Total Synthesis of Spicamycin

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The first total synthesis of one of the spicamycin congeners, SPM VIII (**3**), is described. A preliminary model study for construction of the characteristic *N*-glycoside linkage in spicamycin using tetra-*O*-benzyl-*β*-D-mannopyranosylamine (**13**) and halopurines **5** revealed that Pd-catalyzed conditions successfully provided the coupling products **14** and **15** in good yields. It was also shown that thermal anomerization of the *N*-glycosides easily occurred, which resulted in the predominant formation of the *β*-anomer as the thermodynamically favored compound, and the activation energy of anomerization of **15** was estimated to be ca. 30 kcal/mol. The novel aminoheptose unit of spicamycin **6** was prepared stereoselectively by carbon elongation of an acyclic aldehyde, prepared by ring cleavage reaction of a highly functionalized cyclohexane derived from naturally abundant *myo*-inositol. The Pd-catalyzed coupling reaction of the *β*-heptopyranosylamine **6** with protected 6-chloropurine **5d**, followed by deprotection, provided spicamycin amino nucleoside **2**, whose condensation with dodecanoylglycine completed the total synthesis of **3**. This study confirmed the proposed unique structure of a novel nucleoside antibiotic.

Introduction

In 1983, Hayakawa and co-workers reported the isolation of spicamycin **1** from the culture broth of *Streptomyces alanosinicus* as a potent differentiation inducer of HL-60 human promyelocytic leukemia cells.[1] The structural study by spectral and degradation methods established that spicamycin consists of a novel aminoheptose, adenine, glycine, and fatty acids as a mixture of several congeners differing in their fatty acid moieties.[1] Mild acid hydrolysis of spicamycin afforded spicamycin amino nucleoside (SAN,[2] **2**), whose structure was determined by X-ray analysis and the absolute configuration assigned by the copper complex (TACu) method to be 6-(4-amino-4-deoxy-L-*glycero*-*β*-L-*manno*-heptopyranosylamino)-9*H*-purine.[3] The structure of **2** is quite unique among nucleoside antibiotics with respect to the glycosylation site; while conventional adenine−nucleoside antibiotics bear a sugar at the N-9 position, the present compound is glycosylated at the C-6 amino group of adenine with a *β*-manno anomeric configuration. To our knowledge, the only natural product containing such a glycoside structure is septacidin,[4] a 2′-epimer of spicamycin.

Extensive structure−activity relationship studies of spicamycin analogues by semisynthetic methods utilizing the condensation of **2** derived from natural spicamycin with various amino acids and fatty acids were carried out by the research group at the Kirin Brewery Co., Ltd.,[2,5] to prove that **2** is a useful precursor for the synthesis of spicamycin and its analogues. These investigations generated the promising compounds SPM VIII[2] (**3**) and KRN 5500[6] (**4**), which showed potent antitumor activities against human COL-1 colon carcinoma xenografts and gastric cancer SC-9, respectively. Although the significant biological activity and its intriguing structure have attracted considerable synthetic attention, neither a total synthesis of **1** nor the preparation of the aminoheptose moiety has been completed. Only a few syntheses of compounds related to spicamycin have been reported. Fleet reported the synthesis and biological activities of spicamycin analogues in which the aminoheptose unit was replaced by 4-amino-4-deoxy-L- and D-rhamnose (rhamnospicamycin),[7] and Acton disclosed the preparation and biological properties of septacidin analogues with 4-amino-4-deoxy- and 4-amino-4,6-dideoxy-L-glucose.[8] In this article, we report the effective construction of the *N*-glycoside linkage in spicamycin and the first total synthesis of SPM VIII (**3**), one member of

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(5) (a) Kamishohara, M.; Kawai, H.; Odagawa, A.; Isoe, T.; Mochizuki, J.; Uchida, T.; Hayakawa, Y.; Seto, H.; Tsuruo, T.; Otake, N. *J. Antibiot.* **1994**, *47*, 1305. (b) Sakai, T.; Kawai, H.; Kamishohara, M.; Odagawa, A.; Suzuki, A.; Uchida, T.; Kawasaki, T.; Tsuruo, T.; Otake, N. *J. Antibiot.* **1995**, *48*, 504. (c) Sakai, T.; Kawai, H.; Kamishohara, M.; Odagawa, A.; Suzuki, A.; Uchida, T.; Kawasaki, T.; Tsuruo, T.; Otake, N. *J. Antibiot.* **1995**, *48*, 1467.

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(7) (a) Martin, A.; Butters, T. D.; Fleet, G. W. J. *Chem. Commun.* **1998**, 2119. (b) Martin, A.; Butters, T. D.; Fleet, G. W. J. *Tetrahedron Asymmetry* **1999**, *10*, 2343.

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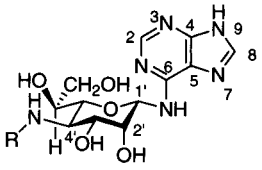


R = $Me_2CH(CH_2)_nCONHCH_2CO$- or
$MeCH_2CH_2(CH_2)_nCONHCH_2CO$- (n = 8~14):
Spicamycin (**1**)
R = H: Spicamycin Amino Nucleoside (SAN) (**2**)
R = $Me(CH_2)_{10}CONHCH_2CO$-: SPM VIII (**3**)
R = $Me(CH_2)_8CH{=}CHCH{=}CHCONHCH_2CO$-: KRN 5500 (**4**)

Figure 1.

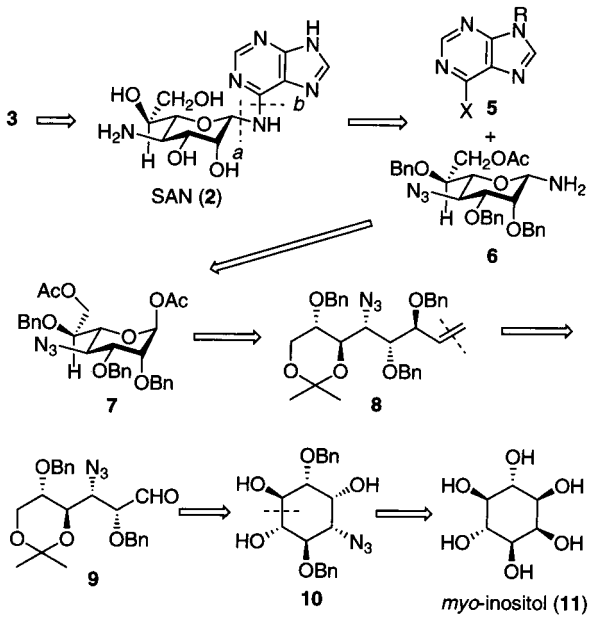


Figure 2. Retrosynthetic analysis of SPM VIII (**3**).

the spicamycin family, starting from the naturally abundant cyclitol *myo*-inositol.[9]

Synthetic Plan

The previous structure−activity relationship studies[2,5] established that SAN (**2**) should be a useful intermediate for spicamycin synthesis. A major obstacle to the synthesis of **2** would be the construction and anomeric control of the novel *N*-glycoside with a β-manno configuration. Two straightforward approaches to the *N*-glycoside are possible (Figure 2), the glycosylation of adenine derivatives (C−N bond formation at *a*) and a coupling of β-glycosylamine with purines having appropriate functional groups (C−N bond formation at *b*). The suspected difficulties of direct glycosylation of adenine[7b] as well as in controlling the β-manno anomeric configuration[10] led us to investigate the latter approach. This idea invites disconnection into β-heptopyranosylamine **6** and halopurine derivative **5**. The β-heptopyranosylamine **6** was envisioned to be synthesized from the corresponding heptopyranose **7**. This cyclic sugar **7** could be generated from aldehyde **9** via acyclic precursor **8**. The aldehyde **9**, in turn, would derive from the ring cleavage reaction of a highly functionalized cyclohexane derivative **10**, which was envisioned as arising from *myo*-inositol (**11**). Although the use of **11**, which is a *meso* compound and generates racemic compounds by conventional transformations, as the starting material requires the optical resolution during the synthetic process, it is advantageous that both enantiomers are available for the determination of the synthetically unconfirmed absolute stereochemistry of spicamycin. In addition, the cyclic structure as well as the established protection−deprotection methods of polyhydroxy groups in *myo*-inositol[11] would be useful for the introduction of desired functional groups stereoselectively on its cyclohexane ring.

Construction of the *N*-Glycoside Model and Its Thermodynamic and Kinetic Properties

The precedent for preparation of spicamycin analogues (rhamnospicamycin) by Fleet[7] revealed that the glycosylamino-9*H*-purine with a β-manno configuration could be synthesized by a multistep procedure. Thus, the coupling reaction of β-rhamnopyranosylamine, prepared from a rhamnopyranose derivative, with 4,6-dichloro-5-nitropyrimidine followed by ammonolysis afforded 6-(β-rhamnopyranosylamino)-4-amino-5-nitropyrimidine, whose reaction with triethyl orthoformate and subsequent hydrogenation provided the desired *N*-glycoside in 4.7% overall yield from the rhamnopyranose. While these classical methods[8,12] were reliable for the construction of the *N*-glycoside, the low efficiency (requiring at least 3 steps and giving products in less than 40% overall yield from pyranosylamines) prompted us to investigate the single-step construction of the glycosidic linkage.[13] For this purpose, the known mannopyranosylamine[14] **13** was chosen as the model substrate[15] and was subjected to the coupling reactions with 6-halopurine derivatives **5a**−**d** (Scheme 1).

Reaction of mannopyranosylamine **13** with 6-chloropurine **5a** under the base-catalyzed conditions (Et_3N,

(9) Suzuki, T.; Tanaka, S.; Yamada, I.; Koashi, Y.; Yamada, K.; Chida, N. *Org. Lett.* **2000**, *2*, 1137.

(10) Formation of a mixture of 6-(α- and β-D-ribofuranosylamino)-9*H*-purines and their pyranosyl isomers by solid-state reaction of D-ribose and adenine (20 mmHg, 100 °C, 6 h, 74% combined yield) has been reported; see: Fuller, W. D.; Sanchez, R. E.; Orgel L. E. *J. Mol. Biol.* **1972**, *67*, 25. A similar solid-state reaction (1 atm, 110 °C, 4 h) afforded 6-(β-D-ribofuranosylamino)-9*H*-purine in 42% yield; however, this method could not be applied to D-xylose and D-glucose. See: Fujishima, T.; Uchida, K.; Yoshino, H. Japan Kokai Tokkyo Koho 50-34040, 1975; *Chem. Abstr.* **1976**, *84*, 106007g.

(11) For recent reviews on utilization of *myo*-inositol in natural product synthesis, see: (a) Billington, D. C. *Chem. Soc. Rev.* **1989**, *18*, 83. (b) Reitz, A. B. *Inositol phosphates and Derivatives, Synthesis, Biochemistry, and Therapeutic Potential*; American Chemical Society: Washington, DC, 1991. (c) Potter, B. V. L. *Nat. Prod. Rep.* **1990**, *7*, 1. (d) Billington, D. C. *The Inositol Phosphates, Chemical Synthesis and Biological Significance*; VCH: Weinheim, Germany, 1993. (e) Chida, N.; Ogawa, S. *Chem. Commun.* **1997**, 807.

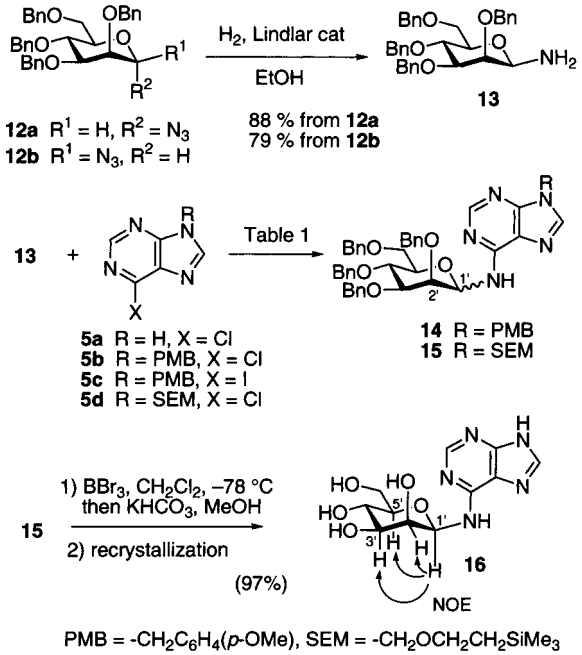
(12) (a) Ikehara, M.; Tada, H. *Chem. Pharm. Bull.* **1963**, *11*, 1102. (b) Fukatsu, S.; Sawa, A.; Umezawa, S. *Bull. Chem. Soc. Jpn.* **1974**, *47*, 917.

(13) Chida, N.; Suzuki, T.; Tanaka, S.; Yamada, I. *Tetrahedron Lett.* **1999**, *40*, 2573.

(14) Rao, C. S.; Ratcliffe, A. J.; Fraser-Reid, B. *J. Chem. Soc., Perkin Trans. 1* **1993**, 1207.

(15) Compound **13** was prepared by hydrogenation of 2,3,4,6-tetra-*O*-benzyl-β-D-mannopyranosyl azide (**12b**) in the presence of Lindlar catalyst in EtOH at room temperature, according to the method reported by Fraser-Reid[14] with slight modification. Interestingly, when α-azide isomer **12a** was subjected to the similar reaction conditions, rapid anomerization of pyranosylamine took place to afford **13** as the sole product, and α-anomer of **13** could not be detected in the reaction mixture, implying the β-anomer is thermodynamically stable compound. A similar phenomenon has been reported by Ogawa in the hydrogenation of 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl azide; see: Ogawa, T.; Nakabayashi, S.; Shibata S. *Agric. Biol. Chem.* **1983**, *47*, 281.

Scheme 1



BuOH, reflux) employed for the successful coupling of aliphatic amines with **5a**[16] led only to the decomposition of **13**, presumably due to the instability of the pyranosylamine as well as the reduced nucleophilicity of the amino group by the presence of an endocyclic oxygen. At this juncture we turned our attention to the Pd-catalyzed conditions,[17] which have been recently reported by Buchwald,[18] Hartwig,[19] and Tanaka[20] to be efficient procedures for the coupling of aliphatic and aromatic amines with aryl halides or triflates. Although the reaction of **13** with **5a** under the conditions reported by Tanaka[20] (Table 1, entry 1) gave no coupling products, we were delighted to find that use of 9-protected 6-chloropurine[21] **5b** afforded the coupling product **14**, though in only 25% yield (entry 2), as an inseparable mixture of α- and β-anomers (ca. 1:5). The dependence on various reaction parameters was then examined. While use of 6-iodopurine[22] derivative **5c** (entry 4) and Hartwig's conditions[19a] (entry 6) did not offer much improvement, Buchwald's conditions[18a] using an excess amount of 9-PMB-6-chloropurine (**5b**) gave **14** in considerably enhanced yield (entry 8). With SEM-protected purine **5d**, which was expected to undergo facile deprotection, similar reaction conditions provided **15** in 75% yield (entry 9). The use of (+)-BINAP gave slightly lower yield (entry 10). It should be noted that when the coupling reaction was terminated at 0.5 h, the anomeric ratio was significantly altered (entry 11).

The structures of the coupling products were established by ^{1}H NMR analysis. The major isomer of **15** showed its anomeric proton at δ 5.76 as a broad singlet, whereas that of the minor isomer appeared at δ 6.40 (broad s). When the broad signal at δ 7.01 (NH of the β-anomer, exchangeable with D_2O) was irradiated, the signal of the anomeric proton sharpened, implying the connectivity of the C-1′ carbon in mannose to the amino group at C-6 of the adenine moiety. The observed NOEs in the major isomer between H-1′ and H-3′, H-1′ and H-5′, and H-1′ and H-2′ clearly showed it to be in the β-manno configuration. Treatment of **15** (α/β mixture) with BBr_3 in CH_2Cl_2 at −78 °C resulted in global deprotection[23] to provide, after recrystallization from water, β-D-mannopyranosylamino-9*H*-purine **16** in 97% yield. The coupling constants ($J_{1',2'} < 1$ Hz, $J_{2',3'} = 3.2$ Hz, $J_{3',4'} = 9.4$ Hz, $J_{4',5'} = 9.4$ Hz) and NOEs observed in the ^{1}H NMR again supported the β-manno configuration.

Whereas an anomerically pure β-mannopyranosylamine **13** was employed as the starting material, the coupling products **14** and **15** were obtained as anomeric mixtures, and the ratio of anomers varied by reaction time. Furthermore, the deprotected product **16** was isolated as a single β-anomer in high yield despite the starting material **15** being a 1:5 anomeric mixture. These phenomenon suggested that the anomerization of **14**−**16** had occurred during the reaction and/or purification processes. With interest to the anomeric behavior of the *N*-glycosides, a kinetic analysis of anomerization of **15** was carried out (Scheme 2). Compound **15**, consisting of α-anomer and β-anomer in a ratio of 30.3:69.7, was dissolved in toluene (5 mg/mL), and the solution was heated at 100 °C in a sealed tube. The time course of the anomeric ratios was followed by HPLC (Figure 3a). From these experiments, it was found that the *N*-glycoside **15** showed thermal anomerization and reached equilibrium at α:β = 10.3:89.7 after 300 h. It was also shown that the α-anomer, which would arise from anomerization of pyranosylamine **13** during the coupling process,[24] is the kinetic product and the β-anomer is the thermodynamic one. With application of eq 1,[25] simple first-order kinetics were observed (Figure 3b) in the anomerization of **15**. Similar reactions were carried out at 105 and 110 °C, and the resulting rate constants obtained from eqs 1 and 2 are summarized in Table 2. Arrhenius plots shown in Figure 3c demonstrated well-fitted linearity, and the activation energies of anomerization of **15** in toluene were estimated to be 28.2 kcal/mol (from α-anomer to β-anomer) and 32.0 kcal/mol (β-anomer to α-anomer), respectively. The deprotected compound **16** was found to show much more rapid anomerization than **15**, even at room

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(20) Reddy, N. P.; Tanaka, M. *Tetrahedron Lett.* **1997**, *38*, 4807.

(21) Compounds **5b,d** were prepared from commercially available 6-chloropurine by essentially the same procedure as that reported for the preparation of 9-benzyl-6-chloropurine; see: Gundersen, L.-L.; Bakkestuen, A. K.; Aasen, A. J.; Overas, H.; Rise, F. *Tetrahedron* **1994**, *50*, 9743.

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(23) Attempted deprotection of compounds **14** and **15** under hydrogenolytic conditions (H_2, Pd catalyst) was unsuccessful; hydrogenation of the heterocyclic moiety was observed. Treatment of **14** with BBr_3/CH_2Cl_2 resulted in the formation of many unidentified products, and no desired product was obtained.

(24) Anomerization of pyranosylamine **13** during the acylation with 2-chlorobenzoyl chloride in pyridine, which resulted in the formation of α-amide and β-amide in a ratio of 1:4.7, has been observed by Fraser-Raid; see ref 14.

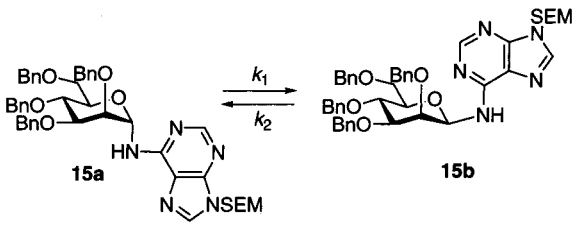
(25) Hough, L.; Richardson, A. C. In *Rodd's Chemistry of Carbon Compounds*, 2nd ed.; Coffey, S., Ed.; Elsevier: Amsterdam, 1967; Vol. I, Part F, Chapter 23, p 173.

Table 1. Coupling Reaction of Mannopyranosylamine (13) with Halopurines[a]

entry	purine	catalyst[b]	additive[c]	base	solvent	time (h)	molar ratio **13**:purine	product	yield[f] (%)
1	**5a**	$Pd(P(cy)_3)_2Cl_2$		NaO*t*Bu	toluene		1.5:1.0		0
2	**5b**	$Pd(P(cy)_3)_2Cl_2$		NaO*t*Bu	toluene	12	1.5:1.0	**14**[d]	25
3	**5b**	$Pd(P(cy)_3)_2Cl_2$		NaO*t*Bu	DMF		1.5:1.0		0
4	**5c**	$Pd(P(cy)_3)_2Cl_2$		NaO*t*Bu	toluene	21	1.5:1.0	**14**[d]	35
5	**5b**	$Pd(P(cy)_3)_2Cl_2$		Cs2CO3	toluene	16	1.5:1.0	**14**[d]	41
6	**5b**	$Pd_2(dba)_3$	DPPF	NaO*t*Bu	toluene	20	1.5:1.0	**14**[d]	28
7	**5b**	$Pd_2(dba)_3$	(−)-BINAP	NaO*t*Bu	toluene	14	1.5:1.0	**14**[d]	45
8	**5b**	$Pd_2(dba)_3$	(−)-BINAP	NaO*t*Bu	toluene	14	1.0:2.0	**14**[d]	79
9	**5d**	$Pd_2(dba)_3$	(−)-BINAP	NaO*t*Bu	toluene	9	1.0:2.0	**15**[d]	75
10	**5d**	$Pd_2(dba)_3$	(+)-BINAP	NaO*t*Bu	toluene	9	1.0:2.0	**15**[d]	68
11	**5d**	$Pd_2(dba)_3$	(−)-BINAP	NaO*t*Bu	toluene	0.5	1.0:2.0	**15**[e]	75

[a] All reactions were carried out with purine [40 μmol (entry 1−7) or 120 μmol (entry 8−11)] and compound **13** (60 μmol) in the presence of catalyst (12 μmol) and base [60 μmol (entry 1−7) or 90 μmol (entry 8−11)] in solvent (2.5 mL) at 130 °C in a sealed tube. [b] cy = cyclohexyl; dba = dibenzylideneacetone. [c] 200 mol % to catalyst. DPPF = 1,1′-bis(diphenylphosphino)ferrocene; BINAP = 2,2′-bis(diphenylphosphino)-1,1′-binaphthyl. [d] Obtained as an anomeric mixture (α:β = ca. 1:5). Ratio was determined with 300 MHz ^{1}H NMR spectra. [e] α:β = 1:2.2. [f] Isolated yields after silica gel chromatography.

Scheme 2



$$\frac{d[\mathbf{15b}]}{dt} = k_1[\mathbf{15a}] - k_2[\mathbf{15b}]$$

$$t = \frac{1}{k_1 + k_2} \ln \frac{[\mathbf{15b}]_0 - [\mathbf{15b}]_\infty}{[\mathbf{15b}]_t - [\mathbf{15b}]_\infty} \quad (1) \qquad K = \frac{k_1}{k_2} = \frac{[\mathbf{15b}]_\infty}{[\mathbf{15a}]_\infty} \quad (2)$$

where $[\mathbf{15b}]_0$ is initial concentration of **15b** at $t = 0$;
$[\mathbf{15b}]_\infty$ is final concentration of **15b** at equilibrium; and
$[\mathbf{15b}]_t$ is concentration of **15b** at time t.

Table 2. Rate Constants k_1 and k_2 and Equilibrium Constant K for Anomerization of 15

T (°C)	K	$k_1 + k_2$ (10^{-3} h^{-1})	k_1 (10^{-3} h^{-1})	k_2 (10^{-3} h^{-1})
100	8.71	18.7	16.8	1.93
105	8.01	29.2	26.0	3.24
110	7.62	51.3	45.3	5.95

temperature;[26] however, the highly crystalline nature of the β-anomer **16**, which easily crystallizes out from most of the solvents, unfortunately did not allow us to carry out the quantitative kinetic analysis.

Having developed the Pd-catalyzed coupling reaction of mannopyranosylamine **13** with SEM-protected 6-chloropurine **5d** followed by deprotection to give **16**, we believed it to be now possible to construct the *N*-glycoside linkage in spicamycin. It is also an important finding that the thermal anomerization of the *N*-glycosides **15** and **16** takes place and the β-anomer was thermodynamically more stable than α-anomer, providing the desired anomer with β-manno configuration as the major product.[27]

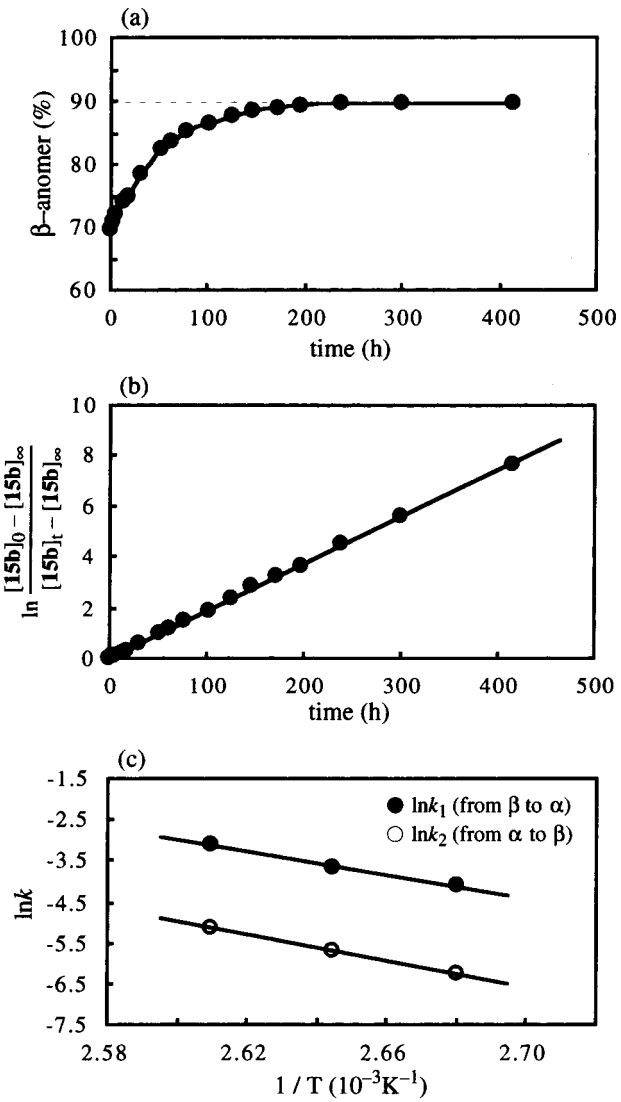


Figure 3. Kinetic data for the anomerization of **15**. (a) The time course of anomeric ratios at 100 °C. Ratios were determined with HPLC (Finepak SIL, JASCO Corp., 4.6 mm i.d., 250 mmL, *i*PrOH/hexane = 1/2; retention volume for **15a** 5.3 mL and for **15b** 7.4 mL). (b) Kinetic constant at 100 °C. (c) Arrhenius plots.

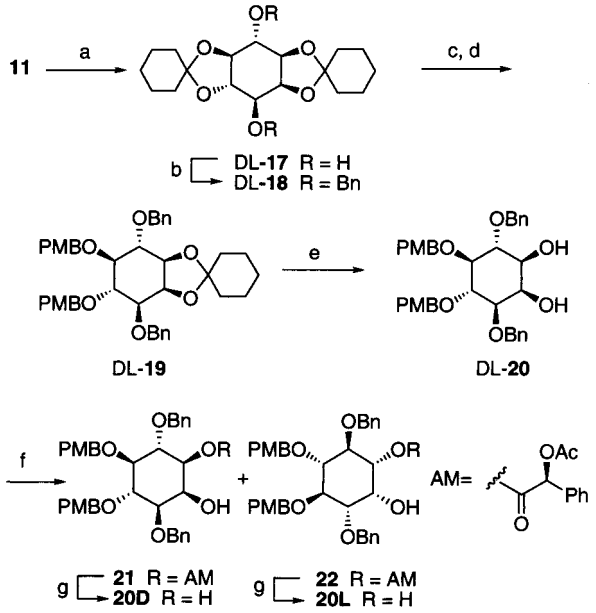
Preparation of the Aminoheptose Moiety

The successful synthesis of model compound **16** now renders our synthetic plan feasible and suggests β-hep-

(26) Facile anomerization of α-1-sulfonamidyl-2-deoxy-2-iodo-mannopyranosides, which resulted in the predominant formation of β-anomers as the thermodynamic products, has been recently reported; see: Owens, J. M.; Yeung, B. K. S.; Hill, D. C.; Petillo, P. A. *J. Org. Chem.* **2001**, *66*, 1484.

(27) Recent reports of anomerization of some *N*-glycosides: (a) Vaino, A. R.; Szarek, W. A. *J. Org. Chem.* **2001**, *66*, 1097. (b) Randell, K. D.; Johnston, B. D.; Green, D. F.; Pinto, B. M. *J. Org. Chem.* **2000**, *65*, 220. (c) Randell, K. D.; Frandsen, T. P.; Stoffer, B.; Johnson, M. A.; Svensson, B.; Pinto, B. M. *Carbohydr. Res.* **1999**, *321*, 143. (d) Andrews, J. S.; Weimar, T.; Frandsen, T. P.; Svensson, B.; Pinto, B. M. *J. Am. Chem. Soc.* **1995**, *117*, 10799.

Scheme 3[a]

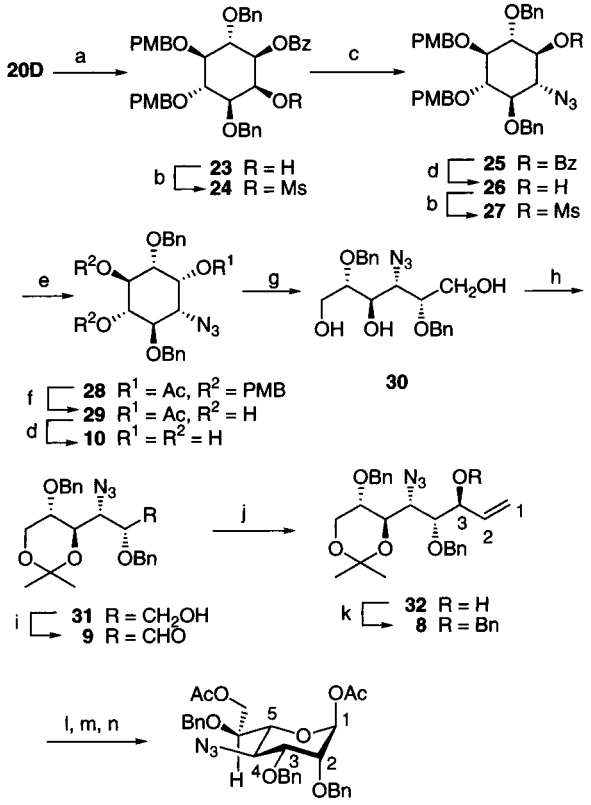


[a] Reagents: (a) 1,1-dimethoxycyclohexane, *p*-TsOH·H_2O, DMF, 100 °C; (b) NaH, BnCl, DMF, rt, 3 h; (c) *p*-TsOH·H_2O, EtOH, 45 °C, 2 h; (d) NaH, PMBCl, Bu_4NI, THF/DMF = 2/1, rt, 12 h; (e) AcOH/H_2O = 4/1, 50 °C, 1 h; (f) L-(+)-*O*-acetylmandelic acid, DCC, DMAP, CH_2Cl_2, −78 to −15 °C; (g) NaOMe, MeOH, rt, 30 min.

topyranosylamine **6** to be a promising intermediate for the total synthesis of spicamycin. As our starting point, the known racemic diol DL-**17**,[28] prepared from **11** in a one-step reaction, was converted into bis(benzyl ether) DL-**18** in 84% yield (Scheme 3). Selective removal of the *trans*-ketal group in DL-**18**, followed by *p*-methoxybenzylation of the diol afforded DL-**19** in 74% yield. Deprotection of the *cis*-ketal function in DL-**19** gave diol DL-**20** in 73% yield. The equatorial hydroxy group in DL-**20** was selectively acylated by a treatment with a slight excess amount of l-*O*-acetyl mandelic acid in the presence of DCC and DMAP[29] to provide a pair of chiral diastereoisomers, **21** and **22**, which were easily separated by silica gel chromatography, in 41% and 37% isolated yields, respectively. Deacylation of **21** gave **20D** (81% yield), and that of **22** afforded the enantiomer **20L** in 89% yield.[30]

Synthesis of the aminoheptose moiety commenced with **20D**, whose equatorial hydroxy group was selectively benzoylated to give **23** in 84% yield (Scheme 4). The remaining hydroxy group in **23** was converted to the azide via mesylate **24** to afford **25** in 86% yield from **23**. Deprotection of the benzoyl group in **25** afforded **26** in 96% yield, whose hydroxy group was mesylated to give **27** in 84% yield. Treatment of **27** with KOAc in the presence of 18-crown-6 ether cleanly provided the inverted acetate **28** in 79% yield. Removal of *O*-PMB and *O*-acetyl protecting groups in **28** gave triol **10** in 95% yield from **28**.

Scheme 4[a]



[a] Reagents: (a) BzCl, pyridine, rt; (b) MsCl, pyridine, rt; (c) NaN_3, DMF, 100 °C; (d) NaOMe, MeOH, rt; (e) KOAc, 18-crown-6 ether, DMF, 100 °C; (f) DDQ, CH_2Cl_2/H_2O = 20/1, 0 °C; (g) $NaIO_4$, MeOH/H_2O = 5/1, 0 °C, and then $NaBH_4$, MeOH/H_2O = 5/1, 0 °C; (h) 2,2-dimethoxypropane, *p*-TsOH·H_2O, DMF, 0 °C; (i) DMSO, DCC, TFA, pyridine, benzene, rt; (j) vinyllithium, Et_2O, −120 °C; (k) NaH, BnBr, DMF, 0 °C; (l) O_3, CH_2Cl_2, −78 °C, and then Me_2S; (m) *p*-TsOH·H_2O, MeOH, 0 °C; (n) Ac_2O, pyridine, 0 °C.

Periodate oxidation of **10**, followed by reduction with $NaBH_4$, afforded acyclic triol **30** in 95% yield. Subsequent treatment of **30** with 2,2-dimethoxypropane provided azidoalditol **31** in 83% yield. Moffatt oxidation of **31** gave unstable aldehyde **9**, which, without isolation, was subjected to the carbon elongation reaction. Although reaction of **9** with vinylmagnesium bromide in CH_2Cl_2 or Et_2O gave the Felkin−Anh product **32** in only ca. 25% yield from **31**, the use of vinyllithium[31] in Et_2O at −120 °C brought considerable improvement and afforded **32** as the major isomer (86% from **31**).[32,33] Protection of the hydroxy group in **32** provided tri-*O*-benzyl ether **8** in 79% yield. Ozonolysis of **8** (Me_2S workup), followed by acidic hydrolysis and subsequent treatment with acetic anhydride and pyridine, furnished 4-azidoheptopyranosyl acetate **7** as the single α-anomer in 81% yield. The coupling constants in **7** ($J_{1,2}$ = 2.0 Hz, $J_{2,3}$ = 2.9 Hz, $J_{3,4}$ = 10.0 Hz, $J_{4,5}$ = 10.0 Hz) clearly revealed that compound **7** has a *manno* configuration.

(28) (a) Garegg, P. J.; Iversen, T.; Johansson, R.; Lindberg, B. *Carbohydr. Res.* **1984**, *130*, 322. (b) Jiang, C.; Baker, D. C. *J. Carbohydr. Chem.* **1986**, *5*, 615.

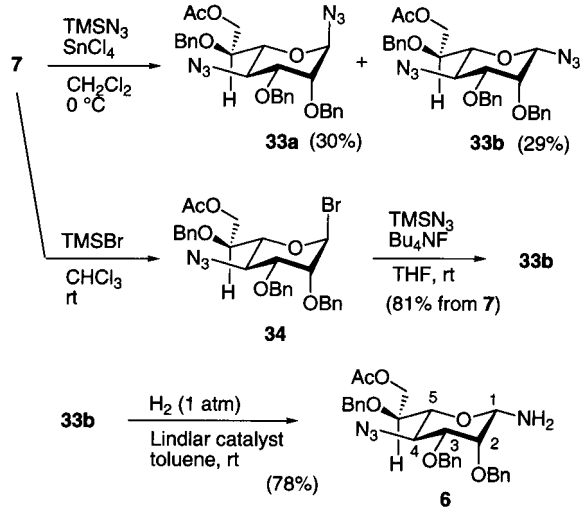
(29) Chida, N.; Yamada, E.; Ogawa, S. *J. Carbohydr. Chem.* **1988**, *7*, 555.

(30) The optical purities of **20D** and **20L** were determined by HPLC analysis [Chiralcel OD, *i*-PrOH−hexane (1:2)] to be both >99% ee, and their absolute structures were confirmed by their transformation into the known compounds, 1D- and 1L-1,4,5,6-tetra-*O*-benzyl-*myo*-inositol (Shvets, V. I.; Klyashchitskii, B. A.; Stepanov, A. E.; Evstigneeva, R. P. *Tetrahedron* **1973**, *29*, 331), respectively. For details, see the Supporting Information.

(31) Vinyllithium was prepared by the procedure of Seyferth and Weiner; see: Seyferth, D.; Weiner, M. A. *J. Am. Chem. Soc.* **1961**, *83*, 3583.

(32) The small amount of C-3 epimer of **32** was detected in the ^1H NMR spectrum (**32**/C-3 epimer: >8/1), but this could not be isolated.

Scheme 5

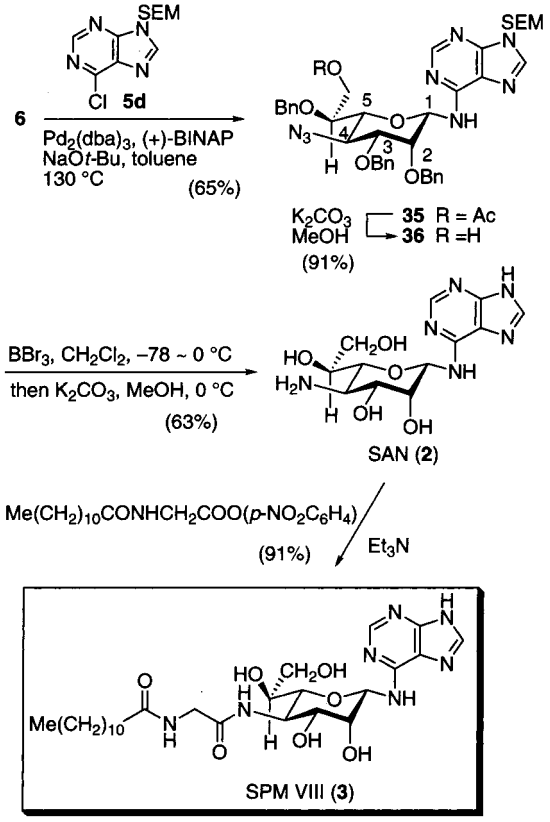


From an earlier observation in preparation of protected mannopyranosylamine **13**, it was expected that both α- and β-anomeric azides such as **33a,b** would afford only β-pyranosylamine upon hydrogenation (vide supra). Acetate **7** was treated with $TMSN_3$ in the presence of $SnCl_4$[34] to afford α- and β-azide compounds **33a,b** in 30 and 29% isolated yields, respectively (Scheme 5). When β-azide **33b** was subjected to hydrogenation conditions in the presence of Lindlar catalyst in toluene, only the anomeric azide group was reduced to give the desired β-heptopyranosylamine **6** in 78% yield as the sole product. The observed NOEs (between H-1 and H-2, H-1 and H-3, and H-1 and H-5) and coupling constants ($J_{1,2} < 1$ Hz, $J_{2,3} = 2.7$ Hz, $J_{3,4} = 9.8$ Hz) unambiguously showed its β-manno configuration. Similar treatment of α-azide **33a**, however, gave none of the desired product; only decomposition of the starting material, presumably induced by the competitive reduction of 4-azide function, was observed. These results led us to explore the selective formation of β-azide **33b** from **7**, and after several attempts, it was found that the two-step reaction sequence involving anomeric bromide **34** gave good results. Thus, treatment of **7** with trimethylsilyl bromide[35] gave unstable α-bromoheptose **34**, which was reacted with trimethylsilyl azide in the presence of TBAF[36] to afford **33b** in 81% yield from **7**.

Total Synthesis of Spicamycin

Having completed the preparation of the heptopyranosylamine unit **6** with the correct stereochemistry, we

Scheme 6



next investigated the key reaction, construction of the *N*-glycoside (Scheme 6). Reaction of amine **6** with **5d** under the conditions optimized in the model experiments (with (+)-BINAP, 130 °C, 2.5 h) worked well, and the desired coupling product, fully protected 6-(β-pyranosylamino)-9-SEM-purine **35** was obtained as the single β-anomer in 65% yield.[37] The observed NOEs between H-1 and H-2, H-1 and H-3, and H-1 and H-5 in the sugar portion clearly supported the β-manno structure of **35**. Deacylation of **35** gave **36** (91% yield), which was treated with excess BBr_3 in CH_2Cl_2 at −78 °C to 0 °C (K_2CO_3/MeOH workup) to provide spicamycin amino nucleoside **2** in 63% yield after purification by silica gel chromatography and reverse-phase chromatography followed by crystallization. It is noteworthy that azide group in **36** was unexpectedly reduced to amine under BBr_3 conditions.[38] The spectral (^1H and ^{13}C NMR) as well as physical properties of synthetic **2** were fully identical with those of SAN,[39] prepared from natural spicamycin by degradation.

Finally, according to the reported procedure,[2] SAN (**2**) was reacted with dodecanoylglycine *p*-nitorophenyl ester[2]

(33) Organometallic reagents have been reported to add to α-alkoxy and α-imino aldehydes and ketones via chelation control to give anti-Felkin−Anh products preferentially. See: (a) Still, W. C.; Schneider, J. A. *Tetrahedron Lett.* **1980**, *21*, 1035. (b) Burke, S. D.; Deaton, D. N.; Olsen, R. J.; Armistead, D. M.; Blough, B. E. *Tetrahedron Lett.* **1987**, *28*, 3905. (c) Iida, H.; Yamazaki, N.; Kibayashi, C. *J. Org. Chem.* **1986**, *51*, 3769. (d) Polt, R.; Peterson, M. A. *Tetrahedron Lett.* **1990**, *31*, 4985. The structural complexity of aldehyde **9** (the presence of an azide group and a dioxolane ring) might prevent the formation of the α-chelation transition structure and gave the Felkin−Anh product **32** as the major product in the reactions with vinyl Grignard and vinyllithium reagents. For reviews on chelation and nonchelation control addition reactions, see: (e) Huryn, D. M. In *Comprehensive Organic Synthesis*; Trost, B. M., Ed.; Pergamon Press: Oxford, U.K., 1991; Vol. 1, Chapter 1.2. (f) Reetz, M. T. *Angew. Chem., Int. Ed. Engl.* **1984**, *23*, 556.

(34) Györgydeàk, Z.; Szilàgyi, L. *Liebigs Ann. Chem.* **1987**, 235. Szilagyi, L.; Györgydeàk, Z. *Carbohydr. Res.* **1985**, *143*, 21.

(35) Gillard, J. W.; Israel, M. *Tetrahedron Lett.* **1981**, *22*, 513.

(36) Soli, E. D.; Manoso, A. S.; Patterson, M. C.; DeShong, P.; Favor, D. A.; Hirschmann, R.; Smith, A. B., III. *J. Org. Chem.* **1999**, *64*, 3171.

(37) When this reaction was terminated at 40 min, a mixture of **35** and its α-anomer (ca. 4:1) was formed in 40% yield, revealing that anomerization of **6** and **35**, as observed in the model experiments, had taken place during the coupling reaction and that β-anomer **35** is the thermodynamically favored product.

(38) Aryl azides are known to undergo reduction (acidolysis) to amines by the action of protic acids (HBr or HI) or Lewis acids ($AlEt_3$ or $AlBr_3$) followed by hydrolysis; see: Abramovitch, R. A.; Kyba, E. P. In *The Chemistry of the Azido Group*; Patai, S., Ed.; Interscience Publishers: London, 1971; Chapter 5. Azides are also known to react with trialkylboranes; see: Petler A.; Smith, K.; Brown, H. C. *Borane Reagents*; Academic Press: London, 1988; p 253. The plausible mechanism for reduction of azide group in **36** would be direct acidolysis or decomposition of $RN(Br)BBr_2$, which might be formed by attack of BBr_3, with HBr liberated by addition of MeOH in the workup process.

to give SPM VIII (**3**) in 91% yield. Again synthetic **3** was identical in all respects with an authentic sample.[39]

Conclusion

The first total synthesis of a member of the spicamycin family, SPM VIII (**3**), starting from *myo*-inositol (**11**) has been accomplished. This study fully confirmed the proposed unique structure of the biologically important natural product and established a synthetic approach to spicamycin and its analogues. The efficiency of the Pd-catalyzed coupling reaction of glycosylamines with protected 6-chloropurine is also noteworthy for the construction of the novel *N*-glycoside. The thermodynamic and kinetic properties of the *N*-glycoside found in this study, i.e., thermal anomerization, preferential formation of the β-anomer, and activation energy for anomerization of **15**, should be important information for the understanding of anomeric behavior of *N*-glycosides.

Experimental Section

Full experimental details and characterization are provided in the Supporting Information.

Acknowledgment. We thank Dr. K. Akimoto (Pharmaceutical Research Laboratory, Kirin Brewery Co., Ltd., Takasaki, Japan) for providing us with the authentic SAN prepared from natural spicamycin. We also thank Prof. Y. Hayakawa (Institute of Molecular and Cellular Biosciences, University of Tokyo, Japan) for valuable information on spicamycin. This work was supported by the Grant-in-Aid for Scientific Research on Priority Area (A) "Exploitation of Multi-Element Cyclic Molecules" from the Ministry of Education, Culture, Sports, Science, and Technology of Japan.

Supporting Information Available: Full experimental procedures, characterization of compounds and ^{1}H NMR copies of compounds **2**, **3**, **6**−**9**, **32**, **33a,b**, **35**, and **36** (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

JO010925C

(39) Synthetic **2**: mp 154−156 °C (dec) (DMF−toluene); $[\alpha]_D^{23}$ +21.5 (*c* 0.14, DMF). Authentic **2**: mp 154−156 °C (dec) (DMF−toluene); $[\alpha]_D^{25}$ +21.7 (*c* 0.36, DMF). Synthetic **3**: mp 198−202 °C (dec); $[\alpha]_D^{24}$ +13.0 (*c* 0.20, MeOH). Authentic **3**: mp 200−203 °C (dec); $[\alpha]_D^{25}$ +13.7 (*c* 0.20, MeOH). These data for authentic samples were measured in our laboratory on material of natural origin, kindly supplied by Dr. Akimoto (Kirin Brewery Co., Ltd.).