SYNTHETIC STUDY OF NEOCARZINOSTATIN CHROMOPHORE: STEREOSELECTIVE SYNTHESIS OF *N*-METHYLFUCOSAMINE AND ITS α-GLYCOSIDE

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Abstract - Stereoselective glycosylation of cyclopentanol with 2-azido-2,6-dideoxygalactopyranose (2-azidofucose) derivatives and its transformation to *N*-methylfucosamine was achieved as a model for the synthesis of neocarzinostatin chromophore.

Recently, considerable effort has been devoted to elucidating the role of the *N*-methylfucosamine moiety of neocarzinostatin chromophore (**1**)[1] in T-selective DNA cleavage[2] and specific binding to apoprotein.[3] The syntheses of **1** and its analogs possessing α-*N*-methylfucosamine are of great interest in clarifying this issue.[4,5] Although remarkable progress has recently been made with the stereoselective glycosylation method,[6] much work remains for establishing a mild and α-selective glycosylation method with *N*-methylfucosamine derivatives in the synthesis of **1**. Glycosylation of *N*-acylated 2-aminosugars generally tends to give β-glycoside *via* participation of the neighboring group. We previously examined glycosylation using a donor with a free *N*-methylamino group, since *in situ* protection as a quaternary

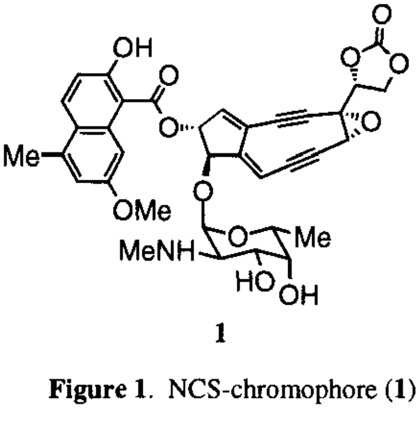

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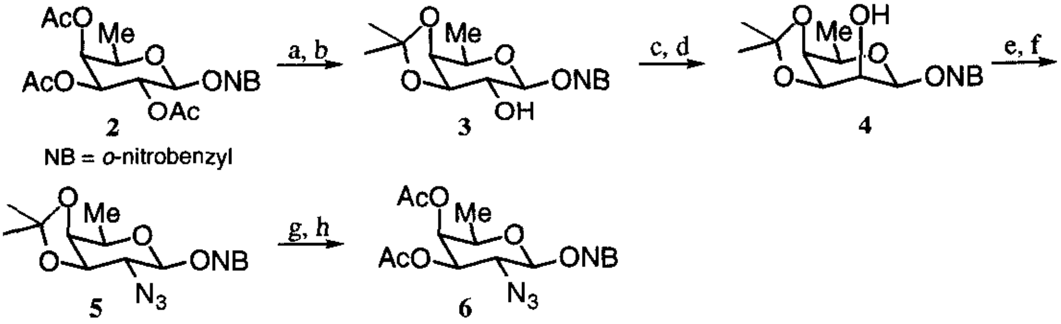
Figure 1. NCS-chromophore (**1**)

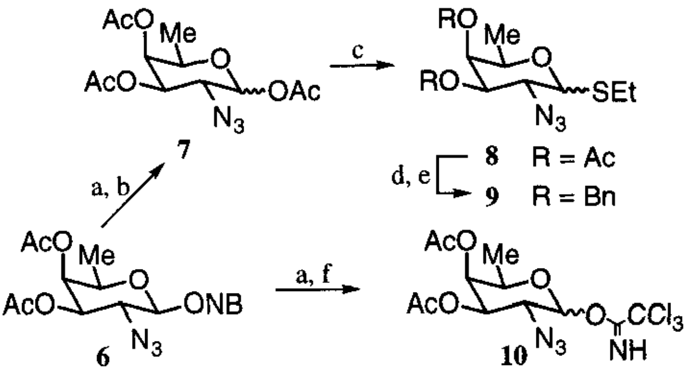
ammonium salt was expected to inhibit such participation. However, β-glycosides were obtained as a sole products. Therefore, we turned to a more conventional method using 2-azidosugars with the lack of such participation. If the azide is transformed to an *N*-methylamino group under mild conditions, it could offer a promising route to **1** and its analogs. In this communication, we examined the stereoselectivity of glycosylation reactions with 2-azido-2,6-dideoxygalactopyranose derivatives under various conditions and the transformation of an azide derivative to *N*-methylfucosamine.

Synthesis of various 2-azido-2,6-dideoxygalactoside derivatives (**7** - **10**) as glycosyl donors was achieved from readily available *o*-nitrobenzylglycoside (**2**),[7] as shown in Schemes 1 and 2.



Scheme 1. Synthesis of 2-azido-2,6-dideoxygalactoside (**6**).

(a) K_2CO_3 (0.1 eq), MeOH, rt, 1 h; (b) $(MeO)_2CMe_2$ (10 eq), TsOH (1.1 eq), DMF, rt, 3 h, 93 % (two steps); (c) $(COCl)_2$ (1.0 eq), DMSO (1.6 eq), Et_3N (3.3 eq), CH_2Cl_2, - 78 °C ~ rt, 2 h; (d) $NaBH_4$ (1.0 eq), MeOH, 0 °C, 10 min, 72 % (two steps); (e) pyridine (6.1 eq), Tf_2O (3.0 eq), CH_2Cl_2, - 30 °C, 1.5 h; (f) Bu_4NN_3 (2.7 eq), DMF, rt, 5 h, 72 % (two steps); (g) $TsOH{\cdot}H_2O$ (2.1 eq), MeOH, rt, 1 h; (h) Ac_2O (15 eq), DMAP (cat), pyridine, rt, 5 h, 93 % (two steps).



Scheme 2. Synthesis of glycosyl donors (**7** - **10**).

(a) hν (high presusure mercury lamp, pyrex filter), THF-pH 7 buffer (10 : 1); (b) Ac_2O (3.5 eq), DMAP (0.8 eq), pyridine, rt, 7 h, 62 % (two steps), α / β = 1 8 / 1; (c) EtSH (1.1 eq), $SnCl_4$ (1.2 eq), CH_2Cl_2, 0 °C, 1.5 h, 71 %, recovery 5 %; (d) K_2CO_3 (cat), MeOH, 2 h; (e) BnCl (4.3 eq), NaH (5.9 eq), DMF, 2 h, 85 % (two steps); (f) CCl_3CN (20 eq), DBU (0.5 eq), CH_2Cl_2, - 30 °C, 3 h, 58 % (two steps), α / β = 3.8 / 1.

We first examined the glycosylation of cyclopentanol with thioglycosides (**8**) and (**9**) using *N*-iodosuccinimide (NIS) / triflic acid[8] (Table 1). The stereoselectivity of the glycosylation with acetate (**8**) depended on the stereochemistry of the anomer of **8** (entries 1 and 2). The α-anomer of **8** gave the α-anomer of **11** predominantly, while the more reactive *o*-benzylthioglycoside (**9**) showed β-selective glycosylation irrespective of the anomer.

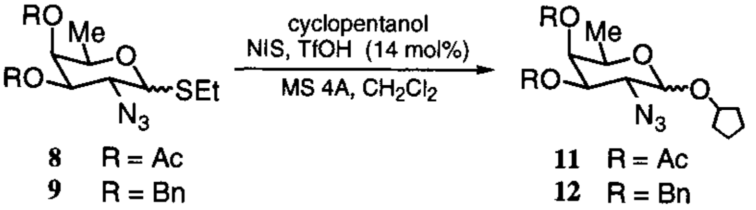


Table 1. Glycosylation of cyclopentanol with **8** or **9**.

entry	donor	temp.	time / h	yield / %	α / β[a]
1	**8** (α-anomer)	- 50 °C ~ rt	13	84	1.5 / 1
2	**8** (β-anomer)	- 50 °C ~ rt	13	89	1 / 2.2
3	**9** (α-anomer)	- 60 °C ~ - 15 °C	3.5	80	1 / 6.1
4	**9** (β-anomer)	- 50 °C ~ rt	15[b]	76	1 / 6.7

[a] The ratio was determined by ^{1}H NMR (200 MHz). [b] The reaction seemed to be completed in a few hours.

Glycosylation of trimethylsilyl ether (**13**) with an anomeric mixture of acetate (**7**) was examined under Mukaiyama conditions using tin(IV) chloride and silver perchlorate as catalysts (Table 2).[9] The α-glycoside (**11**) was obtained as a major product in most solvents,[10] except for acetonitrile.

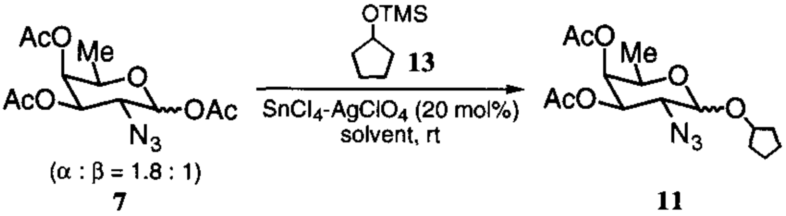
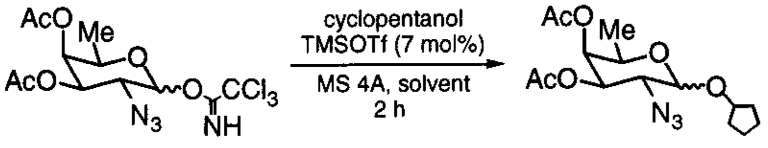


Table 2. Glycosylation of trimethylsilyl ether (**13**) with **7** at room temperature.

entry	solvent	time / h	yield / %	α / β[a]
1	CH_2Cl_2	20	76	2.6 / 1
2	benzene	62	48	1.9 / 1
3	THF	70	12[b]	3.4 / 1
4	MeCN	29	72	1 / 6.1

[a] The ratio was determined by ^{1}H NMR (200 MHz). [b] Recovery of **7** was 65 %.

Furthermore, glycosylation with trichloroacetoimidate (**10**) was examined using trimethylsilyl triflate as a catalyst (Table 3).[11] This reaction appeared to be more rapid than the other reactions (Tables 1 and 2), even at low temperature. Interestingly, the α / β ratio of the product (**11**) in dichloromethane increased with an increase in the reaction temperature. Thus, α / β ratios of 1.4 / 1 and 2.9 / 1 were obtained from the α- and β-anomers of **10**, respectively, at room temperature. On the other hand, improved α-selectivity (2.7 / 1) from the α-anomer of **10** was realized in diethyl ether at - 30 °C.



10 **11**

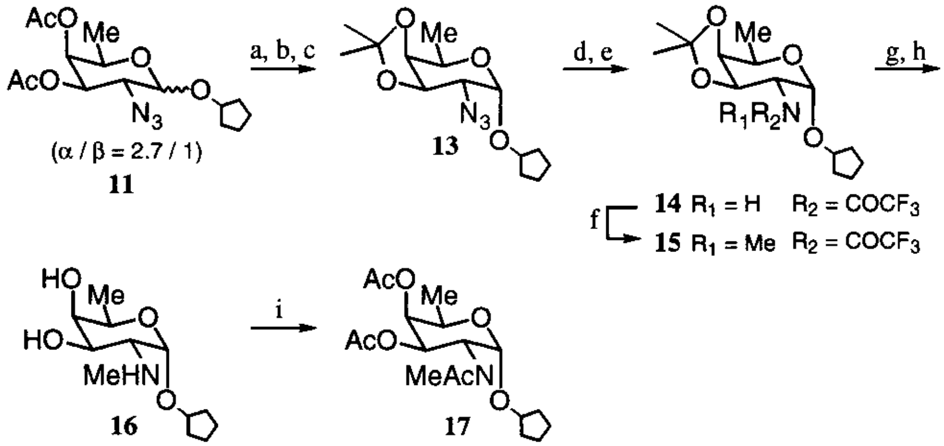
Table 3. Glycosylation of cyclopentanol with **10**.

entry	anomer	solvent	temp.	yield / %	α / β[a]
1	α	CH_2Cl_2	- 30 °C	91	1 / 4.9
2	α	CH_2Cl_2	- 15 °C	67	1 / 3.8
3	α	CH_2Cl_2	0 °C	65	1 / 1.3
4	α	CH_2Cl_2	rt	64	1.4 / 1
5	β	CH_2Cl_2	- 15 °C	65	1 / 1.6
6	β	CH_2Cl_2	rt	83	2.9 / 1
7	β	Et_2O	- 30 °C	65	1.5 / 1
8	α	Et_2O	- 30 °C	84	2.7 / 1
9	α	Et_2O	- 15 °C	85	2.6 / 1
10	α	Et_2O	0 °C	82	2.2 / 1
11	α	Et_2O	rt	80	1.9 / 1

[a] The ratio was determined by ^1H NMR (200 MHz).

Thus, the above examination of glycosylation reactions and the compatibility of the glycosylation conditions with the enediyne functionality in the synthesis of **1** and its analogs[10] suggested that the Schmidt procedure using **10** and trimethylsilyl triflate either in dichloromethane at room temperature or in ether at - 30 °C would be the most appropriate for our purpose.

Conversion of 2-azidoglycoside (**11**) as a 3 / 1 (α / β) anomeric mixture to *N*-methylfucosamine (**16**) was achieved as follows (Scheme 3). The acetate groups of **11** were hydrolyzed and the resulting diol was protected as acetonide (58 % overall yield). Azide (**13**) was treated with triphenylphosphine in aqueous THF, and the resulting amine (**14**) was protected as trifluoroacetamide (86 %). *N*-Methylation[12] of **14** with iodomethane gave **15** in 89 % yield, and deprotection of acetonide in acidic methanol (95 %) followed by carefully controlled alkaline hydrolysis of trifluoroacetamide gave *N*-methylfucosamine (**16**), which was unambiguously identified as triacetate (**17**) (73 % overall yield).[13]



Scheme 3. Conversion from azide to *N*-methylamine derivatives.
(a) K_2CO_3 (cat), MeOH; (b) $(MeO)_2CMe_2$ (15 eq), (-)-10-camphorsulfonic acid (1.8 eq), CH_2Cl_2, 1 h; (c) separation of anomer, 58 % (three steps); (d) Ph_3P (3.5 eq), THF-H_2O (10:1), 16 h; (e) $(CF_3CO)_2O$ (3.5 eq), Et_3N (3.7 eq), CH_2Cl_2, 2 h, 86 % (two steps); (f) KH (8.9 eq), 18-crown-6 (3 6 eq), THF, - 30 °C, 30 min, then MeI (2.3 eq), - 20 °C, 1.5 h, 89 %; (g) TsOH (2.5 eq), MeOH, 2 h, 95 %; (h) K_2CO_3 (1.1 eq), MeOH-H_2O (5:2), rt, 2 h; (i) Ac_2O (28 eq), 4-*N*,*N*-dimethylaminopyridine (1.7 eq), rt ~ 90 °C, 17 h, 73 % (two steps).

In summary, α-selective glycosylation of cyclopentanol with 2-azidoglycoside (**10**) and facile conversion of the resulting azide (**11**) to α-*N*-methylfucosamine (**16**) have been achieved. The present study provides a secure route for the total synthesis of **1** and its α-glycoside analogs.

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13. Representative data for **17**: colorless oil; ^{1}H NMR (200 MHz, $CDCl_3$, a 4:1 mixture of rotational isomers of acetoamide) δ 1.14 (d, J = 6.6 Hz, CH_3 major), 1.15 (d, J = 6.6 Hz, CH_3 minor), 1.59-1.71 (m, cyclopentyl), 1.97 (s, $COCH_3$, minor), 1.98, 2.11 (s, $COCH_3$, major), 2.16 (s, $COCH_3$, major and minor), 2.19 (s, $COCH_3$, minor), 2.82 (s, N-CH_3, minor), 2.87 (s, N-CH_3, major), 4.11-4.25 (m, CH, minor, cyclopentyl), 4.17 (br q, J = 6.6 Hz, CH, major), 4.90 (br d, J = 3.3 Hz, CH, minor), 4.93 (br d, J = 3.3 Hz, CH, major), 5.06 (dd, J = 11.8, 3.9 Hz, CH), 5.29 (d, J = 3.9 Hz, CH, major), 5.33 (d, J = 3.9 Hz, CH, minor), 5.42 (dd, J = 11.8, 3.3 Hz, CH, major), 5.48 (dd, J = 11.8, 3.3 Hz, CH, minor); $[\alpha]_D^{26.5}$ +121° (c 0.61, $CHCl_3$); HRMS (EI, 70 eV) m/z calcd for $C_{17}H_{26}NO_5F_3$: 371.1942 (M^+), found 371.1938 (M^+).

Received, 23rd April, 1997