric acid was fully hydrolysed on boiling for 30 minutes, and the sugars formed were isolated by standard methods. The

product was a clear syrup which, in accordance with the composition of the starting material, should consist of trimethyl glucose (90%) and tetramethyl glucose (10%) (Found: C, 48·9; H, 8·3; OMe, 41·1. The above mixture requires C, 48·9; H, 8·15; OMe, 42·9%).

Deacetylation of the Degraded Cellulose Acetates.—20 G. of the finely-powdered acetate were added to 66 c.c. of 33% aqueous dimethylamine. An appreciable rise of temperature took place and the liquid was cooled to 15° and kept, with frequent shaking, for several days. For the first 48 hours the liquid thickened, but after 96 hours it had become much more mobile. After 9 days, water was added and insoluble dextrins were removed by filtration through charcoal, the filtrate being evaporated to dryness under diminished pressure. A golden syrup remained which was thoroughly mixed with successive quantities of ether to remove acetodimethylamide, the undissolved residue being dissolved in a little water and precipitated by excess of absolute alcohol. A white, amorphous powder was thus obtained (1·85 g.) which sintered at 170° and melted with decomposition at 180° (Found in material dried in a vacuum at 110° until of constant weight: C, 42·3; H, 6·6; *M*, cryoscopic in water, 315. A disaccharide requires C, 42·1; H, 6·4%; *M*, 342). The product is therefore a disaccharide other than cellobiose; it reduced Fehling's solution both before and more particularly after hydrolysis with acids. For $c = 4{\cdot}2735$, $[\alpha]_D^{20^\circ}$ was $+ 14{\cdot}04^\circ$, no mutarotation being observed within 24 hours.

The alcohol mother-liquor which had yielded the above disaccharide gave on evaporation under reduced pressure 6·4 g. of a syrup. This was dissolved in water, the solution agitated with chloroform to remove traces of acetodimethylamide, and the aqueous portion neutralised and recovered. By means of fractional extraction with boiling alcohol, the material was separated into two portions, but it was found impossible to eliminate dimethylaminoglucosides, and consequently this section of the investigation is being continued by different methods.

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