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SYNTHESIS OF A NEW INHIBITOR OF α-FUCOSIDASE

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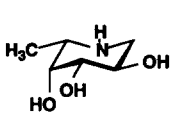
Abstract: A new fucose-type iminosugar in which the nitrogen is placed in the anomeric position was synthesized from D-ribose and was shown to be a potent inhibitor of α-fucosidase (Ki = 8.4 μM).

Deoxyfuconojirimycin (**1**), one of the deoxynojirimycin analogues, is a potent inhibitor of α-L-fucosidases,[1] and its *N*-alkyl derivatives have been demonstrated to inhibit HIV cytopathicity.[2] These iminosugars have, therefore, been considered as a new class of drugs for use in a variety of infectious diseases and diabetes in which carbohydrates play a key role.[3-5] In the course of our study on developing more potent inhibitor of glycosidases, we proposed a new type of iminosugar in which a nitrogen atom is placed in the anomeric position. Several new iminosugars of the glucose- and galactose-type (**2**) have been synthesized by us[6-8] and others[9] and have proven to be more potent inhibitors of β-glycosidases than are the conventional deoxynojirimycin-type iminosugars. In an extension of this work, we herein report the stereoselective synthesis of a new fucose-type iminosugar (**3**) and its analogues (**4-8**) and an evaluation of biological activity as inhibitors of various glycosidases.



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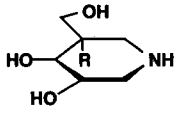
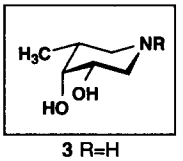
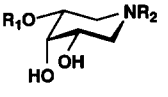


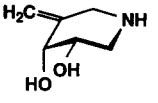
2 R=H or OH



3 R=H
4 R=*n*-Butyl



5 R_1=H, R_2=H
6 R_1=H, R_2=*n*-Butyl
7 R_1=CH_3, R_2=H



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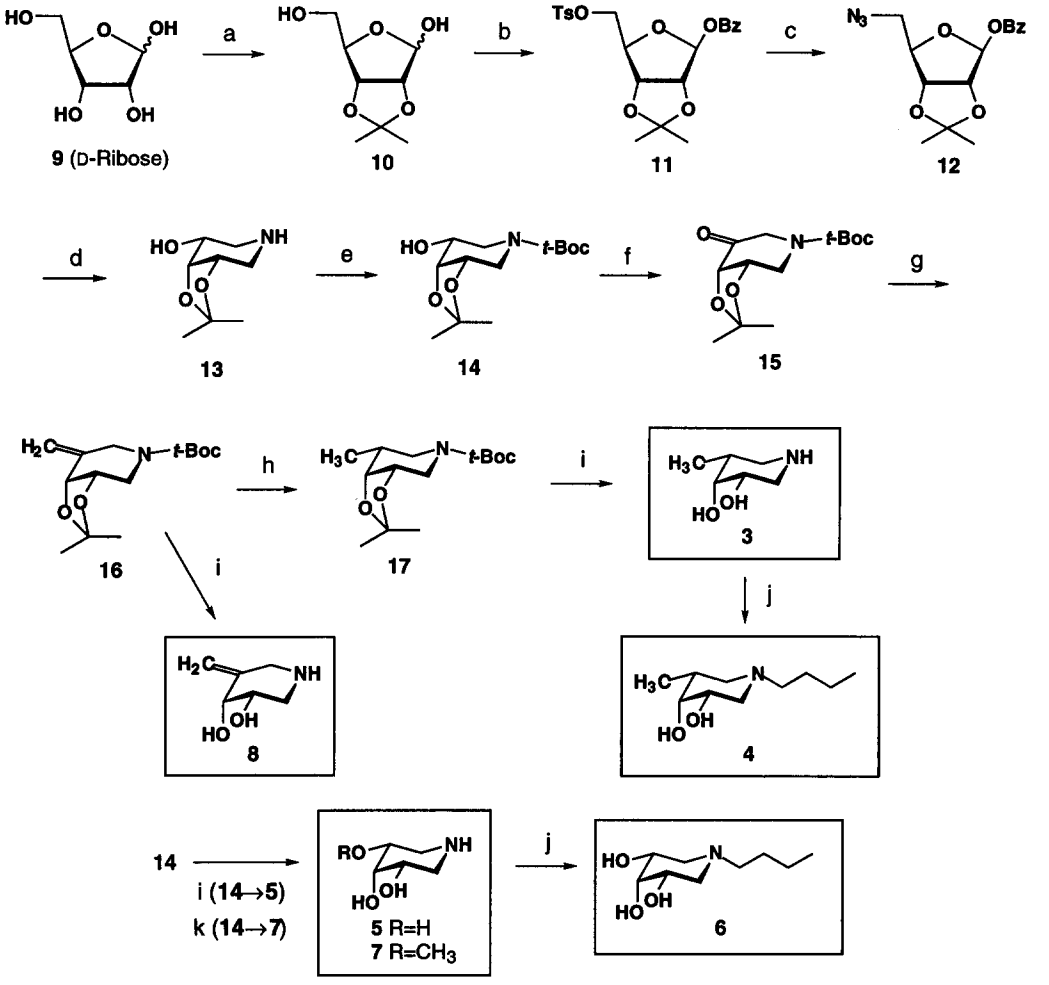
D-Ribose (**9**) was converted to a 2,3-*O*-isopropylidene derivative **10**,[10] which was subsequently treated with TsCl and BzCl to afford **11** as a single isomer in 48% overall yield. Treatment of **11** with NaN_3 gave the azido derivative (**12**) in 91% yield. The benzoyl group of **12** was removed with NaOMe and the subsequent intramolecular reductive amination with H_2-$Pd(OH)_2$ furnished a piperidine derivative **13** in 75% overall yield. To introduce a *C*-5 methyl group of L-fucose structure, we took advantage of the *cis*-fused bicyclic structure of **13**. The amino group of **13** was first protected with BocON[11] to give **14**. Swern oxidation[12] of **14** gave **15**, which was subjected to Wittig methylenation to give **16** in 63% overall yield. Catalytic hydrogenation of **16** preferentially occurred from the less hindered β-face to afford **17** exclusively in 82% yield.[13] Treatment of **17** with 1*N* HCl simultaneously removed both the 3,4-*O*-isopropylidene and the *t*-Boc group to furnish the desired fucose-type 1-*N*-iminosugar **3**. The overall yield was 12% from D-ribose in 11 steps.

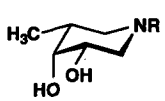


Scheme 1. Synthesis of fucose-type 1-*N*-iminosugar (3) and its analogues (4-8). Reagents and conditions: (a)2,2-Dimethoxypropane/TsOH/acetone/rt/12 h; (b)(i)TsCl/Pyr/0-5 °C/10 h, (ii)BzCl/Pyr/0-5 °C/1 h (48% in 3 steps); (c)NaN_3/DMF/60-65 °C/4 h (91%); (d)(i)NaOMe/MeOH/rt/10 min; (ii)H_2/Pd(OH)$_2$/MeOH-H_2O/rt/12 h (75% in 2 steps); (e)BocON/Et_3N/H_2O-dioxane/rt/8 h (73%); (f)$(COCl)_2$/ DMSO/CH_2Cl_2/-70 °C/30 min then Et_3N/-70 °C to 0 °C/30 min (80%); (g)$CH_3PPh_3^+Br^-$/*n*-BuLi/DME/0 °C to rt/8 h (77%); (h)H_2/Pd-C/MeOH/rt/5 h (82%); (i)1*N* HCl/rt/12 h (90-95%); (j)*n*-Butylaldehyde/BH_3·Pyr/MeOH-phosphate buffer (pH 7.2)/rt/12 h (80%); (k)MeI/NaH/DMF/0 °C to rt/12 h then 1*N* HCl/rt/12 h (90%).

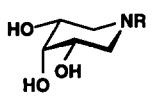
We also synthesized the structurally related fucose-type iminosugar analogues **4**, **5**, **6**, **7**, and **8**. For *N*-alkylation, **3** was treated with *n*-butylaldehyde and BH_3•Pyr in MeOH-phosphate buffer (pH 7.2)[14] to give *N*-butyl iminosugar **4** in 80% yield. For preparation of a triol iminosugar, **14** was deprotected with 1*N* HCl to afford **5** and *N*-butylation of **5** was performed in the same manner described above to give the *N*-butyl derivative (**6**). An iminosugar (**7**) with a *C*-5 O-CH_3 derivative instead of a methyl group was synthesized by methylation

Table 1. Inhibition of glycosidases by iminosugars (3-8).

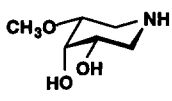
Glycosidase	IC_{50} (μM) for Inhibition by Compound					
	3	4	5	6	7	8
α-Fucosidase from bovine kidney (Sigma F 5884)	26	270	>1000	>1000	>1000	>1000
α-Glucosidase from yeast (Sigma G 7256)	>1000	>1000	>1000	>1000	>1000	>1000
β-Glucosidase from almonds (Sigma G 4511)	500	80	8.8	530	>1000	>1000
α-Galactosidase from green coffee beans (Sigma G 8507)	>1000	>1000	40	725	>1000	>1000
β-Galactosidase from Aspergillus oryzae (Sigma G 7256)	>1000	>1000	40	43	>1000	>1000
α-Mannosidase from jack beans (Sigma M 7257)	>1000	>1000	360	>1000	>1000	>1000



3 R=H
4 R=*n*-Butyl



5 R=H
6 R=*n*-Butyl



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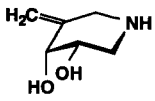

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of **14** with MeI and NaH. Another iminosugar derivative (**8**) with a *C*-5 *exo*-methylene group was prepared by deprotection of **16**.

The inhibitory potency of these iminosugars (**3-8**) was examined[15] against several glycosidases (Table 1). As we anticipated, among the iminosugars tested, **3** was the most potent inhibitor of α-fucosidase from bovine kidney, showing a *Ki* of 8.4 μM (IC_{50} of 26 μM) at pH 6.8; its *N*-butyl analogue **4** also inhibited the enzyme with an IC_{50} of 260 μM. All other analogues (**5-8**) showed no significant inhibition at 1 mM concentration. Contrary to our expectations, **8** did not inhibit the α-fucosidase although its half-chair conformation has been thought to mimic the transition state of the glycosidic bond-cleaving reaction. This lack of inhibitory activity suggests that the enzyme strictly recognizes the substituent at *C*-5.

It was also surprising that the trihydroxypiperidine **5**, in which the methyl group at *C*-5 was replaced with an OH group potently inhibited β-glucosidase with an IC_{50} value of 8.8 μM. Moreover, **5** and its *N*-butyl derivative **6** inhibited both α- and β-galactosidase with an IC_{50} of 40 μM, suggesting that the galactosidases recognize the 3- and 4-OH groups of **5** and **6** as those of a galactoside. To our knowledge, this is the first demonstration of the inhibitory potency of **5** (the *meso*-type 3,4,5-trihydroxypiperidine) against glycosidases; the synthesis and biological activities of other stereoisomers of 3,4,5-trihydroxypiperidine have been reported by others.[16]

In summary, we have synthesized a new fucose-type 1-*N*-iminosugar (**3**) from D-ribose with (i) Wittig methylenation and (ii) stereoselective hydrogenation as the key steps and have shown this iminosugar to be a potent inhibitor of the α-fucosidase.

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References and Notes:

1. Winchester, B.; Barker, C.; Baines, S.; Jacob, G. S.; Namgoong, S. K.; Fleet, G. *Biochem. J.* **1990**, *265*, 277. and references cited therein.
2. Fleet, G. W. J.; Karpas, A.; Dwek, R. A.; Fellows, L. E.; Tyms, A. S.; Petursson, S.; Namgoong, S. K.; Ramsden, N. G.; Smith, P. W.; Son, J.-C.; Wilson, F.; Witty, D. R.; Jacob, G. S.; Rademacher, T.W. *FEBS Lett.* **1988**, *237*, 128.
3. Winchester, B.; Fleet, G. W. J. *Glycobiology* **1992**, *2*, 199.
4. Legler, G. *Adv. Carbohydr. Chem. Biochem.* **1990**, *48*, 319.
5. Look, G. C.; Fotsh, C. H.; Wong, C.-H. *Acc. Chem. Res.* **1993**, *26*, 182.
6. Ichikawa, M.; Ichikawa, Y. *Bioorg. Med. Chem.* **1995**, *3*,161.
7. Ichikawa, M.; Igarashi, Y.; Ichikawa, Y. *Tetrahedron Lett.* **1995**, *36*, 1767.
8. Ichikawa, Y.; Igarashi, Y. *Tetrahedron Lett.* **1995**, *36*, 4585.
9. Jespersen, T. M.; Dong, W.; Sierks, M. R.; Skrydstrup, T.; Lundt, I.; Bols, M. *Angew. Chem. Int. Ed. Engl.* **1994**, *33*, 1778.
10. Kaskar, B.; Heise, G. L.; Michalak, R. S.; Vishnuvajjala, B. R. *Synthesis* **1990**, 1031.
11. Itoh, M.; Hagiwara, D.; Kamiya, T. *Bull. Chem. Soc. Jpn.* **1977**, *50*, 718.
12. Mancuso, A. J.; Huang, S. -L.; Swern, D. *J. Org. Chem.* **1978**, *43*, 2480.
13. The stereochemistry of *H*-5 was determined as the axial configuration by the ^{1}H NMR data of **3**. The coupling constants were $J_{5,6eq}$ 3.5 Hz and $J_{5,6ax}$ 12.4 Hz.
14. Yoshida, T; Lee, Y.-C. *Carbohydr. Res.* **1994**, *251*, 175.
15. Procedures for the inhibition assay of iminosugars, see: Halvorson, H. *Methods Enzymol.* **1966**, *8*, 559; Dale, M. P.; Ensley, H. E.; Kern, K.; Sastry, K. A. R.; Byers, L. D. *Biochemistry* **1985**, *24*, 3530.
16. (a) Bernotas, R. C.; Papandreou, G.; Urbach, J.; Ganem, B. *Tetrahedron Lett.* **1990**, *31*, 3393.
    (b) McCaig, A. E.; Chomier, B.; Wightman, R. H. *J. Carbohydr. Chem.* **1994**, *13*, 397.
    (c) Legler, G.; Stütz, A. E.; Immich, H. *Carbohydr. Res.* **1995**, *272*, 17.
17. Selected ^{1}H and ^{13}C NMR data for the new compounds.

    1-*O*-Benzoyl-2,3-*O*-isopropylidene-5-*O*-*p*-toluenesulfonyl-β-D-ribose 11: ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.35 and 1.57 (s, 3H each, CH_3 of isopropylidene group), 2.39 (s, 3H, $ArCH_3$), 4.05 (dd, 1H, *J* 6.9, 10.3 Hz, H-5a), 4.10 (dd, 1H, *J* 5.1, 10.3 Hz, H-5b), 4.48 (dd, 1H, *J* 5.1, 6.9 Hz, H-4), 4.87 (s, 2H, H-2 and -3), 6.43 (s, 1H, H-1), 7.22 (d, 2H, *J* 8.2 Hz, aromatic H of tosyl group), 7.48 (dd, 2H, *J* 7.7 Hz, aromatic H of benzoyl group), 7.62 (dd, 1H, *J* 7.3 Hz, aromatic H of benzoyl group), 7.66 (d, 2H, *J* 8.2 Hz, aromatic H of tosyl group), 8.00 (d, 2H, *J* 7.3 Hz, aromatic H of benzoyl group); ^{13}C NMR (75 MHz, $CDCl_3$): δ 21.47, 24.87, 26.33, 68.84, 81.09, 84.92, 85.09, 102.73, 113.32, 127.78, 128.45, 129.73, 129.80, 133.48, 145.04, 164.61.

5-Azido-1-*O*-benzoyl-5-deoxy-2,3-*O*-isopropylidene-β-D-ribose 12: ^{1}H NMR (300 MHz, $CDCl_3$) 1.37 and 1.59 (s, 3H each, CH_3 of isopropylidene group), 3.31 (dd, 1H, *J* 6.8, 12.7 Hz, H-5a), 3.55 (dd, 1H, *J* 7.0, 12.7 Hz, H-5b), 4.48 (dt, 1H, *J* 0.93, 6.9 Hz, H-4), 4.76 (dd, 1H, *J* 0.93, 6.0 Hz, H-3), 4.90 (d, 1H, *J* 6.0 Hz, H-2), 7.46 (dd, 2H, *J* 7.4, 7.9 Hz, *m*-aromatic H), 7.60 (tt, 1H, *J* 1.3, 7.4 Hz, *p*-aromatic H), 8.01 (dd, 2H, *J* 1.3, 7.9 Hz, *o*-aromatic H); ^{13}C NMR (75 MHz, $CDCl_3$) 24.9, 26.4, 53.0, 81.7, 85.2, 86.6, 103.1, 113.4, 128.5, 129.6, 133.5, 164.7.

(3*S*, 4*R*, 5*R*)-3,4-*O*-Isopropylidene-3,4,5-trihydroxypiperidine 13: ^{1}H NMR (300 MHz, $CDCl_3$) 1.38, 1.55 (s, 3H each, CH_3 of isopropylidene group), 2.74-3.15 (m, 6H, H-2, H-6, O<u>H</u> and N<u>H</u>), 3.79 (m, 1H), 4.15 (m, 1H), 4.27 (m, 1H); ^{13}C NMR (75 MHz, $CDCl_3$) 25.5, 26.9, 46.6, 47.2, 64.9, 71.8, 74.2, 108.7.

(3*S*, 4*R*, 5*R*)-1-*N*-*t*-Butoxycarbonyl-3,4-*O*-isopropylidene-3,4,5-trihydroxypiperidine 14: ^{1}H NMR (300 MHz, $CDCl_3$) 1.38 and 1.51 (s, 3H each, CH_3 of isopropylidene group), 1.46 (s, 9H, *t*-Bu of *t*-Boc group), 2.79 (m, 1H), 3.18 (dd, 1H, *J* 9.9, 12.2 Hz), 3.45 (dd, 1H, *J* 4.0, 14.0 Hz), 3.56 (m, 2H), 3.90 (m, 1H), 4.38 (m, 2H); ^{13}C NMR (75 MHz, $CDCl_3$) 24.7, 26.6, 28.3, 65.6, 72.2, 79.8, 109.3.

(4*S*, 5*S*)-1-*N*-*t*-Butoxycarbonyl-4,5-*O*-isopropylidene-4,5-dihydroxy-3-piperidone 15: ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.40 and 1.47 (s, 3H each, CH_3 of isopropylidene group), 1.45 (s, 9H, *t*-Bu of *t*-Boc group), 3.51 (m, 1H, H-6), 3.60 (m, 1H, H-6), 4.65 (m, 1H, H-2), 4.83 (m, 2H, H-4 and H-5), 4.95 (m, 1H, H-2); ^{13}C NMR (75 MHz, $CDCl_3$) δ 24.9, 26.3, 80.5, 111.2, 154.2, 202.8.

(3*S*, 4*R*)-1-*N*-*t*-Butoxycarbonyl-3,4-*O*-isopropylidene-5-methylene-3,4-dihydroxy-piperidine 16: ^{1}H NMR (300 MHz, CD_3OD) δ 1.34 and 1.40 (s, 3H each, CH_3 of isopropylidene group), 1.46 (s, 9H, *t*-Bu of *t*-Boc group), 2.94 (m, 1H, H-2), 3.74 (m, 1H, H-6), 3.89 (dd, 1H, *J* 2.4, 14.4 Hz, H-2), 4.24-4.36 (m, 2H, H-3 and -6), 4.71 (d, 1H, *J* 7.5 Hz, H-4), 5.20 (s, 1H, C=CH_2), 5.27 (s, 1H, C=CH_2); ^{13}C NMR (75 MHz, CD_3OD) δ 24.94, 27.05, 28.76, 76.05, 77.33, 80.87, 110.25, 141.24, 156.91.

(3*S*, 4*R*, 5*R*)-1-*N*-*t*-Butoxycarbonyl-3,4-*O*-isopropylidene-5-methyl-3,4-dihydroxy-piperidine 17: ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.05 (d, 3H, *J* 6.9 Hz, *C*-5 C<u>H</u>$_3$), 1.34 and 1.44 (s, 3H each, CH_3 of isopropylidene group), 1.46 (s, 9H, *t*-Bu of *t*-Boc group), 1.90 (m, 1H, H-5), 2.99 (t, 1H, *J* 12.3 Hz), 3.34 (m, 2H), 3.68 (m, 1H), 4.20 (dd, 1H, *J* 2.5, 6.9 Hz), 4.27 (br.s, 1H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 24.7, 26.5, 28.4, 30.7, 72.7, 74.9, 108.2, 164.1.

(3*S*, 4*R*, 5*R*)-3,4-Dihydroxy-5-methylpiperidine 3 (HCl salt): ^{1}H NMR (300 MHz, D_2O) δ 0.93 (d, 3H, *J* 6.75 Hz, *C*-5 C<u>H</u>$_3$), 1.96 (m, 1H, H-5), 2.73 (dd, 1H, *J* 12.6 Hz, H-2ax or 6ax), 2.91 (dd, 1H, *J* 11.7 Hz, H-6ax or 2ax), 3.01 (dd, 1H, *J* 3.65, 12.6 Hz, H-2eq or 6eq), 3.17 (dd, 1H, *J* 4.22, 11.3 Hz,

H-6eq or 2eq), 3.85 (s, 1H, H-4), 3.87 (dm, 1H, *J* 12 Hz, H-3); ^{13}C NMR (75 MHz, D_2O) δ 13.89, 31.88, 41.85, 43.22, 66.25, 69.70.

(3*S*, 4*R*, 5*R*)-1-*N*-*n*-Butyl-3,4-dihydroxy-5-methylpiperidine 4 (HCl salt): ^{1}H NMR (300 MHz, D_2O) δ 0.83 (t, 3H, *J* 7.4 Hz, -$CH_2C\underline{H}_3$ of *N*-Bu), 0.93 (d, 3H, *J* 6.9 Hz, *C*-5 CH_3), 1.29 (tq, 2H, *J* 7.4 Hz, -$C\underline{H}_2CH_3$ of *N*-Bu), 1.63 (tt, 2H, *J* 7.4, 8.0 Hz, -$C\underline{H}_2CH_2CH_3$ of *N*-Bu), 1.97 (m, 1H, H-5), 2.70 (dd, 1H, *J* 12.4 Hz, H-6ax), 2.87 (dd, 1H, *J* 11.6 Hz, H-2ax), 3.08 (dd, 1H, *J* 8.1 Hz, N-CH_2-), 3.14 (dd, 1H, *J* 3.5, 12.4 Hz, H-6eq), 3.27 (dd, 1H, *J* 4.2, 11.7 Hz, H-2eq), 3.82 (s, 1H, H-4), 3.87 (dd, 1H, *J* 4.1, 11.3 Hz, H-3); ^{13}C NMR (75 MHz, D_2O) δ 15.40, 15.95, 21.82, 28.03, 34.26, 52.17, 53.93, 59.77, 68.70, 71.36.

1,5-Dideoxy-1,5-iminoribitol 5 (HCl salt): ^{1}H NMR (300 MHz, D_2O) δ 3.23 (m, 4H, H-2 and -6), 4.03 (dd, 1H, *J* 2.7 Hz, H-4), 4.08 (m, 2H, H-3 and -5); ^{13}C NMR (75 MHz, D_2O) δ 46.71, 67.93, 70.73.

***N*-*n*-Butyl-1,5-dideoxy-1,5-iminoribitol 6** (free amine): ^{1}H NMR (300 MHz, D_2O) δ 0.86 (t, 3H, *J* 7.3 Hz, -$C\underline{H}_3$ of *N*-Bu), 1.25 (tq, 2H, *J* 7.3 Hz, -$C\underline{H}_2CH_3$ of *N*-Bu), 1.44 (tt, 2H, *J* 7.3, 8.0 Hz, -$C\underline{H}_2CH_2CH_3$ of *N*-Bu), 2.20 (dd, 2H, *J* 10.6, 10.7 Hz, H-2ax and -6ax), 2.41 (t, 2H, *J* 8.0 Hz, N-$C\underline{H}_2$), 2.67 (dd, 2H, *J* 3.8, 10.7 Hz, H-2eq and -6eq), 3.72 (dd, 2H, *J* 3.0, 4.0, 10.6 Hz, H-3 and -5), 3.96 (s, 1H, H-4).

(3*S*, 4*S*, 5*R*)-3,4-Dihydroxy-5-methoxypiperidine 7 (HCl salt): ^{1}H NMR (300 MHz, D_2O) δ 3.20 (m, 3H, H-2ax or -6ax, H-2eq and -6eq), 3.29 (dd, 1H, *J* 7.4, 12.9 Hz, H-6ax or -2ax), 3.38 (s, 3H, -$OC\underline{H}_3$), 3.70 (m, 1H, H-5), 4.01 (m, 1H, H-3), 4.10 (dd, 1H, *J* 2.9 Hz, H-4); ^{13}C NMR (75 MHz, D_2O) δ 44.00, 47.30, 59.57, 67.81, 68.94, 77.16.

(3*S*, 4*R*)-3,4-Dihydroxy-5-methylenepiperidine 8 (HCl salt): ^{1}H NMR (300 MHz, D_2O) δ 3.28 (dd, 1H, *J* 2.1, 13.2 Hz, H-2), 3.35 (ddd, 1H, *J* 1.2, 4.8, 13.3 Hz, H-2), 3.64 (d, 1H, *J* 13.5 Hz, H-6a), 3.83 (d, 1H, *J* 13.5 Hz, H-6b), 4.12 (m, 1H, H-3), 4.37 (s, 1H, H-4), 5.31 (s, 1H, *C*-5 C=$C\underline{H}_2$), 5.34 (s, 1H, C=$C\underline{H}_2$); ^{13}C NMR (75 MHz, D_2O) δ 47.36, 47.84, 66.73, 70.45, 116.89, 136.94.

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