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SYNTHESIS OF 1-N-(β-*D*-GLUCOPYRANOSYL)- AND 1-N-(1-DEOXY-2,3,4,5,6-PENTA-O-ACETYL-*D*-GALACTIT-1-YL)ASCORBIGENS

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The 1-N-carbohydrate-containing ascorbigens 1-N-(β-D-glucopyranosyl)ascorbigen and 1-N-(1-deoxy-2,3,4,5,6-penta-O-acetyl-D-galactit-1-yl)ascorbigen were synthesized.

Keywords: ascorbigen, *L*-ascorbic acid, 3-hydroxymethylindole, indole 1-glycosides, 1-N-glucosyl-ascorbigen.

The synthesis of derivatives of ascorbigen – 2-C-[(3-indolyl)methyl]-α-*L*-threo-*L*-glycero-3-hexulofuranosono-1,4-lactone – containing a monosaccharide residue or polyfunctional alcohol residue attached to the nitrogen atom of the indole ring has not so far been described. These compounds are of interest as a new type of hydrophilic derivative of ascorbigen containing simultaneously an N-glycosyl or polyhydroxyalkylindole fragment (the indole analogs of nucleosides) and *L*-ascorbic acid (*L*-AA). As starting compounds we used the previously described 1-N-(2,3,4,5,6-penta-O-acetyl-β-*D*-glucopyranosyl)-3-formylindole (**1**) [1] and 1-N-(1-deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)indole [2].

The usual method for the synthesis of ascorbigens is based on the condensation of 3-hydroxymethylindole or its analogs with *L*-AA under mild conditions [3], where the products from oligomerization of 3-methylindolenine are formed as impurities. Formylindole **1** was reduced by the action of $NaBH_4$ to the corresponding 1-N-(2,3,4,6-tetra-O-acetyl-β-*D*-glucopyranosyl)-3-hydroxymethylindole (**2**) with a yield of 90% (Scheme 1). Compound **2** is more stable than 1-alkyl-3-hydroxymethylindoles, and it can be isolated in the individual state and stored under moderate conditions. However, compound **2** does not enter into condensation with ascorbic acid under the conditions under which 1-alkyl-3-hydroxymethylindoles form ascorbigens (in a citrate–phosphate buffer (CPB) at pH 4.2 in the range of 20-40°C or in ethanol at pH 1-7).

During deacetylation of the derivative **2** by the action of sodium methoxide in methanol 1-N-(β-*D*-glucopyranosyl)-3-hydroxymethylindole (**3**) was obtained. Condensation of the latter with *L*-AA in the CPB at pH 4.2 and 20°C for several days led to the formation of 1-N-(β-*D*-glucopyranosyl)ascorbigen (**4**) with a yield of ~40%. It should be noted that the per-O-acetylated derivatives of 1-N-β-*D*-galactopyranosyl-3-hydroxymethylindole and 1-N-α-*L*-arabinopyranosyl-3-hydroxymethylindole also could not be brought into condensation with ascorbic acid.

The structure of compounds **2** and **4** was confirmed by the data from the NMR spectra. In the glucosides **2** and **4** the constant $J_{1',2'}$ = 8.9 Hz indicates the di-*trans* orientation for protons 1' and 2' and, consequently, the β-*D* configuration and $^4C_1$ conformation for the glucopyranose residue in the obtained compounds. The higher stability and the low reactivity of the hydroxymethylindole **3** in the acidic medium compared with the

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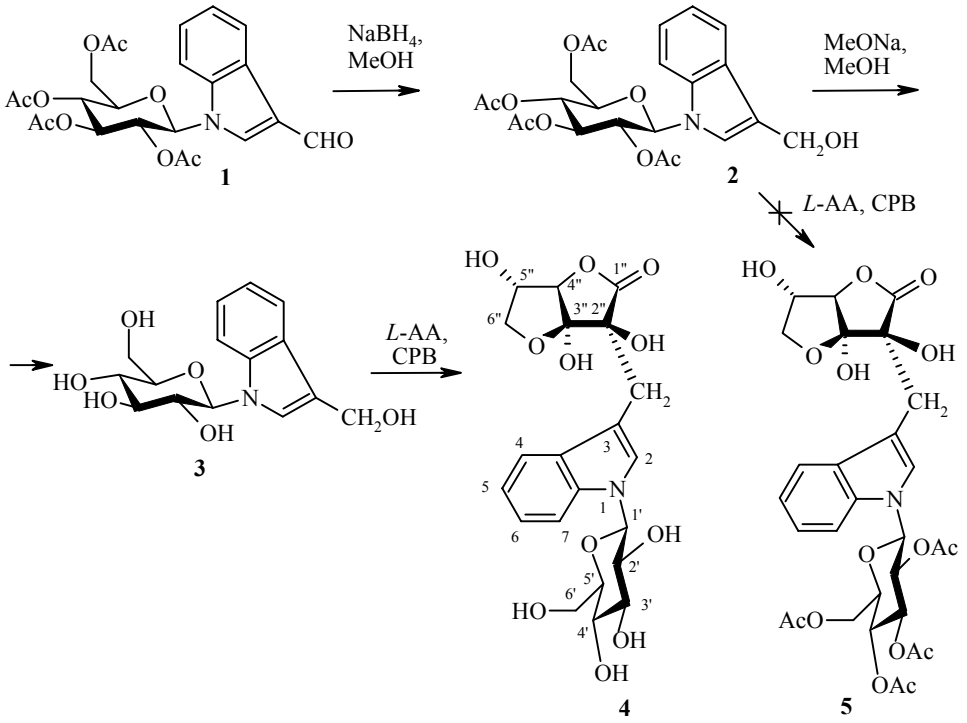
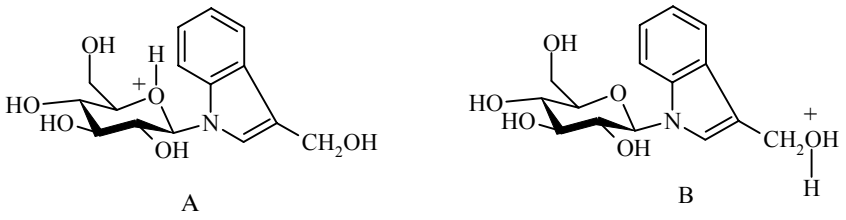
Scheme 1



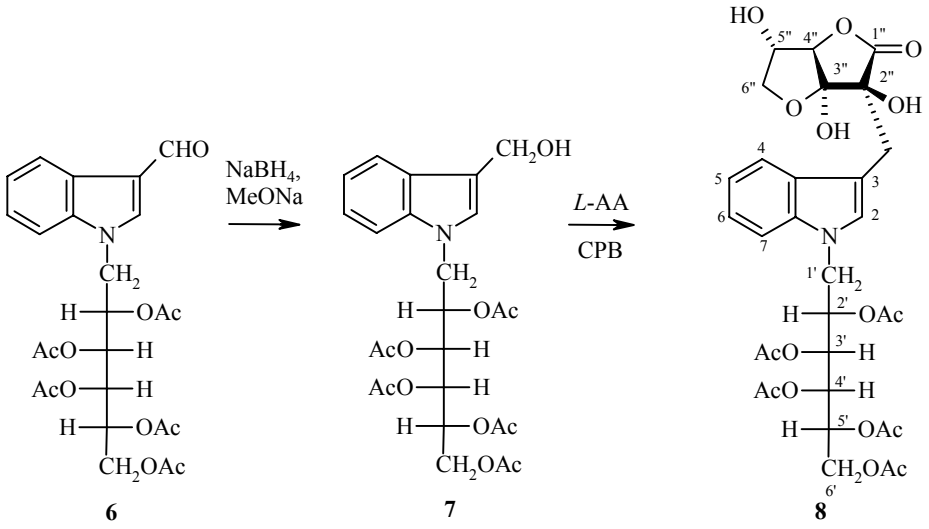
N-unsubstituted or N-alkyl-3-hydroxymethylindole is evidently due to the possibility of alternative protonation of the oxygen of the carbohydrate ring (Scheme 2, structure A) instead of the 3-hydroxymethyl group (Scheme 2, structure B) at the first stage of the reaction, which reduces the electrophilic characteristics of the 3-hydroxymethyl group. The possibility of protonation of the oxygen atom of the carbohydrate ring in 1-glycosylindoles was demonstrated earlier in several examples [4]. In the per-O-acetylated 1-N-glycosyl-3-hydroxymethylindoles the deactivating effect of the carbohydrate ring is strengthened on account of the electron-withdrawing effect of the acetyl groups.

Scheme 2



Derivatives of 3-hydroxymethylindole containing a polyhydroxylated substituent but not having a glycosidic oxygen atom have greater reactivity and are at the same time less stable. The formylation of 1-N-(1-deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)indole gave 1-N-(1-deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)-3-formylindole (**6**), the reduction of which with $NaBH_4$ led to the formation of the corresponding 1-N-(1-deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)-3-hydroxymethylindole (**7**). During the condensation of the latter with *L*-AA in the CPB at pH 4.2 for three days 1-N-(1-deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)ascorbigen (**8**) was formed with a yield of ~40% (Scheme 3).

Scheme 3



EXPERIMENTAL

The NMR spectra were recorded on a Varian 400 VXR instrument at 400 (^{1}H) and 100 MHz (^{13}C). The mass spectra were obtained on a Finnigan MAT 900S instrument by electrospray ionization (ESI MS). As solvents for chromatography we used chloroform–methanol 20:1 (system A), 2:1 (B), 3:1 (C), 25:1 (D), and 10:1 (E). The citrate–phosphate buffer (CPB) with pH 4.2 was prepared from 0.9 g of citric acid and 2 g of Na_2HPO_4 in 100 ml of water. Analytical TLC was conducted on Kieselgel 60 F_{254} plates (Merck). The substances were detected in UV light or were revealed with a 1% solution of *p*-dimethylaminobenzaldehyde in 1 N hydrochloric acid with heat. For column chromatography we used Kieselgel 60 (Merck). Preparative chromatography was conducted on glass plates (20 × 20 cm, 0.5 mm) with Kieselgel 60 $HF_{254+366}$ silica gel (Merck). The melting points were measured on a Buchi SMP-20 instrument and were not corrected. The $[\alpha]_D$ values were determined on a Perkin-Elmer 241 polarimeter.

1-N-(2,3,4,6-Tetra-O-acetyl-β-*D*-glucopyranosyl)-3-hydroxymethylindole (2). To a solution of compound **1** (200 mg, 0.42 mmol) in methanol (10 ml) $NaBH_4$ (32 mg, 0.84 mmol) was added. The reaction mixture was stirred at reduced temperature for 40 min, saturated sodium chloride solution (150 ml) was added, and the product was extracted with ethyl acetate. The extract was washed to a neutral pH with brine and dried over sodium sulfate. After distillation of the solvent 180 mg of the product **2** (yield 90%) was obtained in the form of a syrup; R_f 0.50 (system A). ^{1}H NMR spectrum (deuterochloroform), δ, ppm, *J* (Hz): 7.70 (1H, d, H-6(5)); 7.38 (1H, d, H-5(6)); 7.27 (1H, t, H-7(4)); 7.22 (1H, s, H-2); 7.16 (1H, t, H-4(7)); 5.61 (1H, d, $J_{1',2'}$ = 8.9, H-1'); 5.49 (1H, t, $J_{2',3'}$ = 9, H-2'); 5.43 (1H, t, $J_{3',4'}$ = 8.9, H-3'); 5.25 (1H, t, $J_{4',5'}$ = 9, H-4'); 4.84 (2H, d, $J_{a,b}$ = 5.4, $\underline{CH_2}OH$); 4.28 (1H, dd, $J_{6'a,6'b}$ = 12.4, $J_{6'a,5'}$ = 5.01, H-6'a); 4.20 (1H, dd, $J_{6'b,6'a}$ = 12.4, $J_{6'b,5'}$ = 2.0, H-6'b); 3.96 (1H, m, H-5'); 2.07, 2.06, 2.02, 1.66 (12H, 4s, 4AcO); 1.65 (br. s, combined signal of $CH_2O\underline{H}$ and H_2O in solvent).

1-N-(β-*D*-Glucopyranosyl)-3-hydroxymethylindole (3). 1 N sodium methoxide (0.2 ml) was added to a solution of the carbinol **2** (250 mg, 0.52 mmol) in absolute methanol (10 ml) at room temperature with stirring. After 10-15 min we added water (10 ml) to the reaction mixture and acidified it to pH ~7-8 with 0.1 N hydrochloric acid. The aqueous solution of the product **3** obtained after evaporation of the methanol was used in the next stage; R_f 0.73 (system B).

1-N-(β-*D*-Glucopyranosyl)ascorbigen (4). *L*-AA (0.2 g, 1.2 mmol) in the CPB (50 ml) was added to an aqueous solution of compound **3**, and the mixture was stirred for seven days at room temperature. The reaction mass was then evaporated to dryness and dissolved in absolute methanol. The undissolved part was filtered off, and after evaporation of the filtrate 98 mg of the ascorbigen **4** was isolated by preparative TLC in system B in the form of a yellowish amorphous powder (yield 40%); R_f 0.36 (system B), $[\alpha]_D^{20}$ +1.4° (*c* 0.5, methanol). ^{1}H NMR spectrum (CD_3OD), δ, ppm, *J* (Hz): 7.57 (1H, d, H-7(4)); 7.52 (1H, d, H-4(7)); 7.40 (1H, s, H-2); 7.18 (1H, t, H-5(6)); 7.05 (1H, t, H-6(5)); 5.44 (1H, d, $J_{1',2'}$ = 8.9, H-1'); 4.23 (1H, m, H-5"); 4.18 (1H, dd, $J_{6"a,6"b}$ = 9.8, $J_{6"a,5"}$ = 5.9, H-6"a); 4.11 (1H, s, H-4"); 4.02 (1H, dd, $J_{6"b,6"a}$ = 9.6, $J_{6"b,5"}$ = 3.3, H-6"b); 3.91 (2H, m, H-2', H-6'a); 3.72 (1H, dd, $J_{6'a,6'b}$ = 12.2, $J_{6'b,5'}$ = 5.4, H-6'b); 3.61 (2H, m, H-5', H-3'); 3.52 (1H, t, H-4'); 3.29 (2H, dd, $J_{a,b}$ = 14.3, Ind-3-CH_2). Mass spectrum (ESI), *m/z*: 467.32 $[M]^+$. Found %: N 2.73. $C_{21}H_{25}NO_{11}$. Calculated %: N 3.00.

1-N-(1-Deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)-3-formylindole (6). A solution of phosphorus oxychloride (0.4 ml, 4.3 mmol) in DMF (5 ml) was gradually added to a solution of 1-N-(1-deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)indole (1.0 g, 2 mmol) [2] in absolute DMF (15 ml) at 0°C. The reaction mass was heated at 90-92°C for 1 h. It was then cooled to room temperature, poured onto ice, and neutralized with a solution of potassium carbonate. The precipitate was filtered off, washed with water, and recrystallized from methanol. We obtained 950 mg of the product **6** (yield 90%) in the form of white crystals; mp 179-182°C; R_f 0.55 (system A). ^{1}H NMR spectrum (deuterochloroform), δ, ppm, *J* (Hz): 9.98 (1H, s, CHO_2); 8.25 (1H, d, H-4(7)); 7.73 (1H, s, H-2); 7.52 (1H, d, H-7(4)); 7.36 (1H, t, H-5(6)); 7.31 (1H, t, H-6(5)); 5.41-5.27 (4H, m, 4HC'OAc); 4.25 (1H, dd, $J_{1'a,1'b}$ = 11.7, $J_{1'a,2'}$ = 4.7, H-1'a); 4.18 (2H, m, H-6'a,6'b); 3.78 (1H, dd, $J_{1'b,1'a}$ = 11.7, $J_{1'b,2'}$ = 7.6, H-1'b); 2.23, 2.06, 2.00, 1.95, 1.87 (15H, 5s, 5OAc).

1-N-(Deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)-3-hydroxymethylindole (7). This compound was obtained similarly to compound **2** from compound **6** (812 mg, 1.56 mmol) in the form of a syrup with a yield of 98% (800 mg); R_f 0.50 (system A).

1-N-(1-Deoxy-2,3,4,5,6-penta-O-acetyl-*D*-galactit-1-yl)ascorbigen (8). A solution of the carbinol **7** (800 mg, 1.53 mmol) in ethanol (10 ml) was added to a solution of *L*-AA (537.2 mg, 3.05 mmol) in the CPB (100 ml), and the mixture was stirred at room temperature for three days. The reaction mixture was then extracted with ethyl acetate. The extract was dried over sodium sulfate, and the solvent was evaporated. The product was isolated from the residue by column chromatography (system D). We obtained 400 mg of the ascorbigen **8** (yield 37%) in the form of a white amorphous powder; R_f 0.49 (system E); $[\alpha]_D^{20}$ = +7.4° (*c* 0.5, methanol). ^{1}H NMR spectrum (deuterochloroform), δ, ppm, *J* (Hz): 8.09 (1H, d, H-4(7)); 7.25 (1H, d, H-7(4)); 7.12 (1H, t, H-5(6)); 7.15 (1H, t, H-6(5)); 6.97 (1H, s, H-2); 5.36-5.21 (4H, m, 4CHOAc); 4.24-3.86 (8H, m, H-1',6',4",5",6"); 3.80-3.72 (3H, m, 3OH); 3.26 (2H, dd, $J_{a,b}$ = 17.9, Ind-3-CH_2); 2.15, 2.05, 1.95, 1.92, 1.73 (15H, 5s, 5AcO). ^{13}C NMR spectrum (DMSO-d_6), δ, ppm: 175.75* (1"C); (169.93*, 169.75*, 169.63*, 169.50*, 168.87*) (5 O$\underline{C}$OCH$_3$); 135.89*; 129.09*; 128.35; 120.83; 119.20; 118.50; 109.50; 107.16*; 107.02*; 87.13; 78.68*; 74.11*; 73.80; 68.97; 67.78; 67.56; 67.41; 61.99*; 45.93*; 30.64*; (20.56, 20.51, 20.41, 20.36, 20.19) (5 OCO$\underline{C}$H$_3$). Mass spectrum (ESI), m/z: 679.32 $[M]^+$. Found %: N 1.93. $C_{31}H_{37}NO_{16}$. Calculated %: N 2.06.

REFERENCES

1. M. N. Preobrazhenskaya, V. N. Tolkachev, O. N. Geling, and N. P. Kostyuchenko, *Zh. Org. Khim.*, **10**, 1764 (1974).
2. M. N. Preobrazhenskaya, V. I. Mukhanov, L. D. Manzon, and N. N. Suvorov, *Zh. Org. Khim.*, **8**, 2600 (1972).
3. V. I. Mukhanov, I. V. Yartseva, V. S. Kikot', Yu. Yu. Volodin, I. L. Kustova, N. A. Lesnaya, N. P. Ermakova, Z. P. Sof'ina, and M. N. Preobrazhenskaya, *Bioorgan. Khim.*, **10**, 554 (1984).
4. M. N. Preobrazhenskaya, M. M. Vigdorchik, and N. N. Suvorov, *Khim. Prirod. Soedin.*, **2**, 128 (1968).

*C atoms at which the number of H atoms is even or equal to zero.