An Efficient Synthesis of L-Ascorbic Acid and [5-$^2H_1$]-L-Ascorbic Acid

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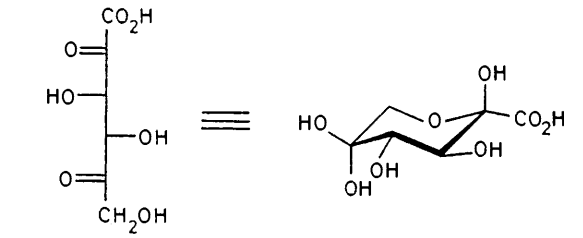
Summary L-Ascorbic acid has been synthesized *via* the regio- and stereo-selective reduction of D-*threo*-2,5-hexodiulosonic acid; this procedure also can be used for preparation of the 5-deuterio-compound.

THERE has been much recent interest in the synthesis of L-ascorbic acid[1] and in specifically labelled derivatives[2] of this biologically important molecule. We report herein a novel synthesis based on the regio- and stereo-selective reduction of D-*threo*-2,5-hexodiulosonic acid (**1**).[3]

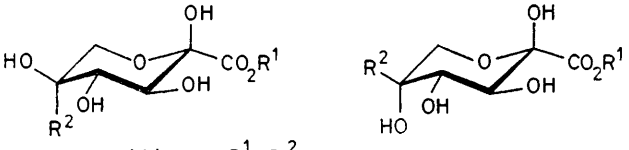
D-*threo*-2,5-Hexodiulosonic acid is readily available in high yield (85%) by the oxidation of glucose with various species of *Acetobacter* and *Pseudomonas* bacteria.[4] The gross structure of (**1**) was first determined by Katznellson and his co-workers,[5] and confirmed by the work of Kondo,[6] Wakisaka,[7] and Bernearts and DeLey.[8] Exhaustive reduction of (**1**) with $NaBH_4$ has been reported to afford a mixture of D-gluconic, D-mannoic, L-idonic, and L-gulonic acids.[5,7,8] Ruff oxidation of the mixture afforded a mixture of D-arabinose and L-xylose in which D-arabinose (containing the undesired configuration at C-5) was claimed to be the major component.[7] Catalytic hydrogenation of (**1**) was reported to afford, regioselectively, a mixture of L-*xylo*- (**2a**) and D-*arabino*-2-hexulosonic acid (**3a**) in which the latter (also containing the undesired configuration at C-5) predominated.[7,9,10]

Our recent determination of the solution structure of (**1**) by ^{13}C n.m.r. spectroscopy as the hydrated pyranose form depicted[11] casts some light on the above observations. If it is assumed that (**1**) is reduced in its stable pyranose form, catalytic reduction of the cyclohexylidene-like 5-keto-unit would be expected to occur predominately from the equatorial face of the molecule, affording the keto-acid isomer (**2b**) which is in agreement with the observed results.† However, ample precedent exists which suggests the *opposite* stereochemical result would be obtained in the reduction of (**1**) with a hydride reductant.[12]

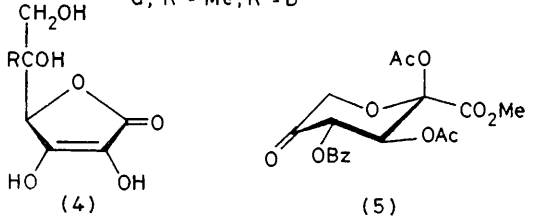
Indeed we have found that the reduction of the sodium salt of (**1**) in water at 0 °C with 0·25 equiv. of $NaBH_4$ affords in 89% yield a mixture of the keto-acids (**2a**) and (**3a**) in the ratio of 86:14.‡ Also formed is a mixture of the four hexonic acids L-gulonic, L-idonic, D-mannoic, and D-gluconic (in a ratio of *ca.* 40:27:19:15)‡ in 3—4% yield. That the observed stereochemical result is due to the axial delivery of hydride in the sterically biased cyclohexylidene-like system (**1**) is supported by the exclusively axial delivery of hydride in the recently reported reduction of the protected 2,5-hexodiulosonate (**5**).[2a]



(**1**)



(**2**) a; $R^1 = R^2 = H$ (**3**)
b; $R^1 = Me, R^2 = H$
c; $R^1 = H, R^2 = D$
d; $R^1 = Me, R^2 = D$



(**4**) a; R = H (**5**)
b; R = D

The mixture of 2-keto-acids derived from the $NaBH_4$ reduction of (**1**) is separable by either ion-exclusion chromatography§ or conversion of the mixture into the corresponding methyl esters followed by silica gel chromatography or fractional crystallization. Thus, lyophilization of the aqueous reduction mixture of (**1**), followed by esterification of the freeze-dried solids in methanol (using Dowex-50 ion exchange resin, hydrogen form) with azeotropic removal of trimethyl borate, affords an 85:15 mixture of the esters (**2b**) and (**3b**) in 88% yield¶ from which (**2b**) is obtained in 62% isolated yield on crystallization from methanol (m.p. 154—155 °C).** Lactonization of (**2b**) with $NaHCO_3$ in refluxing methanol generates sodium L-ascorbate in 93% yield [57% overall from (**1**)]. Conversion into the free acid followed by crystallization from methanol affords L-ascorbic acid (**4a**) {m.p. 188—189 °C, $[\alpha]_D^{25}$ +21° (*c* 10, H_2O)} identical with authentic L-ascorbic acid.

† The catalytic reduction of (**1**) in water at pH 8 with Raney Ni catalyst and hydrogen (40 lb in^{-2}) at ambient temperature was re-examined. On uptake of 1 equiv. of hydrogen, h.p.l.c. analysis of the reaction mixture indicated a regioselective formation of (**2a**) and (**3a**) (20:80, 82% yield) (ref. 12) thus confirming and quantifying Wakisaka's earlier observation.

‡ Ratios were determined by h.p.l.c. analysis using Dowex 50 × 8 column in calcium form. Yields were determined using an Aminex-25 anion exchange column in the formate form.

§ Dowex-50 × 8 Resin in calcium form using water as eluant.

¶ Based on g.l.c. analysis of the per-trimethylsilylated methyl esters on a 2 m OV-210 column.

** All compounds reported give satisfactory combustion analysis and exhibit physical and spectral data consistent with the assigned structures.

The utility of this synthesis for the formation of C-5 labelled L-ascorbic acid is demonstrated by the reduction of (**1**) with $NaBD_4$ at 0 °C in water to give (**2c**) and (**3c**) (85:15) in 81% yield. Isolation of the reduced acids by ion-exclusion chromatography§ afforded the calcium salt of (**2c**) in 70% yield. Esterification as above generated methyl [5-$^2H_1$]-L-*xylo*-2-hexulosonate (**2d**) in 59% isolated yield. Lactonization, followed by isolation of the free acid, produced [5-$^2H_1$]-L-ascorbic acid (**4b**) (m.p. 186—188 °C) in 41% overall yield.

The unexpectedly high regio- and stereo-selective nature of the sodium borohydride reduction of (**1**) offers a convenient and highly efficient synthesis of L-ascorbic acid and its C-5 labelled derivatives.

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