curred spontaneously during this process. Recrystallization from 35 ml. of absolute ethanol yielded 10.1 g. (83%) of material, m.p. 119–120°.

Anal. Calcd. for $C_4H_{10}O_4$ (122.1): C, 39.34; H, 8.25. Found: C, 39.57; H, 8.08.

BERKELEY, CALIF.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, WASHINGTON UNIVERSITY, ST. LOUIS, MO.]

Concerning the Synthesis of D-Mannosamine and D-Glucosamine from D-*arabo*-3,4,5,6-Tetraacetoxy-1-nitro-1-hexene

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RECEIVED NOVEMBER 9, 1959

The action of methanolic ammonia on D-*arabo*-3,4,5,6-tetraacetoxy-1-nitro-1-hexene yields both 2-acetamido-1,2-dideoxy-1-nitro-D-mannitol and 2-acetamido-1,2-dideoxy-1-nitro-D-glucitol, with the former predominating in a ratio of about 6:1. The action of hydrochloric acid on the sodium salts of these two compounds (Nef reaction and *N*-acetyl hydrolysis) yields, respectively, D-mannosamine hydrochloride and D-glucosamine hydrochloride in high yield.

Considerable biochemical interest in D-mannosamine (2-amino-2-deoxy-D-mannose) has been aroused by the recent observation that it occurs naturally as a structural component of *N*-acetylneuraminic acid.[1] Consequently, the development of reasonable methods for the laboratory synthesis of the aminosugar assumes new importance. Of the methods developed to date[2–5] the preparation based on D-arabinose reported by O'Neill[5] appears to be the best route to D-mannosamine. This method involves the conversion of D-arabinose to D-*arabo*-3,4,5,6-tetraacetoxy-1-nitro-1-hexene,[6] reaction of the latter with alcoholic ammonia to give 2-acetamido-1,2-dideoxy-1-nitro-D-mannitol and, finally, application of the Nef reaction followed by acetylation to give D-mannosamine pentaacetate. The latter was converted to D-mannosamine hydrochloride by hydrolysis with hydrochloric acid.

We had independently studied the synthetic route reported by O'Neill but had reached somewhat different conclusions regarding the course of the reaction of D-*arabo*-3,4,5,6-tetraacetoxy-1-nitro-1-hexene with ammonia. In view of this, and since we had developed a somewhat more direct route from the acetylated sugar nitroölefin to D-mannosamine hydrochloride, it seems appropriate to record briefly our experiences with the synthesis.

Contrary to the findings of O'Neill, who concluded that the addition of ammonia to D-*arabo*-3,4,5,6-tetraacetoxy-1-nitro-1-hexene is stereospecific and gives only the D-*manno* isomer, we found rather that the addition is simply stereoselective and that the yield of D-*manno*:D-*gluco* isomer is approximately 6:1. We observed further that the 2-acetamido-1,2-dideoxy-1-nitro-D-mannitol (m.p. 172–173°, $[\alpha]^{25}D$ −16.8°) does not depress the melting point of 2-acetamido-1,2-dideoxy-1-nitro-D-glucitol (m.p. 155–156°, $[\alpha]^{25}D$ −12.7°), so that slight contamination of the former by the latter is difficult to detect. The epimeric substances are, however, readily separated by fractional crystallization from absolute ethanol, in which the D-*gluco* isomer is considerably more soluble than is its epimer.

Application of the Nef reaction, using hydrochloric acid rather than sulfuric acid, to the epimeric acetamidonitroalcohols, followed by heating to hydrolyze the amide linkage yields, respectively, D-mannosamine hydrochloride and D-glucosamine hydrochloride in high yield. The acidic hydrolysis of the *N*-acetyl group in *N*-acetyl-D-glucosamine was observed to be much slower than in *N*-acetyl-D-mannosamine.

The addition of ammonia to the acetylated sugar nitroölefins is reminiscent of its similar addition to the acetylated sugar disulfone olefins.[7] It is noteworthy, however, that whereas the addition to D-*arabo*-3,4,5,6-tetraacetoxy-1,1-bis-(ethanesulfonyl)-1-hexene apparently gives exclusively a product with the D-*gluco* configuration, the addition to the double bond of D-*arabo*-3,4,5,6-tetraacetoxy-1-nitro-1-hexene gives preponderantly a product with the D-*manno* configuration. The stereospecificity in the former case has been explained by Hough and Taylor[8] on the basis of neighboring group participation, whereas the latter case has been rationalized by O'Neill on the basis of rotational conformation. It is apparent that no single explanation is appropriate for both cases.

We are at present examining the addition of ammonia and various amines to other sugar nitroolefins and plan to report on these studies at a later date.

Experimental

2-Acetamido-1,2-dideoxy-1-nitro-D-mannitol and 2-Acetamido-1,2-dideoxy-1-nitro-D-glucitol.—Fifteen grams of D-*arabo*-3,4,5,6-tetraacetoxy-1-nitro-1-hexene,[6] m.p. 114–115°, was covered with 150 ml. of absolute methanol and the mixture was cooled to 0°. Anhydrous ammonia was bubbled into the mixture to approximate saturation, during which operation the acetylated nitroölefin dissolved. The resulting solution was protected from moisture with a drying-tube and allowed to warm to room temperature. After standing overnight, the solution was concentrated in a stream of dry air to a semi-crystalline mass. Filtration, washing with absolute ethanol and recrystallization from absolute ethanol (about 300 ml.) yielded 5.3 g. of pure 2-

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acetamido-1,2-dideoxy-1-nitro-D-mannitol, m.p. 172–173° and $[\alpha]^{25}D$ −16.8° in water (*c* 2.4). O'Neill[5] reports m.p. 172–173° and $[\alpha]^{24}D$ −13.2° in water (*c* 1.1) for this compound.

The mother liquors from the initial reaction mixture were concentrated to a semi-crystalline mass and extracted several times by trituration at room temperature with chloroform to remove acetamide. The resulting residue was combined with the above recrystallization mother liquors, concentrated and recrystallized from ethanol to yield 2.7 g. of mixed acetamidonitroalcohols, m.p. 158–165°. Fractional recrystallization of the latter from absolute ethanol yielded a further 0.75 g. of 2-acetamido-1,2-dideoxy-1-nitro-D-mannitol (total yield, 57.5%), m.p. 172–173°, and 1.1 g. (10.4%) of 2-acetamido-1,2-dideoxy-1-nitro-D-glucitol, m.p. 155–156° and $[\alpha]^{25}D$ −12.7° in water (*c* 3.6).

Anal. Calcd. for $C_8H_{16}O_7N_2$: C, 38.1; H, 6.39; N, 11.1. Found: C, 38.1; H, 6.68; N, 10.7.

D-Mannosamine Hydrochloride and D-Glucosamine Hydrochloride.—A solution of 9.5 g. of 2-acetamido-1,2-dideoxy-1-nitro-D-mannitol in 23 ml. of 2 *N* sodium hydroxide was added dropwise to 19.5 ml. of concentrated hydrochloric acid with vigorous stirring. After the addition, the solution was brought briefly to the boiling point, again cooled to room temperature and saturated with hydrogen chloride gas. The precipitated sodium chloride was removed by filtration (Whatman no. 42 paper) and the filtrate, after dilution with an equal volume of water, was decolorized by filtration through a layer of Celite and decolorizing carbon. The solution was concentrated at reduced pressure and residual hydrogen chloride was removed from ther esulting sirup *in vacuo* over potassium hydroxide. The sirup was dissolved in 15–20 ml. of methanol containing a few drops of water and brought to crystallization by the gradual addition of acetone. Seeding crystals of D-mannosamine hydrochloride are advantageous in this initial crystallization. There was obtained 7.6 g. (93%) of nearly pure D-mannosamine hydrochloride, $[\alpha]^{25}D$ −2.9° in water (*c* 11). A single recrystallization from moist ethanol with the addition of acetone gave material with $[\alpha]^{25}D$ −3.2° in water (*c* 10). The reported[9] value in water is −3°. The product gave an X-ray powder diffraction pattern identical with that of D-mannosamine hydrochloride prepared by the alkaline isomerization of *N*-acetyl-D-glucosamine.[4,10]

2-Acetamido-1,2-dideoxy-1-nitro-D-glucitol was converted to D-glucosamine hydrochloride by the process just described except that the acidic solution from the Nef reaction was refluxed for 4 hours to complete the hydrolysis of the *N*-acetyl function. Crystallized from a small amount of water by the addition of ethanol, the product (85% yield) showed $[\alpha]^{25}D$ +68° equil. in water (*c* 1.7). A single recrystallization from water–ethanol raised this to the reported[9] value of +72°. The product gave an X-ray powder diffraction pattern identical with that of a commercial sample of D-glucosamine hydrochloride (Eastman Organic Chemicals, Rochester, N. Y.).

Acknowledgment.—The authors are pleased to acknowledge the generous support of the Corn Industries Research Foundation, Washington, D.C., during the course of this work.

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(10) We are indebted to Dr. Saul Roseman, Rackham Arthritis Research Unit, University of Michigan, for this latter sample and to Mr. A. V. Guzzo of this Laboratory for the X-ray diffraction measurements.

[CONTRIBUTION FROM RESEARCH LABORATORIES, SYNTEX, S. A.]

Steroids. CXXXI.[1] A New Series of 6-Substituted Progesterone Analogs

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RECEIVED JUNE 18, 1959

Treatment of 5α,6α-oxidoprogesterone bis-ethyleneketal with acetylene dimagnesium bromide led to 6β-ethynylpregnane-5α-ol-3,20-dione bis-ethyleneketal which, following hydrolysis of the ketal functions and dehydration of the 5α-hydroxyl group, provided 6β-ethynylprogesterone. While this substance could not be inverted to the corresponding 6α-ethynyl epimer, it was capable of conversion to 6α-(1-chlorovinyl)-progesterone and 6α-acetyl-progesterone. By controlled reductions of the original ethynylated fission product either 6β-vinyl or 6β-ethylpregnane-5α-ol-3,20-dione bis-ethyleneketal could be obtained. These compounds were then converted by appropriate manipulation to 6α-ethylprogesterone and 6β-vinylprogesterone.

Our recent observation[2] that steroid 5α,6α-epoxides upon treatment with phenylmagnesium bromide may be readily opened to provide 6-phenylated steroids, prompted us to undertake a similar investigation of epoxide openings employing acetylene dimagnesium bromide. While *a priori* it was conceivable that the use of this reagent could lead to a bis-substituted ethyne the possibility appeared remote since it had already been observed[3] that steroidal 17-ketones upon treatment with acetylene dimagnesium bromide provided exclusively the monosubstituted ethyne. On the basis of this earlier work as well as the present instance where only a monosubstituted ethyne was formed, it appears that the steric environment of the reactive center is the controlling factor governing mono- or disubstitution.[4]

Thus when 5α,6α-oxidopregnene-3,20-dione bis-ethyleneketal[5] was treated with acetylene dimagnesium bromide in tetrahydrofuran at reflux temperature the epoxide generally was smoothly opened to provide 6β-ethynylpregnane-5α-ol-3,20-dione bis-ethyleneketal (IIa) in 85% yield. Unexplicably on occasion the yields were less than 10% and in these cases the residues were non-crystalline and on the basis of infrared spectroscopy appeared to be free of either mono- or disubstituted ethynes.[6] No attempt was made to characterize this material.

Upon treatment of IIa with aqueous perchloric acid in tetrahydrofuran[7] the ketal functions were hydrolyzed in high yield to provide the corresponding dione IIIa. Dehydration of IIIa with thionyl chloride in pyridine then led to 6β-ethynylprogesterone (IVa); fully characterized by elemental

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