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Interaction of L-Ascorbic Acid with DL-*N*-Methyl-β-hydroxytryptamine

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Abstract: The interaction of DL-*N*-methyl-ß-hydroxytryptamine **1** with L-ascorbic acid **2** proceeds through the 2-C alkylation of **2** and the intramolecular acylation of the methylamino group to yield diastereomeric 3-hydroxy-4-(indol-3-yl)-1-methyl-3-(2,3,4-trihydroxybutyryl)-pyrrolid-2-ones **4a** and **5b**.

L-Ascorbic acid interacts with 3-hydroxymethylindole or 4-hydroxybenzyl alcohol and their derivatives to give ascorbigens — 2-*C*-arylmethyl-3-ketohexulofuranosono-1,4-lactones[1,2]. The investigation of the interaction of **2** with multifunctional (indol-3-yl)carbinol derivatives is particularly interesting, as β-hydroxytryptamines and β-hydroxytryptophanes and their metabolites are formed in biochemical reactions catalyzed by (indol-3-yl)alkane α-hydroxylase[3]; they also represent analogues of epinephrine, norepinephrine, and their metabolites. Previously we studied the specificity of the interaction of (indol-3-yl)ethane-1,2-diol with L-ascorbic acid (**2**)[4]. In this paper we report the interaction of DL-*N*-methyl-β-hydroxytryptamine (**1**), an indole analogue of epinephrine, with L-ascorbic acid.

The reaction of **1** with an excess of **2** in citric-phosphate buffer at pH 4.2 yielded a mixture of two products (**4a, 5b**)[5] as a result of 2-C-alkylation of ascorbic acid (to the intermediate **3**) followed by the intramolecular acylation; this mixture was separated by preparative TLC[6]. The presence of lactam cycles in **4a** and **5b** was proved by NMR spectroscopy using HETCOR through ^{13}C-^{1}H long-range couplings experiments: cross-peaks were observed between the N-CH_3 protons and the carbonyl carbon of the lactam cycle. The 3-C - 6-C moiety of ascorbic acid in isomer **4a** represents a cyclic hemiacetal (1'-C - 4'-C), whereas in isomer **5b** it exists as an acyclic trihydroxybutyryl residue. This implies that the cyclic furanose intermediate **4b** is unstable. The presence of the furanose cycle in isomer **4a** was confirmed by the specific chemical shift of 1'-C carbon atom (106.38 ppm) and the polarization transfer from 4'-H to 1'-C in a selective INEPT experiment. Signals of carbonyls of the keto and lactam groups were observed in ^{13}C NMR spectrum of **5b**. Signals 4'-H in **5b** are shifted upfield relative to **4a** (Tables 1 and 2). Among chiral atoms of **4a** and **5b**, there are two (3-C and 4-C of the lactam cycle) whose stereochemistry is not preassigned by the stereochemistry of L-ascorbic acid. The absolute configuration of 3-C atom is determined by the direction of the electrophilic attack of the substituted skatyl cation at the 2-C atom of the L-ascorbic acid lactone ring[1,2]. In all the ascorbic acid 2-C-alkylations reported to date, such an attack was directed from the side opposite to the CHOH-CH_2OH fragment,[7] except for 2-C methylation of **2** yielding a mixture of the 2*S* and 2*R* isomers, which can be explained by the small size of the methyl group[8]. This suggests the *S* configuration at the 3-C of compounds **4a** and **5b**.

The absolute configuration at 4-C of the **4a** and **5b** isomers was determined for their tri-*O*-acetyl derivatives **6a** and **b**. Acetylation of compounds **4a** and **5b** with acetic anhydride in pyridine at

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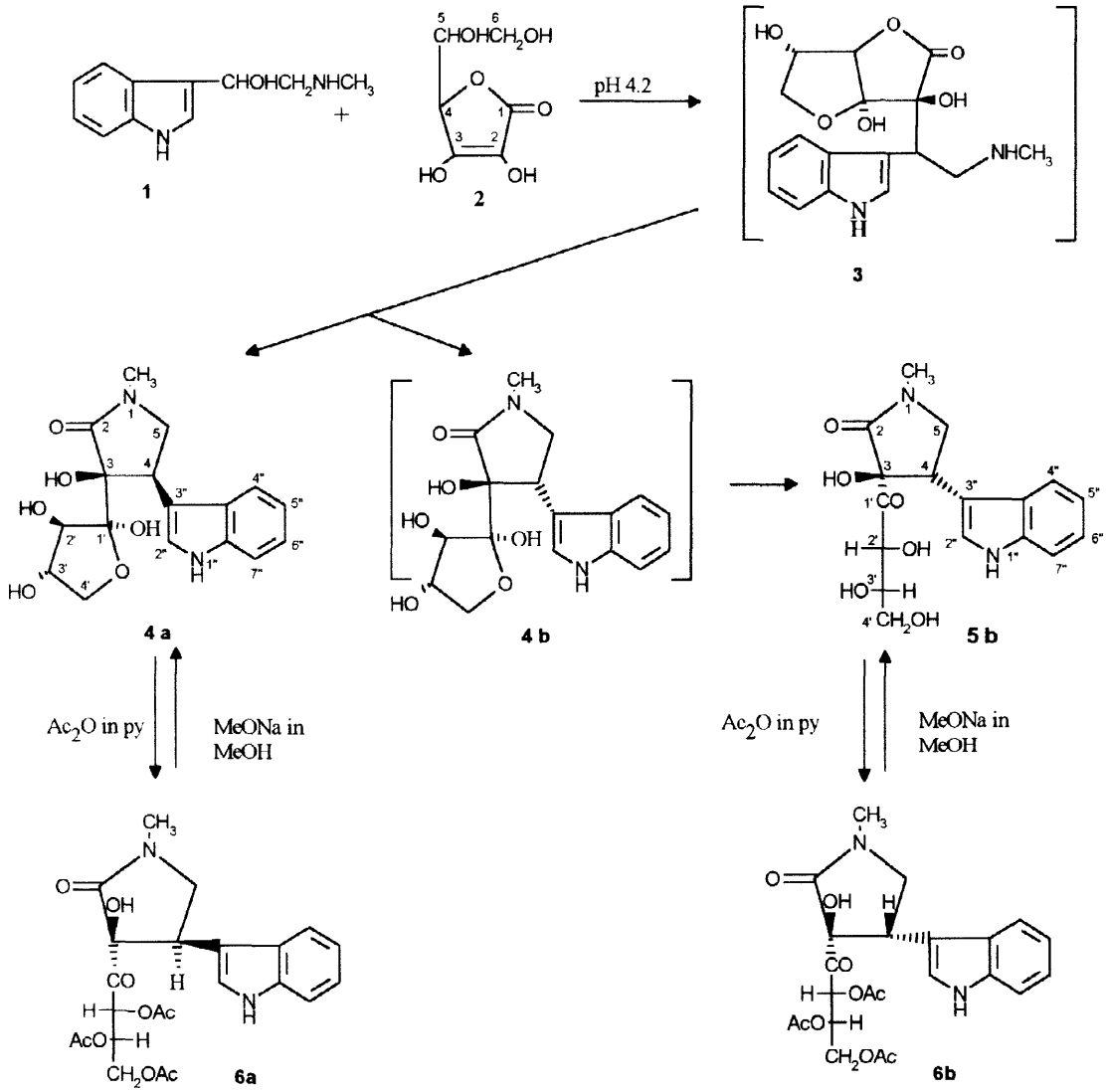
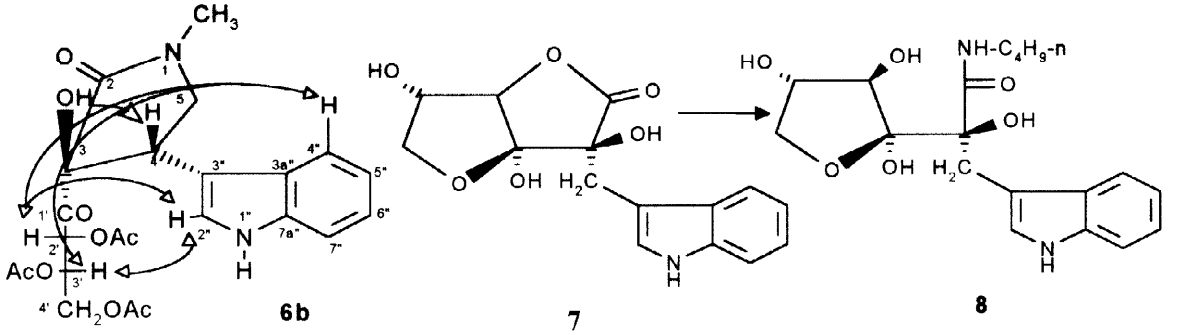
-16°C afforded **6a** and **6b**, respectively,[9] whose deacetylation (MeONa in MeOH) gave again individual **4a** and **5a** (confirmed by HPLC[10]) to show that in the course of the acetylation no racemization occured. ^1H and ^{13}C NMR data showed that the frameworks of **6a** and **6b** are the same as that of **5b** (Table 2). The presence of a sharp signal of the unsubstituted (tertiary) 3-OH group in ^1H NMR spectra of **6a** and **6b** in d_6-DMSO allowed us to use NOE-difference experiments to determine the disposition of the substituents at 4-C. The selective saturation of the 3-OH proton at 6.73 ppm gave rise to 8% signal enhancement of the 4-H multiplet at 3.87 ppm in **6b**, whereas the saturation of the 3-OH proton in **6a** did not affect the 4-H signal. Basing on these data, we assigned to the indole residue at 4-C and the carbohydrate moiety at 3-C a *cis*-arrangement in **6b** and, therefore in **5b**, and a *trans*-arrangement in **6a** and, therefore in **4a**. This led to the conclusion that the absolute configuration at 4-C is *S* in **5b** and *R* in **4a**.

NOE-difference experiments also showed that 2'-H and 3'-H protons of the carbohydrate residue are close to the 2"-H and 4"-H protons of the indole ring in both isomers **6a** and **6b**. This proximity can be accounted for by the lactone cycle deflection from the planar conformation resulting in a *pseudo*-equatorial position of the bulky indole substituent in both isomers, independently of the *cis*- or *trans*-orientation of the substituents at 3-C and 4-C.



The smooth formation of the amide bond in the reaction between **1** and **2** is noteworthy. Earlier, the interaction of ascorbigen with ammonia or primary amines was shown to yield amides of hexulofuranosonic acid, whereas ascorbigen failed to form amides with secondary amines[11]. The interaction of ascorbic acid with amines needs rather drastic conditions and leads to 3-*N*-derivatives[12].



⟶ - NOE connectivities

In our case, intramolecular reaction conditions and thermodynamic stability of the five-membered lactam formed make it possible for the secondary amide to be *N*-acylated by a lactone under mild conditions.

In a model experiment, *N*-butylamide of 3-hexulofuranosonic acid **8**[13] with an intact furanose ring was obtained by incubating ascorbigen **7** (obtained from **2** and 3-hydroxymethylindole[1]) in *N*-butylamine at 25°C.

Table 1. ^{1}H NMR spectra (400 MHz) of compounds **4-6** (aliphatic moiety)

Compound /solvent	4-H	5-H	2'-H	3'-H	4'-H	N-CH$_3$
4a in CD$_3$OD	4.33 J_{5a}8.1 J_{5b}3.7	3.86; 3.46 $J_{a,b}$9.6	4.59 $J_{3'}$3.9	4.11 $J_{4'a}$5.5 $J_{4'b}$4.2	4.06; 3.58 $J_{a,b}$ 9.0	3.00
5b in CD$_3$OD	4.04 J_{5a}10.7 J_{5b} 8.1	3.83; 3.62 $J_{a,b}$9.8	4.00 $J_{3'}$2.2	4.29 $J_{4'}$6.1	3.41	3.02
6a in CDCl$_3$	4.29 J_{5a}.7.4 J_{5b} 3.2	3.86; 3.49 $J_{a,b}$10.0	5.77 $J_{3'}$3.2	5.88 $J_{4'a}$9.5 $J_{4'b}$6.3	4.27; 4.21 $J_{a,b}$11.5	3.00
6b in CDCl$_3$	4.06 J_{5a}11.3 J_{5b} 7.9	3.89; 3.52 $J_{a,b}$9.3	5.08 $J_{3'}$4.1	5.92 $J_{4'a}$5.7 $J_{4'b}$6.6	3.86; 3.80 $J_{a,b}$11.5	3.02

Table 2. ^{13}C NMR spectra (100.6 MHz) of compounds **4-6** *

Aliphatic moiety	**4a** in CD$_3$OD	**5b** in CD$_3$OD	**6a** in CDCl$_3$	**6b** in CDCl$_3$	Indole ring	**4a** in CD$_3$OD	**5b** in CD$_3$OD	**6a** in CDCl$_3$	**6b** in CDCl$_3$
N-CH$_3$	30.30	30.60	30.32	30.35	2"-C	124.94	123.83	123.37	122.70
2-C	175.15	174.27	170.12	168.51	3"-C	113.35	108.91	110.49	108.64
3-C	81.69	88.82	85.07	87.33	4"-C	119.98	119.95	118.92	119.78
4-C	37.27	45.79	39.35	44.86	5"-C	120.19	120.17	120.33	120.03
5-C	55.61	51.19	53.21	50.00	6"-C	122.47	122.72	122.73	122.70
1'-C	106.38	211.23	203.00	202.72	7"-C	112.40	112.16	111.67	111.19
2'-C	78.45	76.27	74.05	74.98	3a"-C	128.56	128.55	126.96	127.32
3'-C	77.64	71.88	68.64	69.23	7a"-C	138.17	137.67	136.35	136.53
4'-C	72.17	63.91	61.57	61.34					

* Signals of three OCOCH$_3$ groups are at 171.26; 170.56; 170.23; 20.79; 20.50 and 19.99 ppm **(6b)** and at 170.39; 170.34; 170.23; 20.73; 20.62 and 20.38 ppm **(6a)**.

The structure of **8**, which was confirmed by ^{13}C NMR, implies that the 2-C alkylation of ascorbic acid precedes the acylation of the methylamino group and that the furanoside residues in compounds **4a** and **8** have an α-configuration.The shift of the tautomeric equilibrium to a furanoside in **4a** and to an acyclic trihydroxybutyryl moiety in **5b, 6a,** and **6b** may be connected with steric hindrances.

RFERENCES AND NOTES

1. Preobrazhenskaya M.N.; Bukhman V.M.; Korolev A.M.; Efimov S.A. *Pharm. Ther.* **1993,** *60,* 301-313.
2. Poss A.; Belter R. *J. Org. Chem.*, **1988,** *53,* 1535-1540.
3. Tsai M.-D.; Floss H.; Rosenfeld H.; Roberts. J. *J. Biol. Chem.* **1979**, *254,* 6437-6443.
4. Preobrazhenskaya M.N.; Lazhko E.I.; Korolev A.M. *Tetrahedron: Asymmetry* **1996**, *70,* 641-644.
5. Mixture **4a+5b**. Yield 83%, HPLC (Diasorb 4x150 column, acetonitrile - 0.01M H_3PO_4, linear gradient 20 ⇒ 50% of acetonitrile): **4a** R_t 10.73 min (65%); **5b** R_t 11.55 min. (34%), Anal. Calcd. for $C_{17}H_{20}N_2O_6 \cdot H_2O$: C 55.73; H 6.05; N 7.65. Found C 55.40; H 5.84; N 7.06.
6. Plates covered with Kieselgel HF_{254} (Merck) were used for preparative TLC. 1-Methyl-3(*S*)-hydroxy-3-[(2*R*, 3*S*)-α-glycerotetrofuranose-1-yl)-4(*R*)-(indol-3-yl)pyrrolid-2-one (**4a**): R_f 0.55 in $CHCl_3$-MeOH 5:1, $[\alpha]^{21}_D$ +20.7° (c 0.5, MeOH), time-of-flight MS, *m/z:* 348 $[M]^+$. 1-Methyl-3(*S*)-hydroxy-3-[(2*R*, 3*S*)-4-trihydroxybutyryl]-4(*S*)-(indol-3-yl)-pyrrolid-2-one (**5b**): R_f 0.44 in $CHCl_3$-MeOH 5:1, $[\alpha]^{21}_D$ -93.1° (c 0.5, MeOH), time-of-flight MS, *m/z:* 348 $[M]^+$.
7. Korolev A.M.; Lazhko E.I.; Yartseva I.V.; Plykhtyak I.L.; Alexandrova L.G.; Rosynov B.V.; Preobrazhenskaya M.N. *Soviet Journal of Bioorg. Chem. (Engl.)* **1991,** *17,* 548-554.
8. Poss A.; Belter R. *Tetrahedron: Asymmetry* **1993, *4,*** 169-172.
9. 1-Methyl-3(*S*)-hydroxy-3-[(2*R*, 3*S*)-4-triacetoxybutyryl]-4(*R*)-(indol-3-yl)pyrrolid-2-one (**6a**): R_f 0.57 in $CHCl_3$-MeOH 20:1, $[\alpha]^{21}_D$ -22.5° (c 1, MeOH), FAB MS, *m/z:* 475 (100%) $[MH]^+$; 229 (75%) $[M - CO(CHOAc)_3H]^+$. 1-Methyl-3(*S*)-hydroxy-3-[(2*R*, 3*S*)-4-triacetoxybutyryl]- 4(*S*)-(indol-3-yl)pyrrolid-2-one (**6b**): R_f 0.71 in $CHCl_3$-MeOH, 20:1, $[\alpha]^{21}_D$ -111.6° (c 1; MeOH), FAB MS *m/z:* 475 (71%) $[MH]^+$; 229 (100%) $[M - CO(CHOAc)_3H]^+$. Mixture **6a+6b**. Yield 59%, Anal. Calcd. for $C_{17}H_{20}N_2O_6 \cdot 2H_2O$: C 54.11; H 5.92; N 5.49. Found C 54.30; H 5.47; N 5.11.
10. HPLC (Diasorb 4x150 column, acetonitrile - 0.01 M H_3PO_4, linear gradient 20 ⇒ 50% of acetonitrile): **6a** R_t 12.51 min; **6b** R_t 9.15 min.
11. Plykhtyak I.L.; Yartseva I.V.; Khan Z.-E.; Preobrazhenskaya M.N. *Bioorg. Khim. (Russ.)* **1988**, *14,* 1437-1443. *CA* **1988**, *109,* 190726f.
12. Picschetsrieder M.; Larish B.; Müller U.; Severin T. *J. Agr. Food Chem.* **1995,** *43,* 3004-3006.
13. 2-C-[(Indol-3-yl)methyl]-α-L-*xylo*-3-hexulofuranosonic acid *N*-butylamide (**8**). $[\alpha]^{21}_D$ -0.7° (c 1; MeOH), FAB Ms, *m/z:* 379 $[MH]^+$. ^1H NMR (CD_3OD, carbohydrate moiety): δ 4.38, 4-H, $J_{4,5}$ 5.5; 4.31, H-5, $J_{5,6a}$ 6.4, $J_{5,6b}$ 5.1; 4.11, H-6a, $J_{6a,6b}$ 9.1; 3.61, H-6b 3.55, 3.20, CH_2", J_{AB} 14.6; ^{13}C NMR(CD_3OD): δ 177.07, C-1; 137.78, C-7'a; 129.60, C-3'a; 125.52, C-2'; 122.04, C-6'; 120.37, C-5'; 119.46, C-4'; 111.95, C-7'; 109.71, 106.92, C-3', C-3; 79.79, C-2; 78.50, C-4; 77.79 C-5; 71.41, C-6; 39.77, 31.74, 20.62, C_4H_9; 31.35, CH_2".