A Convenient Route to an Acetylenic C_{35} Hopanoid and the Absolute Configuration of the Side-chain of Aminobacteriohopanetriol

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The absolute configuration of the side-chain of aminobacteriohopanetriol has been determined as (32*R*, 33*R*, 34*S*) by n.m.r. spectroscopy of its 32,34-*O*-isopropylidene-*N*-acetyl derivative and by chemical correlation with bacteriohopanetetrol yielding an acetylenic bacteriohopane intermediate of potential synthetic value.

Among the numerous types of bacteriohopane derivatives recently isolated from bacteria,[1] aminobacteriohopanetriol (**1**) is a quite widespread triterpenoid.[2] Since we recently described the full stereochemistry of another common hopanoid, bacteriohopanetetrol (**9**), we tried to correlate these two hopanoids in order to determine the absolute configuration of aminotriol (**1**) which might give interesting clues to possible biogenetic pathways for the biosynthesis of the bacteriohopane skeleton.

All attempts to convert directly by several diazotisation

procedures aminotriol (**1**), isolated from *Rhodopseudomonas palustris*[3] or from a *Streptomyces* sp., into tetrol (**9**) failed, so the tetra-acetate of (**1**) was treated according to White[4] with sodium nitrite in acetic acid to give the *N*-nitroso compound (**5**) (yield 90%)† which was further heated at 60 °C in dry 1,4-dioxane in the presence of Na_2CO_3 to give the acetylenic hopanoid (**6**)† instead of the expected tetra-acetate (**8**) (Scheme 1). Few examples of such reactions of 1,2-bifunctional compounds giving acetylenic hydrocarbons have previously been documented.[5] Hydrogenation of (**6**) in the presence of Lindlar catalyst[6] afforded quantitatively the alkene (**7**)† which was oxidized with OsO_4 using Kishi's conditions[7] giving after acetylation a 4:1 mixture of the two C-34 stereoisomers (**8**) and (**10**) of the tetra-acetate of bacteriohopanetetrol (yield 90%). The major compound (**8**) was identical with the tetra-acetate of bacteriohopanetetrol isolated from *Methylobacterium organophilum* or obtained by synthesis from 30-(5′-adenosyl)hopane.[3] The configurations of the asymmetric centres of the side-chain of aminotriol (**1**) are thus (32*R*, 33*R*, 34*S*) according to the stereoselectivity of the osmylation reaction induced by the hydroxy group of the asymmetric centre at C-33.[7]

Confirmation of this configuration was also obtained by another route. The tetra-acetate of (**1**) was quantitatively converted into its *N*-monoacetyl derivative (**2**) by stirring in MeOH–tetrahydrofuran (THF) in the presence of basic Amberlyst A-26.[8] Treatment of (**2**) with acetone in the presence of anhydrous $FeCl_3$ gave the 32,34-*O*-isopropylidene derivative (**3**) (yield 30%)† along with starting material and two other acetonides. Analysis of its 400 MHz ^{1}H n.m.r. spectrum showed that the 33-H proton signal at δ 2.91 for (**3**) [shifted downfield to δ 4.50 for (**4**)† after acetylation] was a triplet (*J* 9.5 Hz) corresponding to a chair conformation of the six membered acetonide ring and to two *trans* diaxial couplings with 32-H and 34-H, in full accordance with the configuration determined by the former method.

† Satisfactory analytical data {m.p., $[\alpha]_D$, i.r., ^{1}H n.m.r. (200 or 400 MHz), ^{13}C n.m.r. (50 or 100 MHz), and eventually m.s.} were obtained for all compounds.

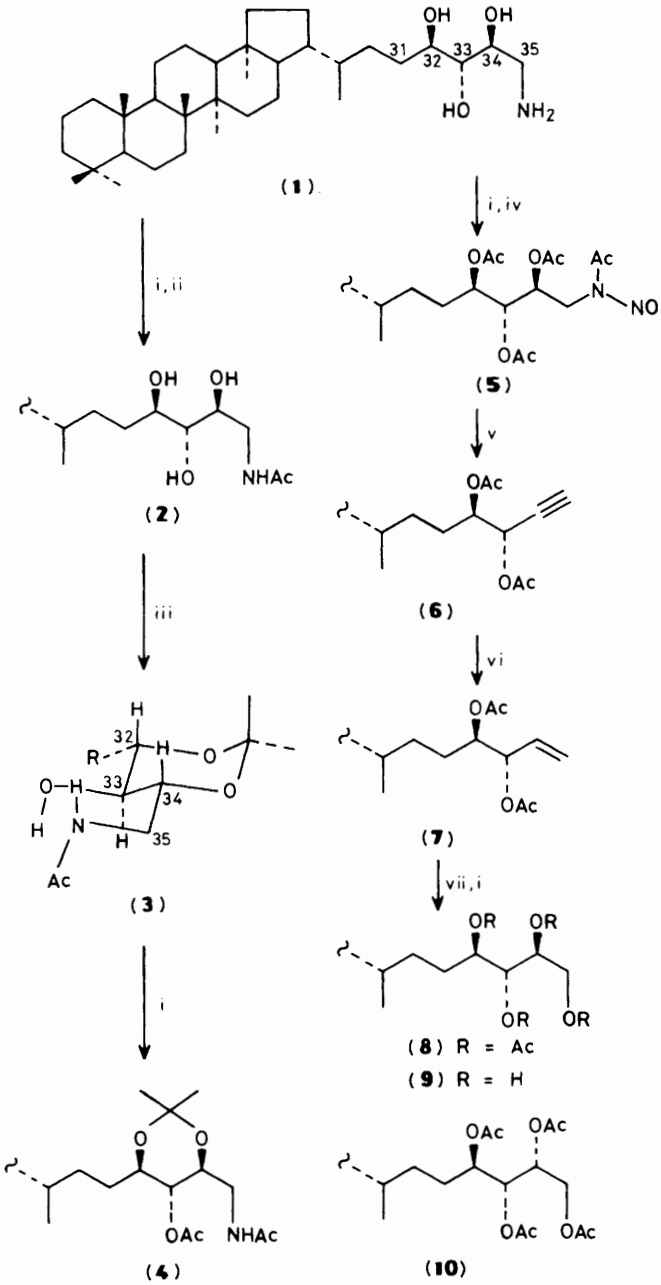
Selected spectroscopic data with i.r. and u.v. measurements in $CHCl_3$; n.m.r. spectra in $CDCl_3$. Compound (**3**): ^{1}H n.m.r., δ 1.373 and 1.457 (2s, 2 × 3H, acetonide methyl groups), 2.076 (s, 3H, CH_3CONH–), 2.91 (t, 1H, $J_{32,33} = J_{33,34}$ 9.5 Hz, 33-H), 2.99 (ddd, 1H, $J_{35a,35b}$ 15, $J_{35a,NH}$ 4.5, $J_{34,35a}$ 2.5 Hz, 35-H_a), 3.64 (m, 2H, 32-H and 34-H), 4.02 (ddd, 1H, $J_{35a,35b}$ 15, $J_{35b,NH}$ 8, $J_{34,35b}$ 2.5 Hz, 35-H_b), 5.94 (dd, 1H, $J_{NH,35a}$ 4.5, $J_{NH,35b}$ 8 Hz, –NH–).

Compound (**4**): ^{1}H n.m.r., δ 1.411 and 1.473 (2s, 2 × 3H, acetonide methyl groups), 2.000 (s, 3H, CH_3CO–), 2.098 (s, 3H, CH_3CO–), 3.15 (dt, 1H, $J_{35a,35b}$ 13, $J_{35a,NH} = J_{34,35a}$ 6 Hz, 35-H_a), 3.57 (ddd, 1H, $J_{35a,35b}$ 13, $J_{35b,NH}$ 6, $J_{34,35b}$ 3 Hz, 35-H_b), 3.69 (ddd, $J_{32,33}$ 10, $J_{31a,32}$ 6, $J_{31b,32}$ 3 Hz, 32-H), 3.81 (ddd, 1H, $J_{33,34}$ 10, $J_{34,35a}$ 6, $J_{34,35b}$ 3 Hz, 34-H), 4.50 (t, 1H, $J_{32,33} = J_{33,34}$ 10 Hz, 33-H), 5.79 (br. t, 1H, $J_{35a,NH} = J_{35b,NH}$ 6 Hz, –N*H*–).

Compound (**5**): i.r., ν_{CO} 1740 cm^{-1} and ν_{NO} 1475 cm^{-1}; u.v., λ 249 (ε 820), 409 (10), and 428 nm (10 m^2 mol^{-1}); ^{1}H n.m.r., δ 1.930 (s, 3H, CH_3CO–), 2.147 (s, 3H, CH_3CO–), 2.148 (s, 3H, CH_3CO–), 2.745 (s, 3H, $CH_3CON(NO)$–), 3.83 (dd, 1H, $J_{35a,35b}$ 14, $J_{34,35a}$ 2.5 Hz, 35-H_a), 4.35 (dd, 1H, $J_{35a,35b}$ 14, $J_{34,35b}$ 9 Hz, 35-H_b), 5.10 (m, 3H, 32-H, 33-H, and 34-H).

Compound (**6**): i.r., $\nu_{\equiv C-H}$ 3300 m, $\nu_{C\equiv C}$ 2350w, and ν_{CO} 1740s cm^{-1}; ^{1}H n.m.r., δ 2.093 (s, 3H, CH_3CO_2–), 2.101 (s, 3H, CH_3CO_2–), 2.51 (d, 1H, $J_{33,35}$ 2.5 Hz, 35-H), 5.06 (dt, 1H, $J_{31a,32} = J_{31b,32}$ 9, $J_{32,33}$ 4 Hz, 32-H), 5.55 (dd, 1H, $J_{32,33}$ 4, $J_{33,35}$ 2.5 Hz, 33-H); ^{13}C n.m.r., δ 56.2 and 64.8 (C-34 and C-35), 73.0 and 75.0 (C-32 and C-33).

Compound (**7**): ^{1}H n.m.r., δ 2.062 (s, 3H, CH_3CO_2–), 2.076 (s, 3H, CH_3CO_2–), 5.02 (dt, 1H, $J_{32,33}$ 9, $J_{31a,32} = J_{31b,32}$ 3.5 Hz, 32-H), 5.32 (m, 3H, 33-H, 35-H_a, and 35-H_b), 5.81 (ddd, 1H, $J_{34,35a}$ 17, $J_{34,35b}$ 10, $J_{33,34}$ 6.5 Hz, 34-H); ^{13}C n.m.r., δ 73.8 and 75.4 (C-32 and C-33), 119.4 (C-35), 132.0 (C-34).



Scheme 1. *Reagents*: i, Ac_2O, pyridine; ii, Amberlyst A-26, OH^-; iii, Me_2CO, $FeCl_3$; iv, 0 °C, $NaNO_2$, H_2SO_4; v, 60 °C, 1,4-dioxane; vi, H_2, Pd–$CaCO_3$, deactivated with lead; vii, OsO_4, *N*-methylmorpholine oxide. All reactions were performed at room temperature unless otherwise indicated.

We have shown that the configurations of the side-chains of aminotriol (**1**) and tetrol (**9**) are identical. Incorporation experiments carried out with ^{13}C labelled acetate indicated that the C_5 polyhydroxylated unit in the bacteriohopane derivatives (**1**) and (**9**) arises from a pentose linked *via* its C-5 carbon atom to the hopane skeleton.[9] According to the stereochemistry the precursor should be a D-ribose derivative. However, most bacteriohopanepolyol conjugates having a polar moiety linked at C-35[2] are difficult to obtain from fermentations in large amounts. Aminotriol (**1**) is invaluable in the synthesis of natural bacteriohopanepolyol conjugates and hemisynthetic structural analogs in amounts sufficient for biological tests.

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References

1 M. Rohmer, P. Bouvier-Navé, and G. Ourisson, *J. Gen. Microbiol.*, 1984, **130**, 1137; G. Ourisson, M. Rohmer, and K. Poralla, *Annu. Rev. Microbiol.*, 1987, **41**, 301, and references cited therein.
2 S. Neunlist, O. Holst, and M. Rohmer, *Eur. J. Biochem.*, 1985, **147**, 561; S. Neunlist and M. Rohmer, *J. Gen. Microbiol.*, 1985, **131**, 1363; S. Neunlist and M. Rohmer, *Biochem. J.*, 1985, **231**, 635.
3 S. Neunlist, P. Bisseret, and M. Rohmer, *Eur. J. Biochem.*, 1988, **171**, 245.
4 E. H. White, *J. Am. Chem. Soc.*, 1955, **77**, 6008.
5 M. S. Newman and S. J. Gromelsky, *J. Org. Chem.*, 1972, **37**, 3220; T. Mandai, T. Yanagi, K. Araki, Y. Morisaki, M. Kawada, and J. Otera, *J. Am. Chem. Soc.*, 1984, **106**, 3670; J. Otera, H. Misawa, and K. Sugimoto, *J. Org. Chem.*, 1986, **51**, 3830.
6 H. Lindlar, *Helv. Chim. Acta*, 1952, **35**, 446.
7 J. K. Cha, W. J. Christ, and Y. Kishi, *Tetrahedron Lett.*, 1983, **24**, 3943.
8 L. A. Reed, P. A. Risbood, and L. Goodman, *J. Chem. Soc., Chem. Commun.*, 1981, 760.
9 G. Flesch and M. Rohmer, *Eur. J. Biochem.*, 1988, in the press.