

THE STRUCTURE OF γ -SUGARS

PART VI

THE SYNTHESIS OF A FURANOSE, A 6-METHYLKETOHEXOSE

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THE γ -sugars are unstable and have not yet been isolated in the free state. They occur in nature only in glycosidal combination, e.g., in sucrose, inulin, graminin and in sinistrin, on hydrolysis of which only the normal form of the sugar is isolated. Derivatives of many of the hexoses and pentoses are known to exist in the γ -form. The simplest derivatives of these labile sugars are the glycosides, which have been found to differ widely in their chemical properties from those of the normal sugars. Thus they are susceptible to hydrolysis by very dilute mineral acid and can be distilled without decomposition *in vacuo*. Furthermore, the γ -glycosides reduce permanganate solution in the cold, a property which has not yet been shown by any of the glycosides of normal sugars.

Haworth and collaborators are responsible for the accepted explanation of the difference in constitution, the γ -sugars being derived from furan, whilst the normal sugars are similarly related to pyran. In this connection it is of interest to note that Hersant and Linnell¹ obtained by synthesis a methylated hexose which could not possess a five-membered ring and which exhibited properties in agreement with those of the normal sugars. Fischer² first isolated γ -methyl-glucoside and Haworth and Law³ established their occurrence in natural products by proving that the fructose residue in sucrose was a γ -form.

Evidence for the furanose structure of γ -fructose rests mainly on the degradation of tetramethyl- γ -fructose obtained on hydrolysis of heptamethylsucrose through trimethylfructofuronic acid to trimethyl- γ -arabinolactone and then to dimethoxysuccinic acid,^{4,5,6}. Other structures^{7,8} have been suggested, but most of them are based upon studies of the oxidation products of derivatives and these oxidations are far from being quantitative. Hartley and Linnell⁹, however, on the basis of parachor studies of partially and fully methylated derivatives of γ -fructose, from the kinetic studies of the hydrolysis of sucrose and on account of the absence of mutarotation in both 3:4:6 trimethyl- γ -fructose and 6-methylfructose solutions, suggested for γ -fructose and its non-glycosidic derivatives an open chain keto-alcohol structure.

The determined parachor values of tetramethyl- γ -fructose and of tetramethyl- γ -methylfructoside were found to be significantly lower than those calculated for the structure assigned to γ -fructose by either Haworth¹⁰ as a five-membered ring or by Hudson¹¹ as a four-membered ring structure. Again five- and six-membered oxygen ring compounds are not normally different in their stability (Linnell and Melhuish¹²).

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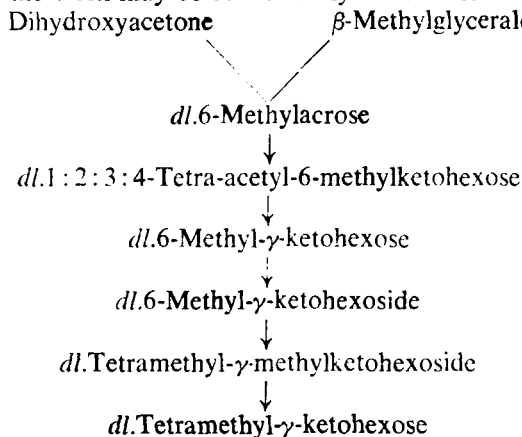
The stability of γ -fructose and the heat of activation of its conversion to normal fructose as obtained from the kinetic studies of sucrose lead to results which are not completely explained by the accepted furanose structure.

The half-life period of γ -fructose as has been shown by Hartley and Linnell¹³ and others^{14,15} to be too small to permit its isolation, and hence any structural enquiries concerning such compounds must rest on a study of their derivatives. The justification for such approach rests on a precise knowledge of the constitution of the derivative used. No derivative of a γ -sugar has yet been synthesised by an unambiguous route, although Hartley and Linnell¹¹ synthesised γ -fructose derivatives which could not exist in pyranose form¹⁶.

It was, therefore, decided to attempt the synthesis of a monomethyl-ketohexose in which the position of the methoxy group would be established beyond doubt by the method used. Such a method would not only avoid the possibility of a ring change during methylation, but would also be free from any criticism which might be directed towards the drastic reactions involved in the oxidative degradation methods adopted by Haworth and his collaborators.

By a modification of Fischer's synthesis of acrose^{17,18}, using pure dihydroxyacetone and monomeric β -methylglyceric aldehyde, a monomethyl-ketohexose was obtained which could not be other than a 6-monomethyl-ketohexose. No 6-monomethyl-ketohexose had been synthesised before.

The preparation of the starting materials for this synthesis have been previously reported¹⁹ and the main series of reactions involved in the remainder of the work may be conveniently summarised as follows:



The condensation of dihydroxyacetone and glyceraldehyde was shown by Fischer¹⁷ to take place in the presence of 1 per cent. of sodium hydroxide. Schmitz²⁰ (see also Jackson²¹ and Neuberg²²) obtained a relatively good yield of acrose using 0.1 per cent. barium hydroxide solution. Hersant and Linnell¹ effected condensation of dihydroxyacetone and methyl- γ -glyceraldehyde by using 0.1 per cent. barium

hydroxide solution for a total period of 5 weeks, sufficient barium hydroxide being added from time to time to maintain the alkalinity of the reaction. In the present research it was decided to employ the latter method as being less likely to induce epimerisation or resinification.

Dihydroxyacetone and β -methylglyceraldehyde in equimolecular proportions were therefore condensed in 5 per cent. aqueous solution in the presence of 0.1 per cent. of barium hydroxide. After a few days the solution became yellow and sufficient barium hydroxide was added from time to time to maintain a well-marked alkalinity, the solution being kept in the dark for a period of six weeks.

Fischer¹⁷ isolated *dl*-fructose from the condensation product through osazone formation, which he converted to osone and then to fructose by reduction. Hersant and Linnell¹ obtained *dl*-5-methylfructose by following the method of Fischer. Isolation was attempted according to Schmitz²⁰, who was able to isolate a crystalline hexose directly. The solution containing the condensation product was therefore neutralised and evaporated to dryness *in vacuo*, the residue being taken up in absolute alcohol, but on evaporation *in vacuo* no crystalline substances could be isolated. Addition of anhydrous ether to the alcoholic solution caused the separation of a white, amorphous substance which gave aggregates of needle crystals (m.pt. 99° to 101°C.) on treatment with phenylhydrazine acetate.

The number of products obtainable in this condensation must include derivations of *dl*-fructose, *dl*-sorbitose, *dl*-tagatose and *dl*-psicose, but previous experience has shown¹⁷ that *dl*-fructose predominates, the only other sugar isolated in small quantity being *dl*-sorbitose. Since dihydroxyacetone in the alkaline medium may enolise, it is not impossible that derivatives of the eight *dl*-aldoses might be present in the product.

It is not surprising, therefore, that little success crowned the efforts to isolate the osazones directly from the alcoholic solution of the product. A small yield of a crystalline product melting at 130° to 132°C. was obtained, however, and this could be *dl*-methylsorbosazone, but the yield was very small. Recourse was, therefore, made to a different method of isolation through the acetylated product. The acetylation was attempted according to the method of Erwig and Koenigs²³, but the yield was too small. Modification of the process of Hudson and Braun²⁴ (by using dry material, increasing the amount of acetic anhydride, stirring for a longer time, and special treatment of the chloroform emulsion obtained on extraction) gave more promise. After removal of the chloroform the main product was a light yellow syrup together with a few crystals. Keeping over potassium hydroxide *in vacuo* for 14 days caused no alteration. The crystals were probably traces of acetylated aldoses and the syrup—the main product—represented a 69 per cent. yield and gave analytical figures for a tetra-acetylmethylhexose. Saponification indicated that the purity was about 99 per cent. This saponification required special care because the methylhexose resulting may be readily attacked by acids or alkalis. Erwig and Koenigs found that the

liberated fructose was slightly attacked by 3 hours' boiling with 0.1N sulphuric acid.

Hudson and Braun observed a similar action of acid on the liberated fructose and after various modifications discarded the method of acid hydrolysis. They found that if 0.1N sodium hydroxide was used for saponification of fructose penta-acetate by shaking at 0°C., the liberated fructose was not attacked by the amount of alkali present.

Determination of the saponification value of the tetra-acetylmethylhexose was then carried out by shaking with 0.1N sodium hydroxide at room temperature for 4½ hours and followed by titration with 0.1N sulphuric acid using phenolphthalein as indicator. After saponification concentration of the liquid *in vacuo* after neutralisation left a brown syrup which reduced Fehling's solution slowly in the cold, vigorously on warming and reduced alkaline permanganate solution in the cold.

Saponification of larger quantities of tetra-acetylmethylhexose was carried out under the same conditions and the liquid was neutralised with dilute acetic acid, filtered and evaporated to dryness *in vacuo*. The residue was then extracted with absolute alcohol, filtered from the sodium acetate, decolorised with charcoal and concentrated *in vacuo*. It was redissolved in alcohol and reconcentrated until sodium acetate was completely eliminated. A pale yellow syrup remained, which after keeping in a vacuum desiccator for a long time became a toffee-like mass. Methoxyl determination of the product gave 4 figures which were slightly high probably owing to the presence of a little 1:6-dimethylhexose formed by condensation of β -methylglyceraldehyde with monomethylidihydroxyacetone formed by the action of alkali on β -methylglyceraldehyde.

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| 1. Molisch's test:— | Positive. |
| 2. Barfoed's reagent:— | Reduction on warming. |
| 3. Fehling's solution:— | Reduction slowly in the cold, vigorous on warming. |
| 4. Ammoniacal silver nitrate solution:— | Reduction in the cold, mirror on warming. |
| 5. Schiff's reagent:— | Magenta colour slowly. |
| 6. Alkaline potassium permanganate solution:— | Instantaneous reduction in the cold. |
| 7. Methyl alcoholic hydrochloric acid cold for 1 hour:— | Syrup did not reduce Fehling's solution, but reduced permanganate in the cold. |
| 8. Phenylhydrazine acetate solution:— | Needle crystalline osazone m.pt. 130° to 132°C. |
| 9. After standing in a vacuum desiccator for 3 months, 6-methylketohexose was found unchanged and still reduced alkaline potassium permanganate in the cold. | |

Reduction of Fehling's solution and potassium permanganate in the cold, reaction with Schiff's reagent and formation of a methylglycoside

in the cold are held to be characteristics of γ -sugars and are not given by normal sugars under the specified conditions.

Glycoside formation was accomplished by dissolving a quantity of *dl*- γ -methylhexose in methyl alcohol containing 1 per cent. of dry hydrogen chloride and keeping the solution at room temperature for 2 hours. On isolation a yellow syrup remained which failed to reduce Fehling's solution even after warming, but reduced alkaline permanganate in the cold. Any glycoside obtained by this established method should be a γ -glycoside; its behaviour towards permanganate together with its syrupy consistency differentiates it from the crystalline normal glycopyranosides.

Further indication of the nature of this glycoside was given on attempting its hydrolysis with N/100 aqueous hydrochloric acid: the hydrolysis was complete in 1 hour, whereas γ -methylglucopyranoside was hardly affected under these conditions.

The mixture of 6-methylhexoses was methylated with dimethylsulphate and sodium hydroxide²⁵ followed by Purdie's²⁶ reagent. The product on isolation did not crystallise and gave analytical figures for a tetramethyl-methylhexoside. It did not reduce Fehling's solution, but reduced permanganate in the cold. On hydrolysis it gave a liquid product which reduced Fehling's solution and potassium permanganate in the cold.

The product of the condensation of β -methoxy- α -hydroxypropionaldehyde and dihydroxyacetone possessed properties which sharply differentiated it from a pyranose. The condensation could not yield an amylenoxide structure owing to the blocking of the 6-hydroxyl group. It could possess a ketone structure or a 1:2, 2:3, 2:4 and 2:5-oxide rings of which the 2:5 is by far the most likely. The results of this synthetic experiment indicates that hexoses possessing other than the amylenoxide structure exhibit properties far more reactive than the normal sugars. These differences are greater than would be expected between derivatives of furane and pyrane, but the results of these experiments are difficult to explain on any other basis.

The separation of the product of this synthesis into individual sugars by partition of the acetylated material by chromatographic methods will be communicated later.

EXPERIMENTAL

Condensation of β -methoxy- α -hydroxypropionaldehyde and dihydroxyacetone.—Freshly prepared β -methoxy- α -hydroxypropionaldehyde (20 g.) dissolved in 400 ml. of water was mixed with a solution of dihydroxyacetone (20 g.) in 400 ml. of water and 1 g. of barium hydroxide dissolved in a little hot water was added to the mixture. The solution was kept protected from light for 6 weeks, sufficient barium hydroxide being added from time to time to maintain a well-marked alkalinity. The solution became yellow in colour during the condensation.

Isolation of the products.—(a) An attempt to isolate by treatment of the above solution with phenylhydrazine-acetate mixture resulted in the

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formation of a black oil from which a small quantity of pale yellow needles melting at 130° to 132°C. was obtained.

(b) The condensate was neutralised with dilute sulphuric acid and the filtered solution was evaporated *in vacuo* at 30° to 35°C. The light brown syrupy residue was dissolved in absolute alcohol and filtered. On removal of the alcohol no crystals appeared and the syrupy residue was redissolved in alcohol and diluted with ether when a voluminous, white, amorphous precipitate formed. After separation, washing with ether and drying over sulphuric acid a hygroscopic creamy product was obtained which reduced Fehling's solution on warming and ammoniacal silver nitrate solution in the cold.

The alcohol-ether filtrate was evaporated at ordinary temperature giving a light brown syrup which reduced Fehling's solution and ammoniacal silver nitrate solution.

6-Methylketo-hexose.—The solution (800 ml.) containing the condensation products was carefully neutralised and the clear filtrate was evaporated under reduced pressure at 30° to 35°C., a little alcohol being added towards the end of the process. A viscous yellow syrup (35 g.) was obtained which gave all the tests characteristic of a γ -sugar. In all some 200 g. was prepared.

6-Methyltetra-acetylketo-hexose.—The dried 6-methylketo-hexose (25 g.) was slowly added to a mixture of 150 ml. of acetic anhydride and 6.25 ml. of sulphuric acid, the mixture being strongly cooled. On stirring in the cold for about 2 hours complete solution was affected. The mixture was poured into 600 ml. of ice-cold water, neutralised with sodium bicarbonate, filtered and the residue, consisting of the acetylated sugar, was dissolved in chloroform. The filtrate was repeatedly extracted with chloroform, the chloroform solutions were washed with water, dried, treated with activated charcoal, filtered and evaporated under reduced pressure. The syrupy residue was kept over potassium hydroxide *in vacuo*, when signs of crystallisation appeared but the bulk of the material remained as a yellow syrupy liquid. Yield 32 g. (69 per cent.) Found C, 49.07; H, 7.03 per cent.; $C_{15}H_{22}O_{10}$ requires C, 49.72; H, 7.08 per cent. 0.5086 g. of syrup required 55.70 ml. of N/10 sodium hydroxide for complete saponification; theory requires 56.10 ml. of N/10 sodium hydroxide.

Purified 6-methylketo-hexose.—The tetra-acetate (75 g.) was hydrolysed with N/10 sodium hydroxide and after isolation in the usual way a yellow, very viscous syrup was obtained which reduced Fehling's solution slowly in the cold but vigorously on warming, reduced potassium permanganate solution in the cold and formed a glycoside on treatment with methyl alcoholic hydrogen chloride at normal temperature for 1 hour. These properties are characteristic of a γ -sugar. A determination of methoxyl using a modified Pregl method gave OCH₃, 16.78 per cent.: $C_6H_{11}O_5(OCH_3)$ requires 15.97 per cent. The slightly high figure was probably due to the presence of a small quantity of dimethyl derivative (*vide supra*).

6-Methyl- γ -methylketo-hexoside.—A solution of 6-methylketo-hexose (1 g.) in 15 ml. of pure methyl alcohol containing 1 per cent. of dry hydrogen chloride was kept at room temperature for 2 hours. The solution was neutralised with silver carbonate, filtered and the solvent removed by distillation. The residue, a brown syrup, failed to reduce Fehling's solution even after warming, but it reduced potassium permanganate solution in the cold. On hydrolysis the power to reduce Fehling's solution was restored.

Hydrolysis of 6-methyl- γ -methylhexoside.—A solution of 6-methyl- γ -methylhexoside (0.69 g.) in 12 ml. of N/100 hydrochloric acid was placed in a water-bath at 95°C. At intervals 0.2 ml. of the solution was removed and added to 1 ml. of Fehling's solution diluted with 2 ml. of water. The solution was replaced in the water-bath and the amount of reduction was observed at the end of 5 minutes. The results were compared with those of methylglucopyranoside treated similarly.

Time in Minutes after Beginning Heating at 95°C.	Reduction Observed	
	6-methyl- γ -methyl-keto-hexoside	methylglucopyranoside
5	Distinct reduction	Very slight reduction
10	" "	" "
15	Strong reduction	Slight "reduction"
30	Rapid "	" "
60	Complete "	" "

Tetramethyl- γ -methylketo-hexoside.—The 6-methylketo-hexose was methylated with dimethyl sulphate and sodium hydroxide by the normal method, at first at 30°C. and then at 70°C. After isolation the process was repeated and the product was worked up in the usual manner. The product was then further methylated, using methyl iodide and silver oxide. On isolation 12 g. of a yellow syrup was obtained which showed no reduction of Fehling's solution, but still reduced potassium permanganate solution in the cold. Found OCH_3 , 60.19 per cent.; $\text{C}_6\text{H}_7\text{O}(\text{OCH}_3)_5$ requires OCH_3 , 62 per cent.

Tetramethyl- γ -keto-hexose.—Hydrolysis of the tetramethyl- γ -methylketo-hexoside (1 g.) was effected with 1 per cent. aqueous hydrochloric acid by heating under reflux for half an hour. After neutralisation and isolation in the usual manner a syrup remained which reduced Fehling's solution and potassium permanganate solution in the cold.

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