

This article was downloaded by:

On: 23 January 2011

Access details: *Access Details: Free Access*

Publisher *Taylor & Francis*

Informa Ltd Registered in England and Wales Registered Number: 1072954 Registered office: Mortimer House, 37-41 Mortimer Street, London W1T 3JH, UK



Journal of Carbohydrate Chemistry

Publication details, including instructions for authors and subscription information:

<http://www.informaworld.com/smpp/title~content=t713617200>

A HIGHLY EFFICIENT SYNTHETIC ROUTE TO $\alpha(2\rightarrow3)/\alpha(2\rightarrow6)$ DISIALYL LEWIS A AS A CANCER ASSOCIATED CARBOHYDRATE ANTIGEN¹

Takayuki Ando^a; Hideharu Ishida^a; Makoto Kiso^a

^a Department of Applied Bioorganic Chemistry, Gifu University, Gifu, Japan

Online publication date: 30 June 2001

To cite this Article Ando, Takayuki, Ishida, Hideharu and Kiso, Makoto (2001) 'A HIGHLY EFFICIENT SYNTHETIC ROUTE TO $\alpha(2\rightarrow3)/\alpha(2\rightarrow6)$ DISIALYL LEWIS A AS A CANCER ASSOCIATED CARBOHYDRATE ANTIGEN¹', *Journal of Carbohydrate Chemistry*, 20: 5, 425 – 430

To link to this Article: DOI: 10.1081/CAR-100105714

URL: <http://dx.doi.org/10.1081/CAR-100105714>

PLEASE SCROLL DOWN FOR ARTICLE

Full terms and conditions of use: <http://www.informaworld.com/terms-and-conditions-of-access.pdf>

This article may be used for research, teaching and private study purposes. Any substantial or systematic reproduction, re-distribution, re-selling, loan or sub-licensing, systematic supply or distribution in any form to anyone is expressly forbidden.

The publisher does not give any warranty express or implied or make any representation that the contents will be complete or accurate or up to date. The accuracy of any instructions, formulae and drug doses should be independently verified with primary sources. The publisher shall not be liable for any loss, actions, claims, proceedings, demand or costs or damages whatsoever or howsoever caused arising directly or indirectly in connection with or arising out of the use of this material.

COMMUNICATION

**A HIGHLY EFFICIENT SYNTHETIC ROUTE TO
 $\alpha(2\rightarrow3)/\alpha(2\rightarrow6)$ DISIALYL LEWIS A AS A CANCER
ASSOCIATED CARBOHYDRATE ANTIGEN¹**

Takayuki Ando, Hideharu Ishida,* and Makoto Kiso*

Department of Applied Bioorganic Chemistry, Gifu University,
Gifu 501-1193, Japan

The carbohydrate determinants, sialyl Lewis A and sialyl Lewis X, which are frequently expressed on human cancer cells,^{2,3} serve as ligands for a cell adhesion molecule of the selectin family.⁴ These carbohydrate determinants are also involved in the adhesion of cancer cells to vascular endothelium and thus contribute to hematogenous metastasis of cancer.^{5,6} We have synthesized a series of the derivatives and the analogs of sialyl Lewis X⁷ and sialyl Lewis A⁸ gangliosides as the versatile probes for elucidation of their biological functions.

The title compound $\alpha(2\rightarrow3)/\alpha(2\rightarrow6)$ disialyl Lewis A (**1**, Figure 1) was first isolated from human colonic adenocarcinoma,⁹ and the determination of the $(2\rightarrow3)$ sialylated Lewis A to $(2\rightarrow3)/(2\rightarrow6)$ disialylated Lewis A ratio in patients with pancreas disease may be helpful for differential diagnosis of pancreas cancer and nonmalignant pancreatic disorders.^{10–12} This paper describes the first, efficient synthetic route to the title compound **1** as its peracetylated derivative **10**, which contains the tri-antennary structure at *O*-3, *O*-4, and *O*-6 of the GlcNAc residue as a structural feature.

The synthetic strategy is illustrated in Figure 1. The crucial point in the synthetic route to the target heptasaccharide **1** was successive glycosylations of a GlcNAc unit of an appropriate trisaccharide in the following order: (A) α -stereo- and regioselective sialylation at *O*-6, (B) regioselective introduction of the sialyl- $\alpha(2\rightarrow3)$ -galactose structure to *O*-3, and (C) fucosylation of the remaining 4-OH of the GlcNAc residue, to give the tri-antennary structure. The order of the glycosylations was decided based on results obtained by our ongoing studies on synthesis of sialoglycoconjugates. It was decided that sialylation should be carried out at the early stage of the synthesis to make isolation of the product less difficult, as sialy-

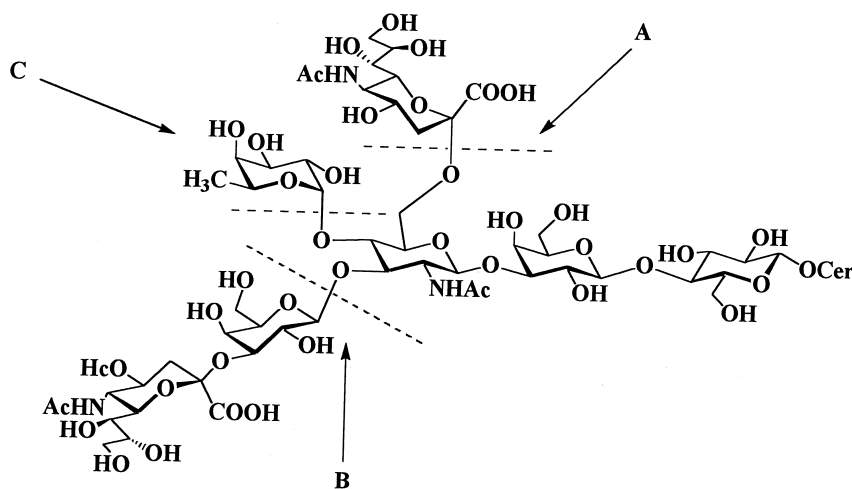


Figure 1. $\alpha(2 \rightarrow 3)/\alpha(2 \rightarrow 6)$ disialyl Lewis A (1).

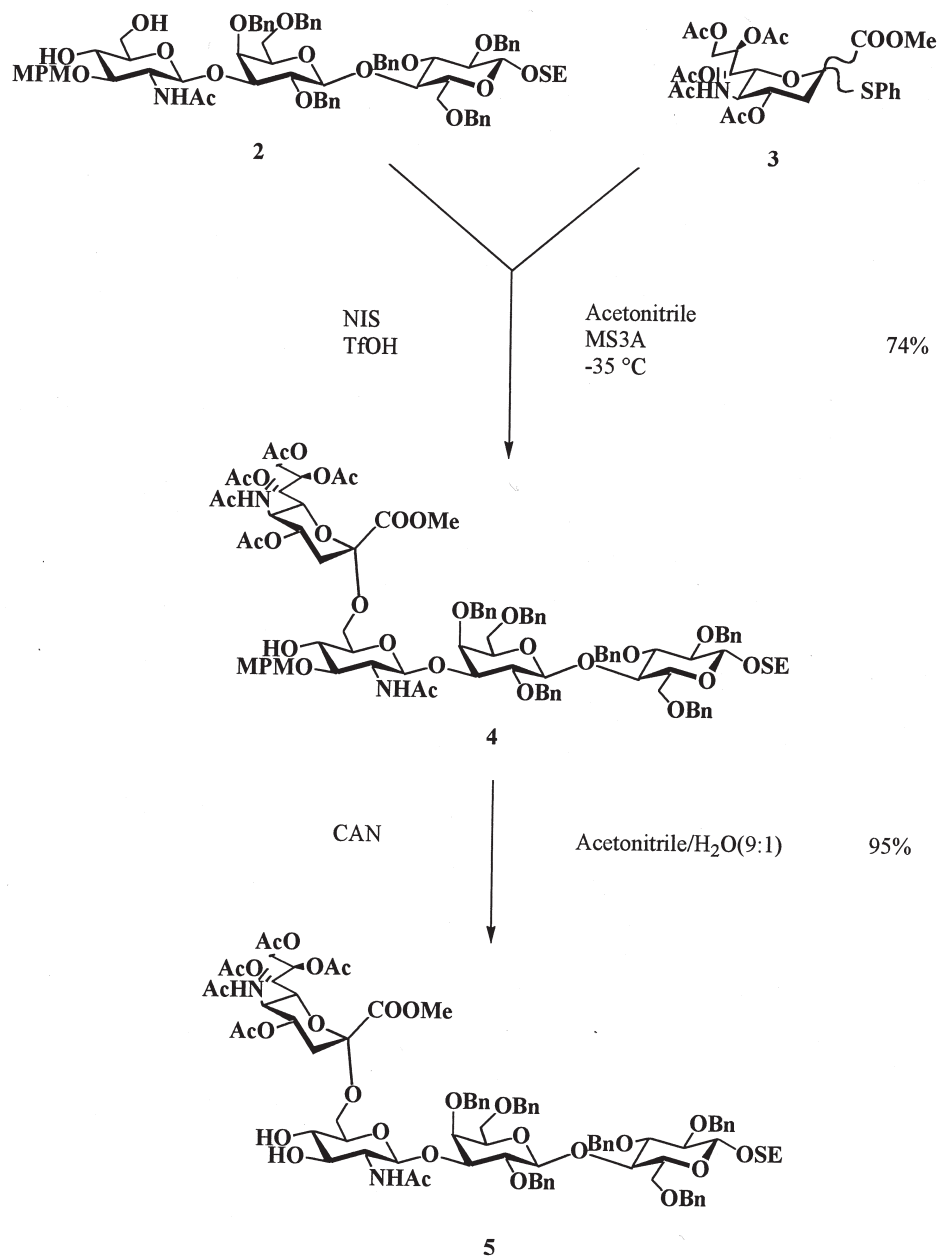
lation of primary hydroxyls often gives anomeric mixtures with relatively low selectivity. Fucosylation was expected to be successfully performed, even at the last stage of the synthesis, because of the observed potency of this reaction.

The α -stereo- and regioselective sialylation of the key trisaccharide acceptor **2**¹³ with **3**,¹⁴ was performed in the presence of *N*-iodosuccinimide (NIS)-trifluoromethanesulfonic acid (TfOH) at $-35\text{ }^{\circ}\text{C}$ in acetonitrile to give the desired sialyl- $\alpha(2 \rightarrow 6)$ -GlcNAc- $\beta(1 \rightarrow 3)$ -Gal- $\beta(1 \rightarrow 4)$ -Glc tetrasaccharide **4** in 74% yield, accompanied by the corresponding β -sialoside (16%) (Scheme 1). The most significant signal in the ^1H NMR spectrum of **4** was a one-proton doublet of doublets ($J_{\text{gem}} = 13.0$, $J_{3\text{eq},4} = 4.8$ Hz) at δ 2.60 due to H-3eq of the newly introduced $\alpha(2 \rightarrow 6)$ -linked sialyl residue.¹⁵ In the spectrum of the β -sialoside, the H-3eq was observed at δ 2.47 ($J_{\text{gem}} = 12.8$, $J_{3\text{eq},4} = 4.76$ Hz).

Treatment of **4** with ceric ammonium nitrate (CAN) in acetonitrile- H_2O (9:1) gave the 3,4-diol tetrasaccharide acceptor **5**, which was glycosylated with the sialyl- $\alpha(2 \rightarrow 3)$ -galactose trichloroacetimidate donor **6**¹⁶ in the presence of trimethylsilyl trifluoromethanesulfonate (TMSOTf) to afford the desired $\beta(1 \rightarrow 3)$ -linked derivative **7** (53%), accompanied by the corresponding $\beta(1 \rightarrow 4)$ -linked hexasaccharide (13%) (Scheme 2). A significant signal in the ^1H NMR spectrum of **7** was a one-proton doublet of doublets at δ 5.48 ($J_{1,2} = 10.3$, $J_{2,3} = 8.01$ Hz, H-2 of Gal) indicating the newly formed glycosidic linkage to be β . The regioselectivity of the glycosylation was confirmed after acetylation of the remaining OH of the GlcNAc residue. The high regioselective glycosylation at O-3 of the GlcNAc residue may be due to steric hindrance at O-4 caused by the (2 \rightarrow 6)-linked sialic acid.

Finally, the remaining, sterically hindered 4-OH of the GlcNAc residue in **7** was efficiently fucosylated with the thiophenyl glycoside of fucose (**8**)¹³ by using the NIS/TfOH-promoted glycosylation procedure in benzene at $7\text{ }^{\circ}\text{C}$. The desired



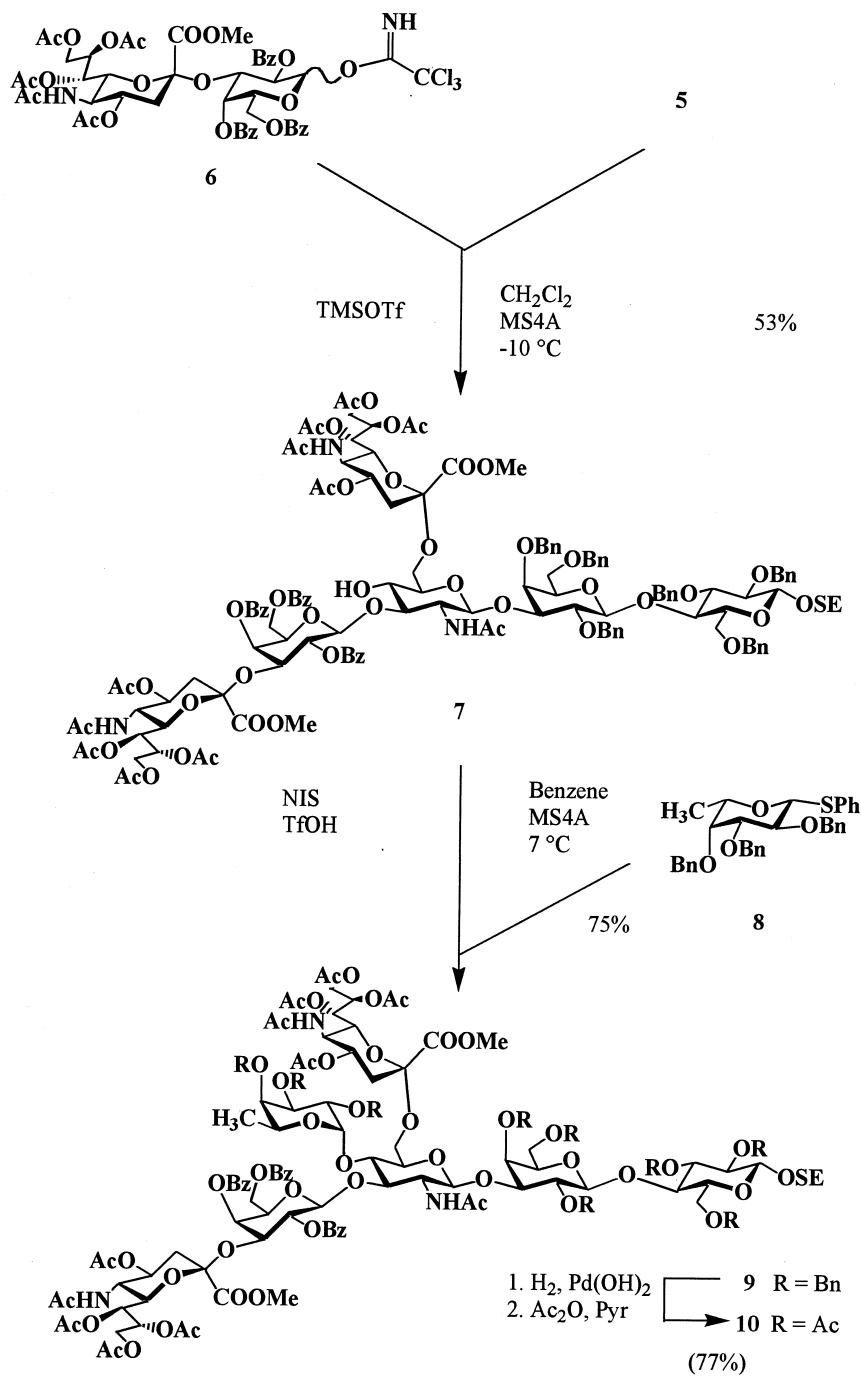


Scheme 1.

heptasaccharide **9** containing the tri-antennary structure based on the GlcNAc residue, was obtained in 75% yield.

Hydrogenolytic removal of the benzyl groups in **9** over Pd(OH)₂ in ethanol, followed by treatment with Ac₂O in pyridine, afforded the peracetylated heptasaccharide **10** in 77% yield.





Scheme 2.



In conclusion, an efficient synthetic route to a derivative form of $\alpha(2\rightarrow3)/\alpha(2\rightarrow6)$ disialyl Lewis A as a cancer associated carbohydrate antigen was achieved for the first time.

ACKNOWLEDGMENT

This work was supported in part by Grans-in-Aid (No. 12306007 and No. 12045228) for Scientific Research from the Ministry of Education, Science and Culture of Japan.

REFERENCES

1. Synthetic studies on sialoglycoconjugates, Part 121. For Part 120, see Otsubo, N.; Yamaguchi, M.; Ishida, H.; Kiso, M. The first, efficient synthesis of novel sLe^x neoglycolipids containing *N*-deacetylated and lactamized sialic acid: key ligand structures for selectin binding. *J. Carbohydr. Chem.* **2001**, in press.
2. Gunnar, C.H.; David, Z. Biosynthesis of the cancer-associated sialyl-Le^a antigen. *J. Biol. Chem.* **1985**, *260*, 9388–9392.
3. Fukushima, K.; Hirota, M.; Terasaki, P.I.; Wakisaka, A.; Togasi, H.; Chia, D.; Suyama, N.; Fukushi, Y.; Nudelman, E.; Hakomori, S. Characterization of sialosylated Lewis X as a new tumor-associated antigen. *J. Biol. Chem.* **1984**, *44*, 5279–5285.
4. Varki, A. Selectin ligands. *Proc. Natl. Acad. Sci. U.S.A.* **1994**, *91*, 7390–7394.
5. Kannagi, R. Carbohydrate-mediated cell adhesion involved in hematogenous metastasis of cancer. *Glycoconjugate J.* **1997**, *14*, 577–584.
6. Nakamori, S.; Furukawa, H.; Hiratsuka, M.; Iwanaga, T.; Imaoka, S.; Ishikawa, O.; Kabuto, T.; Sasaki, Y.; Kameyama, M.; Ishiguro, S.; Irimura, T. Expression of carbohydrate antigen sialyl Le(a): a new functional prognostic factor in gastric cancer. *J. Clin. Oncol.* **1997**, *15*, 816–825.
7. Hasegawa, A.; Kiso, M. Synthesis of sialyl Lewis X ganglioside and analogs. *Methods Enzymol.* **1994**, *242*, 158–173, and references therein
8. Kameyama, A.; Ishida, H.; Kiso, M.; Hasegawa, A. Total synthesis of tumor-associated ganglioside, sialyl Le^a. *J. Carbohydr. Chem.* **1994**, *13*, 641–654.
9. Nudelman, E.; Fukushi, Y.; Levery, S.B.; Higuchi, T.; Hakomori, S.; Novel fucolipids of human adenocarcinoma: disialosyl Le^a antigen (III⁴FucIII⁶NeuAcIV³NeuAcLc⁴) of human colonic adenocarcinoma and the monoclonal antibody (FH7) defining this structure. *J. Biol. Chem.* **1986**, *261*, 5487–5495.
10. Kannagi, R.; Kitahara, A.; Itai, S.; Zenita, K.; Shigeta, K.; Tachikawa, T.; Noda, A.; Hirano, H.; Abe, M.; Shin, S. Quantitative and qualitative characterization of human cancer associated serum glycoprotein antigens expressing epitopes consisting of sialyl-fucosyl type 1 chain. *Cancer Res.* **1988**, *48*, 3856–3863.
11. Itai, S.; Tobe, R.; Kitahara, A.; Kim, Y.C.; Yamabe, H.; Ohtsuki, H.; Kiriara, Y.; Shigeta, K.; Kannagi, R. Significance of 2-3 and 2-6 sialylation of Lewis A antigen in pancreas cancer. *Cancer* **1988**, *61*, 775–787.
12. Itai, S.; Nishikata, J.; Yoneda, T.; Ohmori, K.; Yamabe, H.; Arii, S.; Tobe, T.; Kannagi, R. Tissue distribution of 2-3 and 2-6 sialyl Lewis A antigens and significance



- of the ratio of two antigens for the differential diagnosis of malignant and benign disorders of the digestive tract. *Cancer* **1991**, *67*, 1576–1587.
13. Komba, S.; Ishida, H.; Kiso, M.; Hasegawa, A. Synthesis and biological activities of three sulfated sialyl Le^x ganglioside analogues for clarifying the real carbohydrate ligand structure of L-selectin. *Bioorg. Med. Chem.* **1996**, *4*, 1833–1847.
 14. Marra, A.; Sinaÿ, P. Stereoselective synthesis of 2-thioglycosides of *N*-acetylneuraminic acid. *Carbohydr. Res.* **1989**, *87*, 35–42.
 15. Kanie, O.; Kiso, M.; Hasegawa, A. Glycosylation using methyl thioglycosides of *N*-acetylneuramic acid and dimethyl (methylthio) sulfonium triflate. *J. Carbohydr. Chem.* **1988**, *7*, 501–506.
 16. Hasegawa, A.; Suzuki, N.; Ishida, H.; Kiso, M. Synthesis of ganglioside GM3 and GM4 analogs containing 2- or 3-branched fatty-alkyl residues in place of ceramide. *J. Carbohydr. Chem.* **1996**, *15*, 623–627.

Received April 10, 2001

Accepted May 7, 2001



Request Permission or Order Reprints Instantly!

Interested in copying and sharing this article? In most cases, U.S. Copyright Law requires that you get permission from the article's rightsholder before using copyrighted content.

All information and materials found in this article, including but not limited to text, trademarks, patents, logos, graphics and images (the "Materials"), are the copyrighted works and other forms of intellectual property of Marcel Dekker, Inc., or its licensors. All rights not expressly granted are reserved.

Get permission to lawfully reproduce and distribute the Materials or order reprints quickly and painlessly. Simply click on the "Request Permission/Reprints Here" link below and follow the instructions. Visit the [U.S. Copyright Office](#) for information on Fair Use limitations of U.S. copyright law. Please refer to The Association of American Publishers' (AAP) website for guidelines on [Fair Use in the Classroom](#).

The Materials are for your personal use only and cannot be reformatted, reposted, resold or distributed by electronic means or otherwise without permission from Marcel Dekker, Inc. Marcel Dekker, Inc. grants you the limited right to display the Materials only on your personal computer or personal wireless device, and to copy and download single copies of such Materials provided that any copyright, trademark or other notice appearing on such Materials is also retained by, displayed, copied or downloaded as part of the Materials and is not removed or obscured, and provided you do not edit, modify, alter or enhance the Materials. Please refer to our [Website User Agreement](#) for more details.

[Order now!](#)

Reprints of this article can also be ordered at

<http://www.dekker.com/servlet/product/DOI/101081CAR100105714>