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Silylene/Oxazolidinone Double-Locked Sialic Acid Building Blocks for Efficient Sialylation Reactions in Dichloromethane

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We describe efficient sialylation reactions in CH_2Cl_2 with the use of silylene/oxazolidinone double-locked sialic acid building blocks. The building blocks were synthesized from 4,5-oxazolidinone-protected phenylthiosialoside. In sialylation reactions towards primary and relatively reactive secondary hydroxy groups on the galactosides, the double-locked building blocks provided desired coupling products in good yields with excellent α-selectivities. In the sialylation reaction with the C3-OH of the galactoside, the double-locked building blocks expressed significantly better α-selectivity in comparison with the results obtained by using the oxazolidinone-locked building block.

Introduction

Chemical sialylation is an important method used to synthesize homogeneous sialylated glycans because of the tertiary anomeric center and the 3-deoxy nature of the sialic acid building blocks.[1] To improve the reaction yield and α-selectivity, three different strategies have been adopted, namely, the use of nitrile solvent, pre-introduction of artificial participating groups, and the tuning of the C5 N-protecting groups.

The use of a nitrile solvent in the sialylation reaction has been adopted for formation of α-sialosides as the major isomer. The nitrile solvent preferentially associates to the β-face of the oxonium ion intermediate upon activation of the anomeric leaving groups.[2] Even though the traditional strategies are very simple, the stereoselective effect is moderate because of the weak association properties of the nitrile group.

To obtain a stereoselective neighboring group participation effect, various artificial auxiliaries have been developed. Such auxiliary groups were loaded on either the C1 carboxyl moiety or at the C3 position. The use of a readily removal C1 ester-type participating group in the sialylation reactions afforded moderate levels of α-selectivity, because of the flexibility of the anomeric configuration.[3] In contrast, sialic acid building blocks containing a C3 thiophenyl or thiocarbonyl group have expressed excellent α-selectivities and reaction yields.[4,5] However, such artificial participating groups demand an additional removal step involving radical reductions.

Direct α-selective sialylation methods using C5 N-substituted sialic acid building blocks such as *N*,*N*-diacetyl, *N*-trifluoroacetyl, phthalimide, and several kinds of carbamates have been developed.[6] Such substitutions of the C5 protections augment the reactivity of the sialic acid building blocks in comparison to that of the C5 acetamide,[6d] and they are expected to provide a higher reaction yield. The combined use of nitrile solvents with this strategy has often assisted stereoselective sialylation reactions.

Recently, potent C4,5-oxazolidinone-protected sialic acid building blocks have been reported by the group of Tanaka–Takahashi.[7] Specifically, building blocks involving the primary alcohol functions at C6 of galactose and at C9 of sialic acid exhibit good reactivity as well as excellent α-selectivity without the nitrile effect. In contrast, such excellent α-selectivity was dramatically diminished in reactions involving the C3-OH group of galactosides.[7b] In such cases, the employment of a nitrile solvent re-established the good α-selectivities.[7d] Additionally, the oxazolidinone-locked building blocks have been remarkably employed for the efficient synthesis of α(2–8/9) oligosialic acids and a huge ganglioside GP1c.[7a,8]

In other reports about glycosylation reactions involving α-galactosylations as well as arabinofuranosylations, bulky silicon-protecting groups on the glycosyl donor conferred stereoselectivity due to its bulkiness and ring constraining effects.[9] We expected that these effects would be applicable to sialylation reactions, and thus we designed novel sialic

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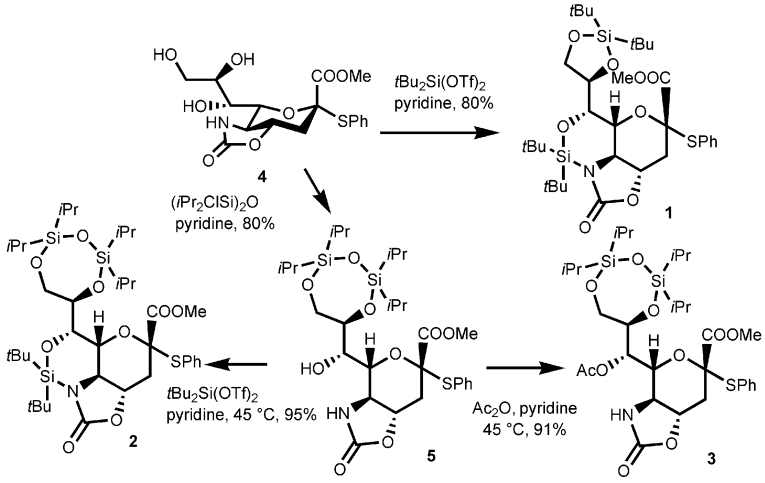


Scheme 1. Synthesis of double-locked sialic acid building blocks **1** and **2** and oxazolidinone building block **3**.

acid building blocks **1** and **2** (Scheme 1). To achieve improved stereoselectivities in the sialylation reactions by tuning the highly reactive oxazolidinone building blocks, bulky di-*tert*-butylsiloxanylidene (DTBS) groups were introduced at the C5,7-positions. The central pyranose ring of building blocks **1** and **2** were doubly constrained with oxazolidinone and DTBS groups. The double-locking might augment the stereoselectivity because the α- and β-faces of the oxonium intermediate could be highly distinguishable. For the purpose, C8,9-DTBS protected building block **1** and C8,9-tetraisopropyldisiloxanylidene (TIPDS) protected building block **2** were prepared to disclose additional steric effects of the C8,9-protecting groups. All sialylation reactions were performed in CH_2Cl_2 to examine whether the unique building block structures are able to control stereoselectivity without the use of a nitrile solvent.

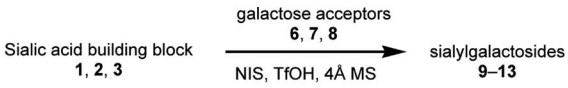
Results and Discussion

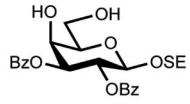
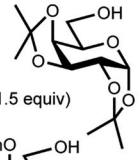
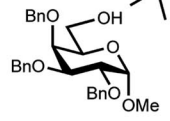
Before investigating the sialylation reactions, the silylene/oxazolidinone double-locked sialic acid building blocks **1** and **2**, as well as oxazolidinone-type building block **3**, were synthesized (Scheme 1). Building block **1**, whose pyranose ring was fixed with a DTBS group at the C5,7-positions and a oxazolidinone at the C4,5-positions, was synthesized from known C4,5-oxazolidinone derivative **4**.[7a,7c] The single-step treatment of **5** with $t\mathrm{Bu_2Si(OTf)_2}$ in pyridine produced building block **1** in 80% yield. Building block **2**, which has bulkier C8,9-TIPDS protections, was synthesized by a two-step procedure. Treatment of **4** with $(i\mathrm{Pr_2SiCl})_2\mathrm{O}$ in pyridine produced **5** in 80% yield, and subsequent introduction of the DTBS group onto the C5,7-positions produced desired **2** in 95% yield. Building block **3**, which has no C5,7-DTBS lock, was also synthesized from intermediate **5** by acetylation in 91% yield.

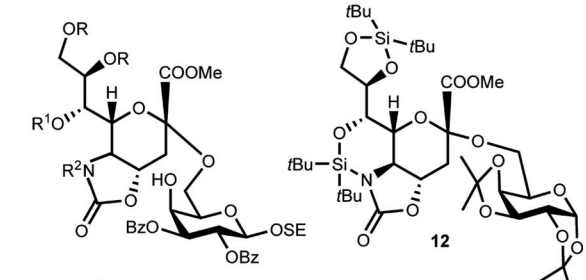
After obtaining building blocks **1**, **2**, and **3**, sialylation reactions to the C6-OH group of galactose acceptors **6**, **7**, and **8** were investigated (Table 1). In all entries, phenylthiosialosides **1**–**3** were activated with *N*-iodosuccinimide (NIS) and TfOH in CH_2Cl_2 at –40 °C in the presence of 4 Å molecular sieves. Initially, C6-OH galactoside **6** was employed and coupled with building blocks **1**–**3**. With the use of double-locked **1**, desired disaccharide **9** was obtained in 92% with excellent α-selectivity (α/β = 22:1; Table 1, Entry 1). Double-locked building block **2** provided disaccharide **10** in 80% yield with complete α-selectivity (Table 1, Entry 2). The use of single-locked **3** similarly provided disaccharide **11** in 83% yield with excellent α-selectivity (α/β = 15:1; Table 1, Entry 3). For primary acceptors, such excellent α-selectivity of oxazolidinone-protected sialic acid building blocks was identical with previous reports.[7] Actually, simple-to-prepare building block **1** expressed potent α-selectivities with galactose acceptors **7** and **8** to give corresponding disaccharides **12** and **13** in good yields (Table 1, Entries 4 and 5). Additionally, no further differences between building block **1** and **2** were found in terms of the stereoselectivity of the sialylation reactions.

Our interest was then shifted toward the secondary C3-OH group of the galactose acceptors (Table 2). In all entries, the reaction conditions were similar to those reported in Table 1. When excellent α-selective results were obtained, the α-configuration was ascertained by $^3J_{C1\text{-}H3ax}$ coupling constants by measuring the phase-sensitive gradient-enhanced HMBC spectra.[10] The other cases, each α/β-isomer was determined by empirical rule.[11] Galactose C3-OH acceptor **14** was initially adopted, and sialylations with double-locked building blocks **1** and **2** and single-locked **3** were investigated. Building block **1** was used with galactose

Table 1. Sialylation reactions using double-locked sialic acid building blocks **1** and **2** and oxazolidinone-type **3** toward the C6-OH group of galactosides **6**, **7**, and **8**.



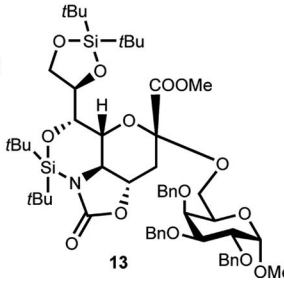
Entry	Acceptor (equiv.)	B.B. (equiv.)	Product (% yield, α/β ratio)
1	 **6** (1.0 equiv)	**1** (1.2)	**9**: (92%, α/β=22:1)
2	**6** (1.5 equiv)	**2** (1.0)	**10**: (80 %, α)
3	**6** (1.5 equiv)	**3** (1.0)	**11**: (83 %, α/β= 15:1)
4	 **7** (1.5 equiv)	**1** (1.0)	**12**: (88%, α)
5	 **8** (1.0 equiv)	**1**(1.2)	**13**: (89%, α)



9: R = R^1, R^2 = DTBS,

10: R = TIPDS, R^1, R^2 = DTBS

11: R = TIPDS, R^1 = Ac, R^2 = H



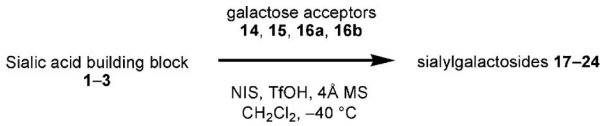
acceptor **14** to give disaccharide **17** in 83% yield in a highly α-selective manner (Table 2, Entry 1). A similar result was obtained with the use of building block **2**. After sialylation, desired disaccharide **18** was stereoselectively produced in 85% yield (Table 2, Entry 2). In contrast, the use of single-locked building block **3** dramatically diminished the α-stereoselectivity, and desired sialoside **19** was afforded in 79% yield with an α/β ratio of 1:2.5 (Table 2, Entry 3).

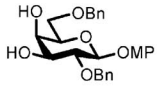
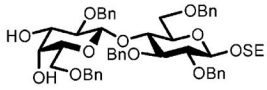
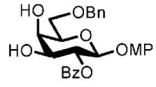
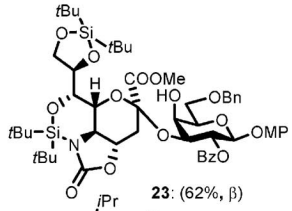
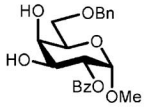
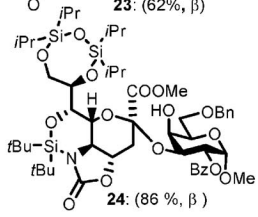
As next task, the C3-OH group of lactose **15**, which would be a practical acceptor toward several gangliosides syntheses, was employed. The sialylation reaction between building block **1** and acceptor **15** provided trisaccharide **20** in 83% yield with moderate α-selectivity (α/β = 1.6:1; Table 2, Entry 4). In contrast, the use of building block **2** produced trisaccharide **21** in 80% yield in a highly α-selective manner (Table 2, Entry 5). As expected, the reaction between single-locked building block **3** and lactoside **15** provided moderate β-selectivity (α/β = 1:1.4) and gave **22** in 61% yield (Table 2, Entry 6). Unfortunately, the high α-selectivity vanished with the use of 2-OBz galactose acceptors **16a/b**. Neither building block **1** nor **2** produced the desired α-anomer of disaccharides **23** or **24** in CH_2Cl_2 (Table 2, Entries 7 and 8). With regard to the C3-OH group of the C2-OBn-protected galactose acceptors, which is relatively reactive, potent α-selectivities were expressed with the use of silylene/oxazolidinone double-locked sialic acid building blocks **1** and **2**. In the comparison with single oxazolidinone-locked building block **3**, both **1** and **2** showed better α-selectivities. When lactose acceptor **15** was submitted for sialylations, building blocks **2** exhibited excellent α-selectivity.

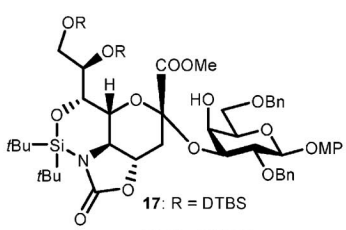
In previous reports, C5,7-oxazolidinone-locked sialic acid building blocks yielded the undesired β-anomer predominantly.[12] Thus, the excellent stereoselectivity of C4,5-oxazolidinone and C5,7-silylene double-locked sialic acid building blocks **1** and **2** were highly intriguing.

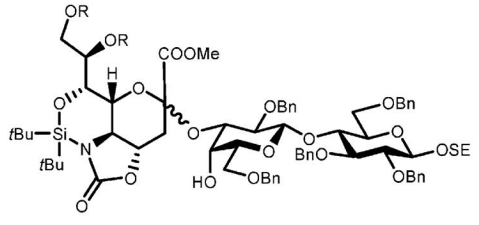
We propose the plausible reaction mechanism indicated in Scheme 2. Before presenting the mechanism, the global minimum structures of key building blocks **1** and **2** were estimated by molecular modeling (MacroModel ver8.1). The global minimum structure of **2** and the superimposed structures within 5 kJ mol^{-1} are shown in Scheme 2. From the model, the tricyclic ring on the C4,5-and C5,7-positions of building block **2** completely separate the *Re* and *Si* faces of the corresponding oxonium ion, and the bulky TIPDS group at the C8,9-positions covers the *Si* face. After NIS–TfOH activation, TfOH approaches from the *Re* face to give presumable intermediates **2a** and **2a′** as described by Crich.[13] With regard to the reaction of reactive alcohols, that is, primary alcohols, as well as C2-OBn-protected **14** and **15**, the HO group would attack from the *Si* face through an S_N2-like pathway. In contrast, less reactive C2-OBz-protected **16** would remain difficult to react with relatively stable intermediate **2a′**. Then, **2a′** would decompose to give highly reactive oxonium ion **2a′′**, which could react with **16** from the *Re* face to settle into the β-anomer. Additionally, the silylene/oxazolidinone double-lock on **1** and **2** would stabilize the conformation of the pyranose ring more than the single oxazolidinone-type building block **3**. Such conformational constraining would stabilize plausible intermediates **2a**/**2a′**, which provide the stereoselective effects for building block **1** and **2**.

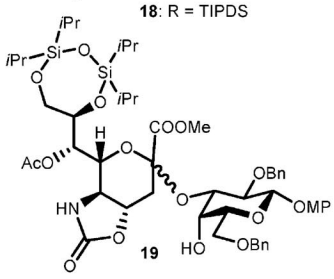
Table 2. Sialylation reactions using double-locked sialic acid building blocks **1** and **2** and oxazolidinone-type **3** toward the 3-OH group of galactosides **14**, **15**, and **16a/b**.

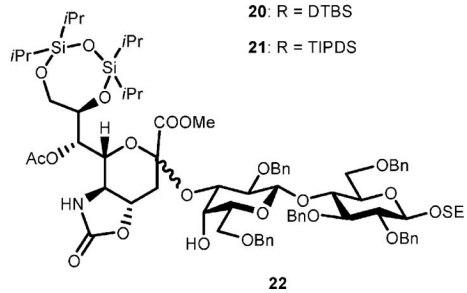


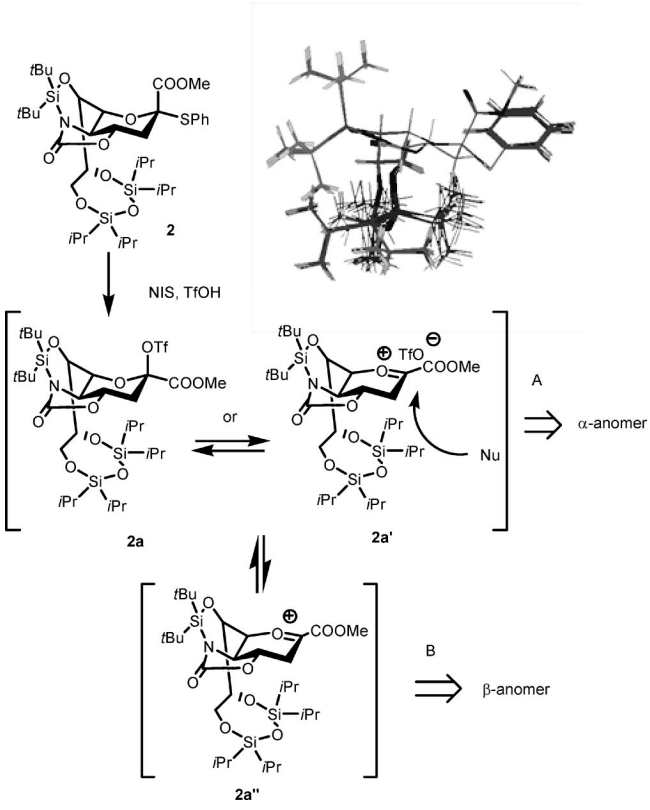
Entry	Acceptor (equiv.)	B.B. (equiv.)	$^3J_{C\text{-}H}$ [Hz] (α-anomer)	Product (% yield, α/β ratio)
1	 **14** (1.0)	**1** (1.2)	5.8	**17**: (83%, α)
2	**14** (1.5)	**2** (1.0)	5.2	**18**: (85%, α)
3	**14** (1.5)	**3** (1.0)	–	**19**: (79% α/β=1:2.5)
4	 **15** (1.5)	**1** (1.0)	5.1	**20**: (83%, α/β = 1.6:1)
5	**15** (1.5)	**2** (1.0)	4.1	**21**: (80%, α)
6	**15** (1.5)	**3** (1.0)	4.4	**22**: (61%, α/β = 1:1.4)
7	 **16a** (1.0)	**1** (1.2)	–	 **23**: (62%, β)
8	 **16b** (1.5)	**2** (1.0)	–	 **24**: (86 %, β)











Scheme 2. Plausible reaction mechanism and global minimum structure of compound **2**. The results of MD simulation within 5 kJ mol^{-1} were overviewed.

Conclusions

We here described efficient sialylation reactions in CH_2Cl_2 using silylene/oxazolidinone double-locked sialic acid building blocks **1** and **2**. Initially, building blocks **1** and **2** were prepared from known oxazolidinone **4**. Then, sialylation reactions using building blocks **1**–**3** were demonstrated in CH_2Cl_2, with no nitrile-supported system. In comparison to the sialylation results obtained using **3**, the results for building blocks **1** and **2**, which have additional C5,7-DTBS lock, provided significant α-selectivities. In the sialylation reaction toward the C6-OH group of galactose acceptors, double-locked **1** and **2** as well as simple oxazolidinone-locked **3** provided the desired α-anomer with excellent α-selectivity. When double-locked building blocks, especially compound **2**, were submitted for sialylation with the C3-OH group of galactosides **14** or **15**, desired coupling products were obtained with excellent α-selectivity.

Experimental Section

Experimental details can be found in the Supporting Information.

Molecular modeling was performed with MacroModel ver8.1 through conformational search program. Conformational profiles were generated by 2000 step Monte Carlo (MCMM) searches with MMFFs force fields in $CHCl_3$ and then reminimized by multiple minimization program with the force fields in order to give a sufficient global minimum when the structure did not reach a gradient to <0.05 kJ Å^{-1} mol^{-1} by C-search.

Supporting Information (see footnote on the first page of this article): Synthetic procedures, spectroscopic data, and ^{1}H and ^{13}C NMR spectra of compounds **1**–**3**, **5**, **9**–**13**, and **17**–**24**.

Acknowledgments

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