THE STRUCTURE OF FUCOSE.

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ABSTRACT.

The complete structure of fucose has not hitherto been known, the position of the hydroxyl group on carbon atom 5 being the part in doubt. In order to clear up this point a lactone of the methyl tetronic acid has been prepared, and from its optical properties the position of the hydroxyl group on carbon atom 4 of this substance, corresponding to carbon atom 5 of fucose, has been established, thus giving the complete structure of fucose. The position of the hydroxyl groups on carbon atoms 2, 3, and 4 of fucose has also been verified by a study of the optical properties of the amides of the methyl tetronic acid and of fuconic acid (prepared for the first time), together with fuconic lactone. An improved method for preparing fucose is also recorded.

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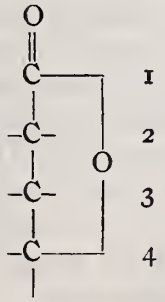
I. INTRODUCTION.

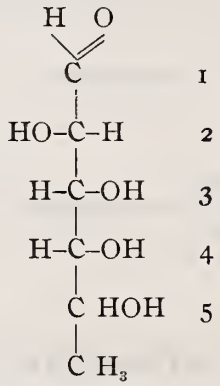
Some relationships existing between the constitution and optical rotatory powers of certain sugar derivatives have been discovered by Hudson and by Levene. Hudson [1] found that when the formula of a sugar acid lactone—that is, a gamma lactone—is written vertically, thus,



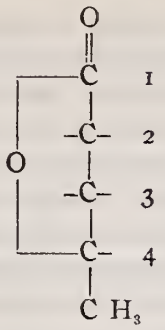
[1] This and succeeding reference numbers refer to bibliography at end of this paper.

the compound is dextrorotatory when the lactone ring is on the right of the carbon chain; and, vice versa, when the ring is on the left of the chain, the compound will be levorotatory. As the position of the ring is determined by the position which the hydroxyl group had on the carbon atom 4 before the ring was formed, a means of determining the position of the hydroxyl group on this carbon atom is thus available. He also found [2] that the dextrorotatory amides of the sugar acids have the hydroxyl group on carbon atom 2 on the right of the carbon chain, while the levorotatory amides have the hydroxyl on the left. Levene [3] found that the same principle holds for metallic salts and phenylhydrazides of the sugar acids.

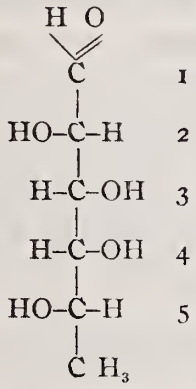
Now the configuration of fucose has been established[4] to be that of a methylpentose,



the position of the hydroxyl group on carbon atom 5 having not yet been determined. It would follow from the above that if a methyl tetronic acid lactone could be obtained from fucose, it would be possible to apply the above rule to it and thus determine the position of the hydroxyl group on carbon atom 4 of the lactone, which corresponds to carbon atom 5 in fucose. Previous to the present investigation this compound had not been prepared; but Hudson and Chernoff [5] obtained a methyl tetronic acid lactone from rhamnose by applying a method devised by Nef, Hedenburg, and Glattfeld [6] for the oxidation of pentoses to the corresponding tetronic acids. The same results should be expected from fucose, giving the methyl tetronic lactone desired. This proved to be the case. A yield of 7.5 g of the substance was obtained from 50 g of fucose. The lactone was strongly levorotatory (−63.°65), which indicates that its configuration is



and hence, the configuration of fucose is



This is in accordance with the conclusion reached by Mayer and Tollens,[7] based upon the similarity of the specific rotations of l-galactose (−81°) and fucose (−75.°5).

Incidentally, the position of the other three hydroxyl groups in fucose may be verified by the same line of reasoning. Fuconic acid lactone[8] is dextrorotatory (+71.°7); therefore, the hydroxyl on the carbon atom 4 will, according to the rule, be on the right of the carbon chain. Fuconic amide, on the other hand, as will be shown in the experimental part, is levorotatory (−31.°13): hence, the position of the hydroxyl group on carbon atom 2 is on the left of the carbon chain. The amide of the methyl tetronic acid was prepared, as described below, so that the position of the hydroxyl group on carbon atom 2 in this compound, corresponding to carbon atom 3 in fucose, could be verified. This substance was found to be dextrorotatory (+18.°48); therefore, the hydroxyl group on this carbon atom is on the right of the chain. Its position is further established, as Hudson has pointed out, by the fact that the methyl hexonic lactone prepared by Mayer and Tollens from fucose[9] is dextrorotatory (+33.°3). It will be seen

from the formula that the carbon atom 2 in the methyl tetronic acid becomes carbon atom 4 in either of the two fucohexonic acids.

Thus by the preparation of the methyl tetronic acid lactone from fucose and the application of the above rules to this substance, the position of the hydroxyl group on the carbon atom 5 of fucose has been determined. Likewise, the positions of the hydroxyl groups on carbon atoms 2, 3, and 4 of this sugar, heretofore determined by purely chemical means, have been verified.

The present communication also contains an improved method for the preparation of fucose.

II. EXPERIMENTAL.

1. PREPARATION OF FUCOSE.

About 10 kg. of seaweed* which has been air dried sufficiently to become brittle are coarsely ground in a meat chopper. The ground material is then treated at room temperature for two days with 3 per cent HCl (3 liters to each kilogram), after which it is repeatedly washed with water by decantation until the washings are no longer acid to litmus. The residue is pressed out and dried.

One kilogram of the dried substance is added to 8 liters of boiling 2 per cent sulphuric acid and simmered for 3 hours. The undissolved material is filtered upon large Büchner funnels and thoroughly washed. The sulphuric acid in the filtrate and washings is neutralized with a paste of precipitated barium carbonate. The resulting barium sulphate is removed by filtering the solution through a thin layer of decolorizing carbon on moistened filter paper in a Büchner funnel. The filtrate is treated with basic lead acetate until no further precipitate is formed. The solution is next freed from the lead precipitate, and the excess of lead in the filtrate removed by the cautious addition of dilute sulphuric acid. After the removal of the lead sulphate the solution is evaporated under diminished pressure to a sirup of about 175 cc. This sirup is dissolved in an equal volume of methyl alcohol by warming on a water bath and sufficient ethyl alcohol added to make 2 liters. The precipitate formed is filtered upon decolorizing carbon, as outlined above, and the filtrate evaporated to 150 cc. The sirup is washed from the flask with small portions of absolute alcohol by warming, and when all is removed sufficient absolute alcohol is added until the solution has a volume of

* The material used in this work was *Ascophyllum nodosum*, which was obtained from Woods Hole, Mass., through the courtesy of Dr. R. E. Coker of the U. S. Bureau of Fisheries.

450 cc. Seventy-five grams of phenylhydrazine is added and the mixture placed in an ice box over night to crystallize completely. The hydrazone is filtered and washed, first with absolute alcohol and finally with ether. The yield is 100 g.

To obtain the sugar, the hydrazone thus prepared is sufficiently pure. Seventy-five grams of the material is suspended in 1,800 cc of water which is heated to about 90° C., and 36 g of benzaldehyde is added while the mixture is rapidly stirred. The stirring is continued for ½ to ¾ hour. It is advantageous to have an atmosphere of carbon dioxide in the flask to prevent the benzaldehyde from becoming oxidized. After removing the benzaldehyde phenylhydrazone the filtrate is treated with carbon. The fucose solution is then concentrated to a thick sirup and dissolved in 800 cc of hot absolute alcohol. Some impurities are thrown out by this process, so the solution is filtered, after which it is concentrated to about 50 cc. This sirup is removed to a beaker and the flask washed out with small portions of absolute alcohol by warming, 50 cc in all being used for this purpose. The sugar begins to crystallize almost at once, and the crystallization is completed over night in an ice box. It is freed from mother liquor, washed with absolute alcohol and ether, and dried. The yield is 38 to 40 g. A sample of this material had the following rotation:

$$\left[\alpha\right]_{D}^{20} = -75.^{\circ}55 \text{ (2.5015 g in 50 cc water solution).}$$

Generally, another small crop of crystals may be obtained by concentrating the mother liquors.

To recrystallize the sugar it is dissolved in distilled water making a 25 per cent solution and treated with decolorizing carbon. The filtrate is concentrated under diminished pressure to a thick sirup and dissolved from the flask by warming on the water bath with three volumes of absolute alcohol, then allowed to crystallize. The crystals are washed with absolute alcohol and ether, and dried. The mother liquors may be worked over so that almost a quantitative yield is obtained. The purified sugar had the following specific rotation:

$$\left[\alpha\right]_{D}^{20} = -75.^{\circ}48 \text{ (5.0742 g of sugar in 100 cc water solution.)}$$

$$\left[\alpha\right]_{\lambda 5461\ \text{Å}}^{20} = -88.^{\circ}92 \text{ (5.0742 g of sugar in 100 cc water solution.)}$$

2. METHYL TETRONIC ACID LACTONE FROM FUCOSE.

Fifty grams of fucose was dissolved in 250 cc of water and added to a solution of 93 g of KOH dissolved in 5,250 cc of water. Air freed from carbon dioxide was continuously drawn through the mixture at a fairly rapid rate for 60 hours by means of a water pump. During working hours the solution was heated to 40 to 45°. The KOH was then neutralized and the organic acids were liberated by the cautious addition of HCl. This was determined by means of thymol blue. Just enough HCl was added to give a distinct red color showing the presence of free HCl. The solution was evaporated under diminished pressure until a sufficient amount of KCl had separated to cause bumping. This was filtered off and washed with a little cold water, and the filtrate was adjusted to the point where all the free HCl was exactly neutralized. Concentration was continued to almost dryness and the resulting mass dissolved in a little water and again evaporated to dryness to insure the removal of formic acid.

The residue was extracted three times with boiling absolute alcohol, 250 cc being used for each extraction. The alcoholic liquor was filtered and evaporated to a thick sirup, dissolved in about 100 cc of absolute alcohol, and again evaporated. The resulting sirup was extracted with 500 cc of dry ethyl acetate. An insoluble gum remained, while the supernatant liquid was cloudy; but after a day an insoluble gum separated, leaving a clear solution. This solution was evaporated under diminished pressure to a mobile liquid which was exhausted with dry ether by vigorous shaking for some time. After a day the ether layer was clear, and a gumlike substance separated. Upon standing two days more, beautiful, long crystals appeared on the walls of the flask, while the insoluble material was a semisolid mass of crystals. The ether was decanted and the crystals dissolved by warming in 30 cc of ethyl acetate. Upon cooling, the solution immediately crystallized. The yield of the crude product was 7.5 g. The substance was removed from the mother liquor and recrystallized from a few cc of ethyl acetate.

Its melting point was 111°C. uncorrected. The substance when first dissolved in water was neutral, but gradually became acid, showing its lactone character. 0.0928 g of the material was dissolved in 10 cc of 0.1 normal KOH, warmed to the temperature of a boiling water bath, and allowed to cool. The excess of KOH was titrated with 0.1 normal HCl using phenolphthalein as an indicator. In this way the sample required 7.01 cc of 0.1 normal KOH for neutralization; calculated for $C_5H_8O_4$, 7.03 cc. The optical rotation of the substance was as follows: 1.2529 g dissolved in 25 cc of water at 20° C rotated the plane of polarized light 6°38 to the left in a 2 dm tube using sodium light. Therefore,

$$\left[\alpha\right]_D^{20}=\frac{(100)\,(-6.38)}{4\,(1.2529)\,(2)}=-63^\circ.65.$$

0.2028 g of substance gave on combustion 0.1101 g of water and 0.3379 g of CO_2.

Calculated for $C_5H_8O_4$	C, 45.43 per cent	H, 6.10 per cent.
Found	45.44	6.07

3. METHYL TETRONIC AMIDE.

Dry ammonia was passed into a solution of 3.3 g of the lactone dissolved in 30 cc of absolute alcohol until it became saturated. Crystallization of the amide could not be induced; so after a day the liquid was evaporated under diminished pressure to about 10 cc, and 10 cc of dry ether added. After a week crystallization commenced and was complete in a very short time after it started. The crystals were filtered, washed with absolute alcohol and ether, and dried. The yield was 2.5 g. The melting point of the crude substance was 112° C. uncorrected. To recrystallize the compound, it was dissolved, by warming, in 15 cc of absolute alcohol, filtered, and 15 cc of dry ether added.

The melting point was 112°.5 C. Its specific rotation was as follows:

0.5006 g of substance in water to make 25 cc rotated the plane of polarized light 0.74° to the right, using a 2 dm tube; hence,

$$\left[\alpha\right]_D^{20}=\frac{(100)\,(+0.74^\circ)}{(4)\,(0.5006)\,(2)}=+18^\circ.48.$$

Nitrogen was determined by the Kjeldahl method.

0.2537 g of substance used 17.07 cc of 0.1 normal HCl.

Calculated for $C_5H_{11}O_4N$	N = 9.40 per cent.
Found	N = 9.43

4. FUCONIC AMIDE.

This substance has not hitherto been recorded. 15 g of fucose were dissolved in 75 cc of water and 15 g of bromine added. Upon shaking occasionally, during an hour, the bromine was dissolved. The solution was allowed to stand for two days, after which the excess of bromine was removed by distillation under reduced pressure. The resulting liquid was made neutral with sodium hydroxide and then treated with slightly more than the calculated amount of barium chloride. Barium fuconate immediately began to crystallize, the crystallization being complete as soon as the solution cooled to room temperature. The yield was 19.5 g. The barium was removed from 10 g of this salt by the addition of sulphuric acid. The resulting solution was freed from barium sulphate and evaporated under diminished pressure to a thick sirup, after which it was heated, *in vacuo*, at the temperature of a boiling water bath for ½ hour. The residue was dissolved by warming in 150 cc of absolute alcohol, and filtered.

The filtrate was evaporated to a thick sirup, dissolved in 100 cc more of absolute alcohol, and again evaporated to a sirup. This was dissolved in 60 cc of absolute alcohol and the solution was saturated with dry ammonia. Crystallization of the amide began within a few minutes.

The melting point of the crude product was 180°.5 uncorrected. Upon recrystallizing from 85 per cent alcohol its melting point was unchanged.

1.0021 g of substance in water to make 50 cc rotated the plane of polarized light 1°.248 to the left, using a 2 dm tube; hence,

$$\left[\alpha\right]_D^{20}=\frac{(100)\,(-1.248)}{2\,(1.0021)\,(2)}=-31°.13.$$

By Kjeldahl estimation 0.3037 g of substance used 16.86 cc of 0.1 normal HCl.

Calculated for $C_6H_{13}O_5N$.....N 7.82 per cent
Found..... 7.78

III. SUMMARY.

I. An improved method for the preparation of fucose is described.

II. The methyl tetronic lactone from fucose has been prepared, and from its optical properties the position of the hydroxyl group on carbon atom 5 of fucose has been determined, thus giving the complete structure of fucose.

III. The amides of the methyl tetronic acid and fuconic acid have been prepared, and from a study of their optical properties together with those of fuconic lactone, the positions of the hydroxyl groups on carbon atoms 2, 3, and 4 of fucose, heretofore determined by purely chemical means, have been verified.

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