Pergamon

SCIENCE DIRECT®

Bioorganic & Medicinal Chemistry Letters 13 (2003) 2737–2740

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

Structure–Activity Relationships of Azasugar-Based MMP/ADAM Inhibitors

Hideki Moriyama,[a,b] Takahiro Tsukida,[a,b,*,†] Yoshimasa Inoue,[b] Hirosato Kondo,[b,†] Kohichiro Yoshino[b] and Shin-Ichiro Nishimura[c,*]

[a]*Japan Bioindustry Association, Hokkaido Collaboration Center, N-21, W-12, Kita-Ku, Sapporo 001-0021, Japan*
[b]*R&D Laboratories, Nippon Organon K.K., 1-5-90, Tomobuchi-cho, Miyakojima-ku, Osaka 534-0016, Japan*
[c]*Division of Biological Sciences, Graduate School of Science, Hokkaido University, N-21, W-12, Kita-Ku, Sapporo 001-0021, Japan*

Received 17 February 2003; revised 16 May 2003

Abstract—In order to investigate structure–activity relationships of azasugar series toward metalloproteinases, we synthesized and evaluated several azasugar-based compounds. As a result, it was found that 4-phenoxybenzene derivative **3** having 2*R*,3*R*,4*R*,5*S*-configurations exhibited most potent inhibitory activities against matrix metalloproteinase-1, -3 and -9 and TACE.

Metalloproteinases, contained matrix metallo-proteinases (MMPs) and a disintegrin and metallo-proteinases (ADAMs), are a family of zinc-containing enzymes. MMPs, comprised collagenases, stromelysins, gelatinases and membrane-type MMPs (MT-MMPs), mediate the breakdown of connective tissue and are therefore targets for therapeutic inhibitors in many inflammatory, malignant and degenerative diseases.[1–4] On the other hands, ADAMs,[5] structurally related to MMPs, mediate the processing of membrane-bound cytokine such as tumor necrosis factor α (TNF-α) into soluble form. TNF-α is a major mediator of inflammatory and immune responses[6] and a strong inducer of other cytokines such as IL-lβ, IL-6 and IL-8. Elevated TNF-α levels are implicated in pathologies of rheumatoid arthritis,[7] multiple sclerosis,[8] type II diabetes,[9] and so on. Therefore, TNF-α converting enzyme (TACE or ADAM17) is an attractive target for medicinal chemists.[10]

As previously reported, we succeeded to find novel azasugar-based MMP/ADAM inhibitors such as compound **1a**,[11] having 2R,3S,4R,5S-configuration, which exhibited moderate inhibitory activity against MMP-1, -3, -9 and TACE. Next, we focused on the optimization of the inhibitory activity toward MMPs and TACE by synthesizing new analogue of compound **1a**. In the present report, we investigated the effects of stereochemistry at C-3, C-4, and C-5 positions, and property of arylsulfonylamide moiety on biological activity of azasugar-based inhibitors.

Chemistry

Figure 1 shows a synthetic strategy of new analogues of azasugar-based metalloproteinases inhibitors from **1a** as a key compound. Schemes 1 and 2 indicate synthetic routes of analogues appeared in this study.

The azide compounds **6b–d**,[12] which were easily prepared from L-gulono-1,4-lactone **6b**, L-glucono-1,5-lactone **6c** and D-gulono-1,4-lactone **6d**, respectively, were hydrogenated in the presence of palladium on carbon, and then the corresponding amines were reacted with 4-methoxybenzenesulfonyl chloride **7** to give compounds **8b–d** in 72-87% yields (Scheme 1). The terminal isopropylidene group were selectively cleaved by treatment of Muromac® (H^+-form), in 90% MeOH[13a] or cerium chloride heptahydrate and catalytic amount of oxalic acid in acetonitrile[13b] to provide diols **9b–d** in moderate yields. The primary hydroxyl group of diols **9b–d** were selectively mesylated by treatment of mesyl chloride at −40 °C in dichloromethane to afford mesylates **10b–d** in moderate yields. The intramolecular cyclization of compounds **10b–d** in the presence of potassium carbonate

*Corresponding author. Tel.: +81-6-6401-8199; e-mail: takahiro.tsukida@shionogi.co.jp (T. Tsukida); Tel.: +81-11-706-9043; fax: +81-11-706-9042; e-mail: shin@glyco.sci.hokudai.ac.jp (S.I. Nishimura).
†Present address: Manufacturing Technology R&D Laboratories, Shionogi & Co., Ltd., 1-3 Kuise Terajima 2-chome, Amagasaki, Hyogo 660–0813, Japan.

0960-894X/03/$ - see front matter © 2003 Elsevier Ltd. All rights reserved.
doi:10.1016/S0960-894X(03)00530-4

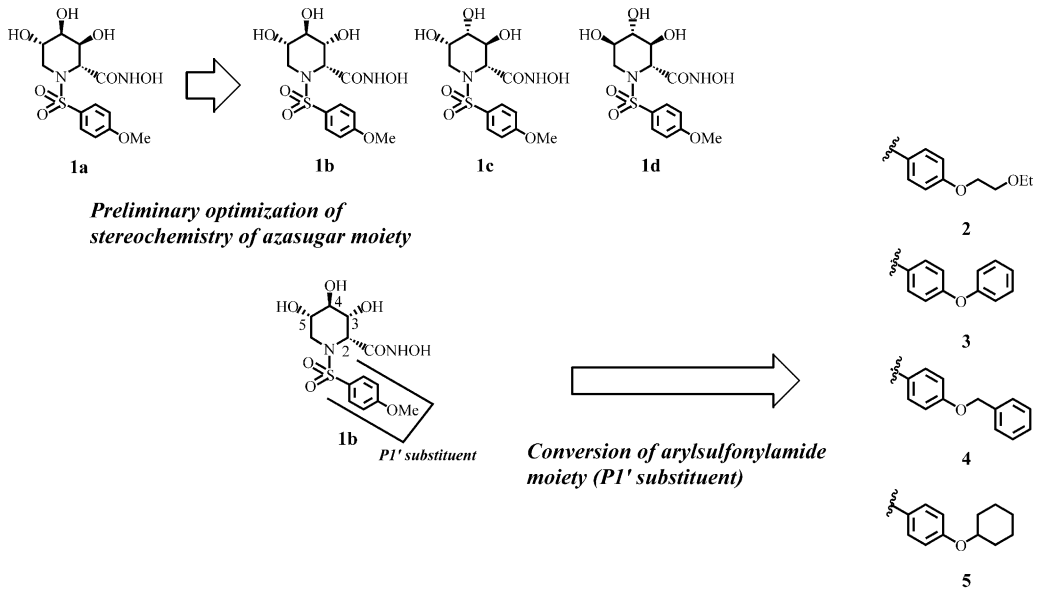
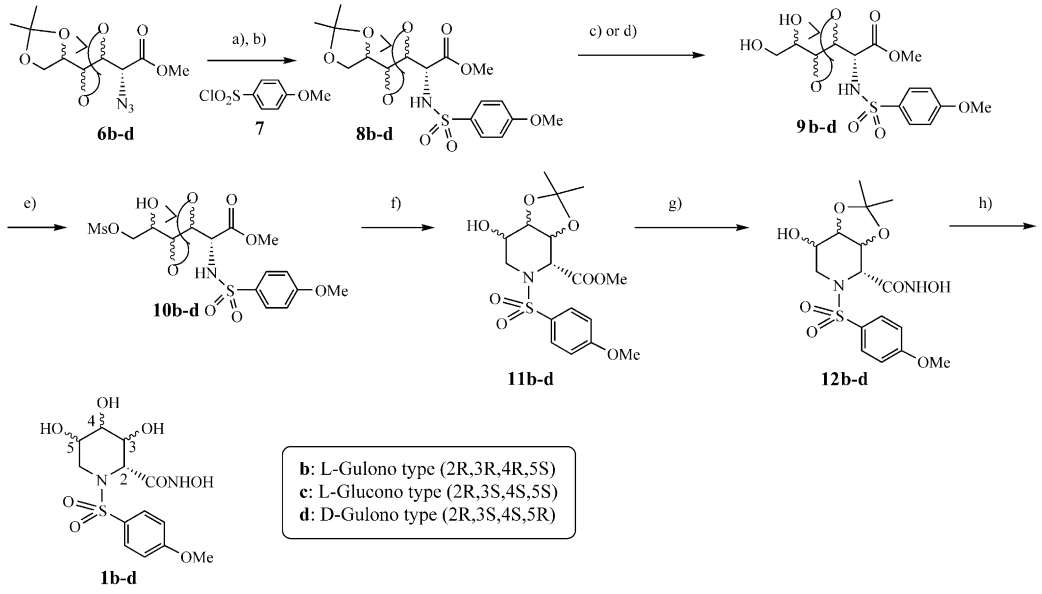


Figure 1. Synthetic strategy of new analogues of azasugar-based metalloproteinase inhibitor.

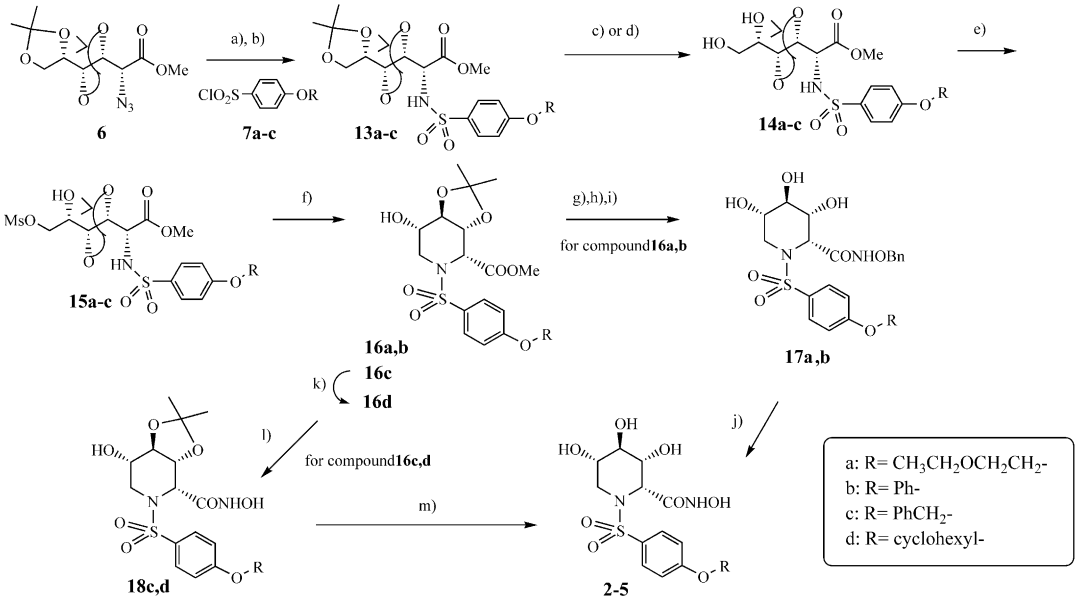


Scheme 1. (a) 10% Pd-C/H_2, EtOAc; (b) 7, 4-DMAP, DMF, 72–87% from **6b–d**; (c) Muromac, 90% MeOH; (d) cerium chloride heptahydrate, oxalic acid, CH_3CN, 53–64%; (e) methanesulfonyl chloride, Et_3N, CH_2Cl_2, 31–58%; (f) K_2CO_3, DMF, 56–100%; (g) 50% NH_2OH aq., NaCN, MeOH, 51–72%; (h) Muromac, MeOH, 66–78%.

gave compounds **11b–d**. Compounds **11b–d** were subjected to aminolysis by the treatment of 50% hydroxylamine aqueous solution in the presence of sodium cyanide in methanol, to afford compounds **12b–d** in 51-72% yield. Finally, 3,4-*O*-isopropylidene group in **12b–d** were cleaved by treatment of Muromac® to provide the target compounds **1b–d** in 66–78% yields.[14] Compounds **2–5** were synthesized, according to Scheme 2, which was same manner to Scheme 1. Compounds **16a**,**b** were subjected to hydrolysis by 1 N sodium hydroxide, followed by the condensation of benzyloxyamine hydrochloride (NH_2OBn) in the presence of 1-(3-dimethylamino-propyl)-3-ethyl-carbodiimide hydrochloride (WSC) and 1-hydroxy-triazole (HOBt) and the deprotection of isopropylidene group using Muromac®, to give compounds **17a**,**b** in moderate yield (Scheme 2). Finally, compounds **17a**,**b** were hydrogenated in the presence of 10% palladium-carbon under hydrogen atmosphere to afford target compounds **2**,**3** in 69–71% yields.[14] On the other hand, after conversion of compound **16c** into **16d**, compounds **16c**,**d** were subjected to aminolysis, to afford compounds **18c**,**d** in 49–62% yields. Finally, 3,4-*O*-isopropylidene group in **18c**,**d** were cleaved by treatment of Muromac® to provide the target compounds **3**,**4** in good yields.[14]

Biological Evaluation

Inhibitory activities against TACE and MMPs (MMP-1, MMP-3, MMP-9) of compound **1a–d** and **2–5** were summarized in Table 1.[15] At first, we focused on the



Scheme 2. (a) 10% Pd-C/H_2, EtOAc; (b) **7a–c**, 4-DMAP, DMF, 69–87% from 6; (c) Muromac, 90% MeOH; (d) cerium chloride heptahydrate, oxalic acid, CH_3CN, 49–68%; (e) methanesulfonyl chloride, Et_3N, CH_2Cl_2, 47–61%; (f) K_2CO_3, DMF, 79–93%; (g) 1N NaOH; (h) NH_2OBn, WSC, HOBt, DMF, 46–51% from **16a**,**b**; (i) Muromac, 90–93%; (j) 10% Pd-C/H_2, MeOH, 69–71%; (k) (i) 10% Pd-C/H_2, EtOAc; (ii) cyclohexanol, DEAD, Ph3P, THF, 49%; (l) 50% NH_2OH aq, NaCN, MeOH, 49–62%; (m) Muromac, 82–85%.

Table 1. Inhibitory activities against MMP-1,3,9, and TACE

Compd	rMMP-1 K_i (nM)	rMMP-3 K_i (nM)	rMMP-9 K_i (nM)	TACE K_i (nM)
1a	84	1.7	157	71
1b	25	7.7	4.8	12
1c	>850	490	780	510
1d	450	85	82	340
2	>850	42	64	8.7
3	8.0	0.51	0.06	2.3
4	850	2.6	6.1	1.6
5	100	1.8	0.93	67
Marimastat	1.1	84	11	0.40

stereochemistry of azasugar component. Compound **1b** bearing 2*R*,3*R*, 4*R*,5*S*-configuration exhibited potent inhibitory activities against all metalloproteinases (MMP-1, -3, -9 and TACE), K_i values were 25 nM against MMP-1, 7.7 nM against MMP-3, 4.8 nM against MMP-9 and 12 nM against TACE, respectively. Accordingly, it was clarified that **1b** showed 2–33 times more potent inhibitory activities against MMP-1, -3, -9 and TACE than compound **1a** having 2*R*,3*S*,4*R*,5*S*-configuration. On the other hand, compound **1c** having 2*R*,3*S*,4*S*,5*S*-configuration was less against all four target enzymes. Moreover, compound **1d** with 2*R*,3*S*,4*S*,5*R*-configuration was also a weak-moderate metalloproteinase inhibitor. Although *R*-configuration at the 2-position of azasugar compound would be essential for binding to MMPs and TACE alike other sulfonamide-based inhibitors,[15,16] it was found that stereochemistry at the 3-, 4-, 5-positions as well as the 2-position of azasugar scaffold would be crucial for the interaction with MMP-1, -3, -9 and TACE. In addition, appropriate stereochemistry at the 3-, 4-, 5-positions of azasugar skeleton could be expected to show broad or selective spectrum for inhibitory activities toward MMPs versus TACE.

Next, we investigated the conversion of arylsulfonylamide (P1′-substituent). The stereo-chemistry of azasugar scaffold was fixed as 2*R*,3*R*,4*R*,5*S*-configuration, due to the potent inhibitory activities of compound **1b** against MMPs and TACE. Compounds **2** and **4**, having 2-ethoxyethoxy and benzyloxy moieties, respectively, exhibited weak activity against MMP-1. This result indicated that 4-(2-ethoxyethoxy)- and 4-benzyloxy-benzenesulfonylamide units would not be accommodated to shallow S1′ pocket in MMP-1.[17] On the other hand, compound **3**, bearing phenoxy moiety, showed most potent inhibitory activities against MMP-1, -3, -9 and TACE, and was 5-80 times more potent than methoxy type **1b**. In addition, inhibitory activities of compound **3** toward MMP-3 and -9 were 165–183 times potent than those of Marimastat, a well-known MMP inhibitor. Interestingly, in the case of cyclohexyloxy type **5**, it was found that inhibitory activities against MMP-1, -9 and TACE were dramatically decreased, compared to compound **3**. From these results, it was suggested that the tail end phenoxy moiety such as in compound **3** would be more preferable for inhibitory activity against MMPs and TACE than the other substituents.

In conclusion, we have synthesized a series of azasugar-based MMP/ADAM inhibitors to develop compound **1a**. As a result, we succeeded to find compound **3**, bearing 2*R*,3*R*,4*R*,5*S*-configuration and 4-phenoxybenzenesulfonylamide moiety, which exhibited desirable potent inhibitory activities against TACE and MMP-1, -3, -9. In addition, azasugar could be expected to improve water solubility, compared to other classes of metalloproteinase inhibitor. Therefore, azasugar-based

compound would be a promising candidate of therapeutic agent for diseases associated with metalloproteinases. We are now investigating in vivo test of azasugar compound **3**.

Acknowledgements

The present work was supported by a grant for 'Research and Development on Glycocluster Controlling Biomolecules' from the New Energy and Industrial Technology Development Organization (NEDO).

References and Notes

1. Michaelides, M. R.; Curtin, M. L. *Curr. Pharm. Des.* **1999**, *5*, 787.
2. Whittaker, M.; Floyd, C. D.; Brown, P.; Gearing, A. J. H. *Chem. Rev.* **1999**, 2735.
3. Beckett, R. P.; Whittaker, M. *Exp. Opin. Ther. Pat.* **1998**, *8*, 259.
4. Rothenberg, M. L.; Nelson, A. R.; Hande, K. R. *Oncologist* **1998**, *3*, 271.
5. Black, R. A.; White, J. M. *Curr. Opin. Cell. Biol.* **1998**, *10*, 654.
6. Bemelmans, M. H.; van Tits, L. J.; Buurman, W. *Crit. Rev. Immunol.* **1996**, *16*, 1.
7. Moreland, L. W.; Baumgartner, S. W.; Schiff, M. H.; Tindall, E. A.; Fleischmann, R. M.; Weaver, A. L.; Ettlinger, R. E.; Cohen, S.; Koopman, W. J.; Mohler, K. *N. Engl. J. Med.* **1997**, *337*, 141.
8. Clements, J. M.; Cossins, J. A.; Wells, G. M.; Corkill, D. J.; Helfrich, K.; Wood, L. M.; Pigott, R.; Stabler, G.; Ward, G. A.; Gearing, A. J.; Miller, K. M. *J. Neuroimmunol.* **1997**, *74*, 85.
9. Morimoto, Y.; Nishikawa, K.; Ohashi, M. *Life Sci.* **1997**, *61*, 795.
10. Nelson, F. C.; Zask, A. *Exp. Opin. Ther. Pat.* **1999**, *8*, 383.
11. Moriyama, H.; Tsukida, T.; Inoue, Y.; Kondo, H.; Yoshino, K.; Nishimura, S. *Bioorg. Med. Chem. Lett.* in press.
12. (a) Kim, J. H.; Yang, M. S.; Lee, W. S.; Park, K. H. *J. Chem. Soc., Perkin Trans. 1* **1998**, 2877. (b) Lee, B. W.; Jeong, I.-Y.; Yang, M. S.; Choi, S. U.; Park, K. H. *Synthesis* **2000**, 1305.
13. (a) Park, K. H.; Yoon, Y. J.; Lee, S. G. *Tetrahedron Lett.* **1994**, *35*, 9737. (b) Xiao, X.; Bai, D. *Synlett* **2001**, 535.
14. All new compounds gave satisfactory characteristics data. Characteristics are given for a representative compound: **1b**: ^{1}H NMR (DMSO-d_6, 250 MHz) δ 3.0–3.7 (m, 5H), 3.84 (s, 3H), 4.20 (d, 1H, J = 5.3 Hz), 7.09 (d, 2H, J = 9.0 Hz), 7.67 (d, 2H, J = 9.0 Hz), 8.82 (s, 1H), 10.67 (s, 1H). MALDI-TOF: 385 ($M + Na^+$), 401 ($M + K^+$).
15. (a) Recombinant human collagenase-1 (MMP-1), stromelysin-1 (MMP-3), gelatinase B (MMP-9) and TNF-α converting enzyme (TACE) were used in our studies. Assay conditions were referred as below: Sawa, M.; Kiyoi, T.; Kurokawa, K.; Kumihara, H.; Yamamoto, M.; Miyasaka, T.; Ito, Y.; Hirayama, R.; Inoue, T.; Kirii, Y.; Nishiwaki, E.; Ohmoto, H.; Maeda, Y.; Ishibushi, E.; Inoue, Y.; Yoshino, K.; Kondo, H. *J. Med. Chem.* **2002**, *45*, 919. (b) Yoshiizumi, K.; Yamamoto, M.; Miyasaka, T.; Ito, Y.; Kumihara, H.; Sawa, M.; Kiyoi, T.; Yamamoto, T.; Nakajima, F.; Hirayama, R.; Kondo, H.; Ishibushi, E.; Ohmoto, H.; Inoue, Y.; Yoshino, K. *Bioorg. Med. Chem.* **2003**, *11*, 433.
16. Tamura, Y.; Watanabe, F.; Nakatani, T.; Yasui, K.; Fuji, M.; Komurasaki, T.; Tsuzuki, H.; Maekawa, R.; Yoshioka, T.; Kawada, K.; Sugita, K.; Ohtani, M. *J. Med. Chem.* **1998**, *41*, 640.
17. (a) Yamamoto, M.; Tsujishita, H.; Hori, N.; Ohishi, Y.; Inoue, S.; Ikeda, S.; Okada, Y. *J. Med. Chem.* **1998**, *41*, 1209. (b) Lovejoy, B.; Welch, A. R.; Carr, S.; Luong, C.; Broka, C.; Hendricks, R. T.; Campell, J. A.; Walker, K. A. M.; Martin, R.; Van Wart, H.; Browner, M. F. *Nat. Struct. Biol.* **1999**, *6*, 217.