Bioorganic & Medicinal Chemistry Letters 18 (2008) 5245–5248

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Structure–activity relationship of *ortho*- and *meta*-phenol based LFA-1 ICAM inhibitors

Edward Yin-Shiang Lin *, Kevin M. Guckian, Laura Silvian, Donovan Chin, P. Ann Boriack-Sjodin, Herman van Vlijmen, Jessica E. Friedman, Daniel M. Scott

Research and Development, Biogen Idec Inc., 14 Cambridge Center, Cambridge, MA 02142, USA

ARTICLE INFO

Article history:
Received 28 July 2008
Revised 14 August 2008
Accepted 18 August 2008
Available online 22 August 2008

Keywords:
LFA-1
LFA-1/ICAM inhibitors
ICAM

ABSTRACT

LFA-1 ICAM inhibitors based on *ortho*- and *meta*-phenol templates were designed and synthesized by Mitsunobu chemistry. The selection of targets was guided by X-ray co-crystal data, and led to compounds which showed an up to 30-fold increase in potency over reference compound **1** in the LFA-1/ICAM1-Ig assay. The most active compound exploited a new hydrogen bond to the I-domain and exhibited subnanomolar potency.

LFA-1 (leukocyte function-associated antigen-1) is a cell surface adhesion protein expressed on all leukocytes, and plays a key role in the inflammatory process. Interaction of LFA-1 with its counter-receptors (intracellular adhesion molecules ICAM-1, -2, -3) promotes the migration of leukocytes to the site of inflammation. Blocking the LFA-1/ICAM interaction is of great interest and has potential in the treatment of autoimmune diseases.[1–4]

Inhibition of the LFA-1/ICAM-1 interaction is possible at two sites, including the metal-ion dependent adhesion site (MIDAS) that is the location of the direct interaction between LFA-1 and the ICAMs. Alternatively, inhibitors may target the I-domain allosteric site (IDAS). Binding at this remote site prevents conformational changes in LFA-1 necessary for interaction with the ICAMs.[5,6]

Previous studies at Abbott Laboratories[7–13] had identified *p*-arylthio cinnamide compounds, as shown in Figure 1, that inhibit the interaction of LFA-1 and ICAM-1 at the allosteric site with moderate potency. This series of compounds is characterized by A and B phenyl rings, connected by a sulfide linker. Appended to the B-ring is a third ring (C-ring) linked via a *trans*-cinnamide moiety. These studies indicate that 2,3-bis(trifluoromethyl)phenyl and 4-carboxypiperidine were the preferred B- and C-rings, respectively. Compound **1** (Fig. 2) was among the most active[12] in inhibiting the interaction of LFA-1 and ICAM-1, and showed 12 nM potency in the LFA-1/ICAM1-Ig assay.[9]

The scope of this study is to improve on the potency of **1** by exploring the structure–activity relationship of substitution on

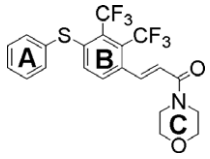


Figure 1. General structure for LFA-1 ICAM inhibitors.

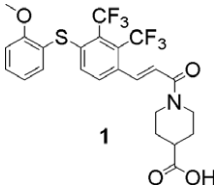


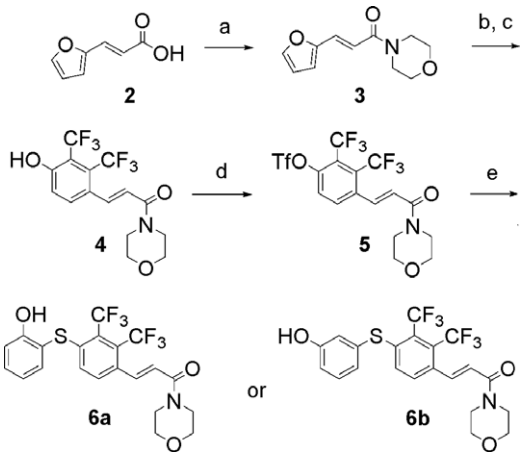
Figure 2. Abbott LFA-1 ICAM inhibitor compound **1**.

the A-ring, while the B- and C-rings are kept constant. Modeling studies suggested potential hydrophobic interactions and hydrogen bonding interactions when the A-ring is properly substituted at the *ortho*- or *meta*-position. This study intends to examine these interactions through the synthesis of appropriate A-ring ethers. Note that morpholine was chosen as a C-ring replacement for 4-carboxypiperidine, as this substitution simplifies the synthesis and had earlier been shown to result in no loss of potency.[12]

Previously reported chemistry[14,15] (Scheme 1) was applied to prepare the common phenol intermediates, compounds **6a** and **6b**. Amide coupling of carboxylic acid **2** to morpholine gave amide **3**. Dienophile hexafluorobutyne was condensed with **3,** which was

* Corresponding author.
E-mail address: ted.lin@biogenidec.com (E.Y.-S. Lin).

0960-894X/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2008.08.062



Scheme 1. Synthetic scheme for preparation of **6a** and **6b**. Reagents and conditions: (a) i—$SOCl_2$, CH_2Cl_2, 0 °C to RT, o/n; ii—morpholine, CH_2Cl_2, RT, 2 h; (b) hexafluoro-2-butyne, dichloroethane, −78 °C to 115 °C, o/n; (c) boron trifluoride diethyl etherate, dichloroethane, reflux, 2 h; (d) triflic anhydride, pyridine, CH_2Cl_2, 0 °C to RT, 2 h; (e) 2-mercaptophenol or 3-mercaptophenol, K_2CO_3, acetone, reflux, 1 h.

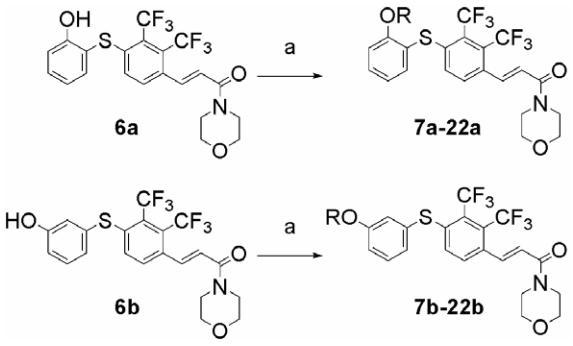
followed by Lewis-acid catalyzed rearrangement to give phenol **4**. Conversion of phenol **4** to the triflate **5** with triflic anhydride was followed by reaction with 2- or 3-mercaptophenol to give the key intermediates, **6a** and **6b**. Phenols **6a** and **6b** were found to be active in the LFA-1/ICAM1-Ig assay, with IC_{50} of 9.7 and 17 nM, respectively.

From phenols **6a** and **6b**, elaboration to the A-ring ethers was accomplished by Mitsunobu reaction with the appropriate alcohols to give the ethers **7a**–**22a** and **7b**–**22b** (Scheme 2). Note that for certain multifunctional alcohols, a subsequent deprotection was required to complete the synthesis.

A series of saturated-ring alcohols was used and the data for the ether products is summarized in Table 1. Each ether shows moderate to high potency in the LFA-1/ICAM1-Ig assay.

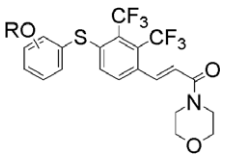
In general, the *ortho*-substituted ethers (**7a**–**14a**) are either equipotent or more active than the *meta*-substituted analogs (**7b**–**14b**).[16] The presence of a polar heteroatom in the ring increased potency, as the piperidine (**8a**, 2 nM) and tetrahydropyran compounds (**9a**, 2.5 nM) were more active than the corresponding cyclohexyl compound (**7a**, 8.9 nM). This suggests that a polar group in this position is preferred. This result is supported by the substituted cyclohexane compounds, **11a**–**14a**. Comparison of 4-methylcyclohexyl (*cis*-**11a**, 18 nM; *trans*-**12a**, 51 nM) and 4-carboxycyclohexyl (*cis*-**13a**, 4 nM; *trans*-**14a**, 1.8 nM) shows that the more polar 4-carboxycyclohexyl is favored.

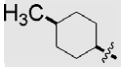
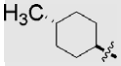
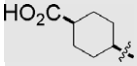
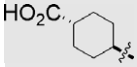
To complement the data on saturated ring ethers, a series of arylmethanols was used. These were chosen to determine the



Scheme 2. Synthetic scheme for preparation of **7a**–**22a** and **7b**–**22b**. Reagents and conditions: (a) R-OH, PPh_3, diisopropyl azodicarboxylate, THF, RT, o/n.

Table 1
Activity of ether compounds: saturated rings[a]



R-	LFA-1 ICAM1-Ig IC_{50} (nM)			
	ortho-		*meta-*	
	**7a**	8.9	**7b**	530
	**8a**	2	**8b**	3.9
	**9a**	2.5	**9b**	34
	**10a**	55	**10b**	2.1
	**11a**	18	**11b**	860
	**12a**	51	**12b**	810
	**13a**	4	**13b**	3
	**14a**	1.8	**14b**	18

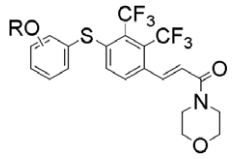
[a] Assay performed as described in the Ref. 9.

effect of aryl rings on the structure–activity relationship, and to see the effect of inserting an additional methylene into the linker. The data for the ether products is summarized in Table 2. As in the previous series, the *ortho*-substituted ethers (**15a**–**20a**) were more potent than the *meta*-substituted analogs (**15b**–**20b**). As a whole, the series shows retention of activity, and in some cases improved activity relative to phenols **6a** or **6b**. Benzyl (**17a**, 24 nM) was tolerated but was less active than each of the pyridinemethyl isomers, **18a**–**20a**. This is consistent with the preference for polar substitutions.

3- and 4-pyridinemethyl-substituted, **19a** and **20a,** were the most active compounds in Table 2, and have IC_{50} of 2.5 and 3.7 nM, respectively. Crystal structure of **19a** and LFA-1 complex (Fig. 3) suggests that no new hydrogen bonds are formed between the protein and the ether-modified A-ring of the small molecule; most of the improved potency is likely due to increased hydrophobic interactions. Note that in the co-crystal structure, the pyridine moiety is exposed to solvent, leading to the preference for pyridinemethyl over the less hydrophilic benzyl. Interestingly, the crystal structure showed that the pyridine ring of **19a** is in close proximity to the phenol OH of Tyr-257 (5.45 Å). Interaction of the small molecule with this residue directly (via a longer linker) or through a water molecule would be expected to increase potency significantly.

The saturated ring series led to the discovery of the polar 4-carboxy substitution. The unsaturated series confirmed the preference for substitution by polar groups and also suggested that an additional methylene in the linker could bring the compound into contact with Tyr-257. Close examination of the X-ray co-crystal structure suggested that the non-planar cyclohexane would be a better scaffold than the planar phenyl for reaching and interacting

Table 2
Activity of ether compounds: unsaturated rings[a]



R-	LFA-1 ICAM1-Ig IC_{50} (nM) ortho-		meta-	
	**15a**	9.5	**15b**	68
	**16a**	11	—	—
	**17a**	24	**17b**	220
	**18a**	9.7	**18b**	70
	**19a**	2.5	**19b**	69
	**20a**	3.7	**20b**	52

[a] Assay performed as described in the Ref. 9.

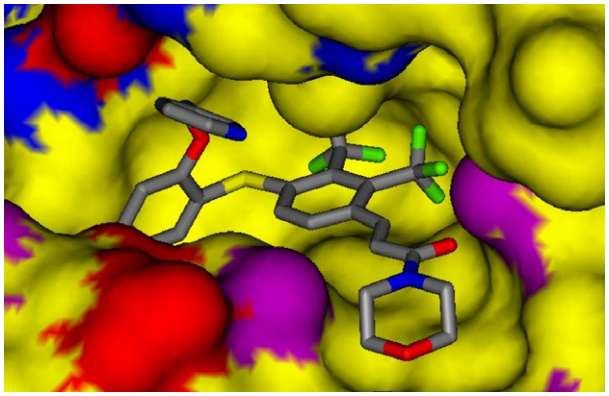


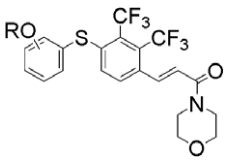
Figure 3. X-ray co-crystal structure of **19a** in the LFA-1 I-domain. RCSB file name (3BQN).

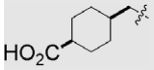
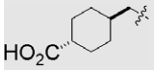
with Tyr-257. Thus, 4-carboxycyclohexanemethyl substituted ethers were synthesized and the data summarized in Table 3.

A significant increase in potency was seen in the best compounds in this series over the compounds in Tables 1 and 2. The *cis*-4-carboxycyclohexanemethyl-substituted **21a** showed 10-fold improvement in potency over the one carbon shorter **13a** and was the most active compound in the series: 0.4 nM in the LFA-1/ICAM1-Ig assay. It is also 6 times more potent than the best aromatic compound shown in Table 2 and 100 times more active than the reference compound, **1**.

An X-ray co-crystal structure of **21a** and LFA-1 was obtained (Fig. 4), which showed a new interaction between the inhibitor and the allosteric site. As hypothesized, the carboxyl group of **21a** interacts with Tyr-257, making a new direct hydrogen bond (3.06 Å) to the side-chain phenol OH. Close analysis of the crystal structure also suggests an additional water-mediated interaction between the carboxylate of **21a** and the side-chain carboxylate of Glu-284. These two interactions are the likely contributors to the observed increase in potency.

Table 3
Activity of ether compounds: 4-carboxycyclohexanemethanol derived ethers[a]



R-	LFA-1 I CAM1-Ig IC_{50} (nM) ortho-		meta-	
	**21a**	0.4	**21b**	3.4
	**22a**	0.6	**22b**	3.4

[a]Assay performed as described in the Ref. 9.

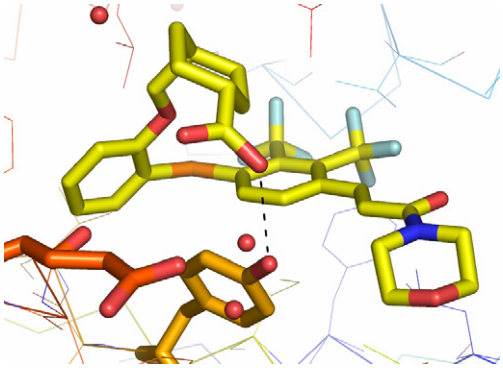


Figure 4. X-ray co-crystal structure of **21a** in the I-domain. RCSB file name (3E2M).

The ether **21a** exhibits poor pharmacokinetic properties when dosed orally in male rats: 9% bioavailability and a 3.6 h iv half-life. This trend was observed for each of the representative *ortho*-analogs studied. However, the less potent *meta*-analog, **13b**, showed improved PK (31% OBA, 2.6 h iv half-life). Further investigation is needed to determine if other *meta*-analogs show similar or better PK, and whether the PK of the *ortho*-series can be improved.

In summary, a new series of potent LFA-1 antagonists featuring A-ring ethers has been discovered. A new hydrogen bond interaction between the carboxylate of the inhibitor and Tyr-257 of the protein was discovered. The best compound in the series is two orders of magnitude more potent than the reference compound **1** and is subnanomolar in the LFA-1/ICAM-Ig assay. These compounds are under further study.

References and notes

1. Giblin, P. A.; Lemieux, R. M. *Curr. Pharm. Des.* **2006**, *12*, 2771.
2. Gottlieb, A.; Krueger, J. G.; Bright, R.; Ling, M.; Lebwohl, M.; Kang, S.; Feldman, S.; Spellman, M.; Wittkowski, K.; Ochs, H. D.; Jardieu, P.; Bauer, R.; White, M.; Dedrick, R.; Garovoy, M. *J. Am. Acad. Dermatol.* **2000**, *42*, 428.
3. Shimaoka, M.; Springer, T. A. *Curr. Topics. Med. Chem.* **2004**, *4*, 1485.
4. Staunton, D. E.; Lupher, M. L.; Liddington, R.; Gallatin, W. M. In *Advances in Immunology*, Academic Press: London, Vol. 91, 2006, p 111.
5. Luo, B.-H.; Carman, C. V.; Springer, T. A. *Ann. Rev. Immun.* **2007**, *25*, 619.
6. Gadek, T. R.; Burdick, D. J.; McDowell, R. S.; Stanley, M. S.; Marsters, J. C., Jr.; Paris, K. J.; Oare, D. A.; Reynolds, M. E.; Ladner, C.; Zioncheck, K. A.; Lee, W. P.; Gribling, P.; Dennis, M. S.; Skelton, N. J.; Tumas, D. B.; Clark, K. R.; Keating, S. M.; Beresini, M. H.; Tilley, J. W.; Presta, L. G.; Bodary, S. C. *Science (Washington, DC, United States)* **2002**, *295*, 1086.
7. Link, J. T.; Sorensen, B.; Liu, G.; Pei, Z.; Reilly, E. B.; Leitza, S.; Okasinski, G. *Bioor. Med. Chem. Lett.* **2001**, *11*, 973.
8. Liu, G.; Huth, J. R.; Olejniczak, E. T.; Mendoza, R.; DeVries, P.; Leitza, S.; Reilly, E. B.; Okasinski, G. F.; Fesik, S. W.; von Geldern, T. W. *J. Med. Chem.* **2001**, *44*, 1202.
9. Liu, G.; Link, J. T.; Pei, Z.; Reilly, E. B.; Leitza, S.; Nguyen, B.; Marsh, K. C.; Okasinski, G. F.; von Geldern, T. W.; Ormes, M. *J. Med. Chem.* **2000**, *43*, 4025.

10. Pei, Z.; Xin, Z.; Liu, G.; Li, Y.; Reilly, E. B.; Lubbers, N. L.; Huth, J. R.; Link, J. T.; von Geldern, T. W.; Cox, B. F.; Leitza, S.; Gao, Y.; Marsh, K. C.; DeVries, P.; Okasinski, G. F. *J. Med. Chem.* **2001**, *44*, 2913.
11. Stewart, A. O.; Bhatia, P. A.; McCarty, C. M.; Patel, M. V.; Staeger, M. A.; Arendsen, D. L.; Gunawardana, I. W.; Melcher, L. M.; Zhu, G.-D.; Boyd, S. A.; Fry, D. G.; Cool, B. L.; Kifle, L.; Lartey, K.; Marsh, K. C.; Kempf-Grote, A. J.; Kilgannon, P.; Wisdom, W.; Meyer, J.; Gallatin, W. M.; Okasinski, G. F. *J. Med. Chem.* **2001**, *44*, 988.
12. Winn, M.; Reilly, E. B.; Liu, G.; Huth, J. R.; Jae, H.-S.; Freeman, J.; Pei, Z.; Xin, Z.; Lynch, J.; Kester, J.; von Geldern, T. W.; Leitza, S.; DeVries, P.; Dickinson, R.; Mussatto, D.; Okasinski, G. F. *J. Med. Chem.* **2001**, *44*, 4393.
13. Zhu, G.-D.; Arendsen, D. L.; Gunawardana, I. W.; Boyd, S. A.; Stewart, A. O.; Fry, D. G.; Cool, B. L.; Kifle, L.; Schaefer, V.; Meuth, J.; Marsh, K. C.; Kempf-Grote, A. J.; Kilgannon, P.; Gallatin, W. M.; Okasinski, G. F. *J. Med. Chem.* **2001**, *44*, 3469.
14. Zhu, G.-D.; Staeger, M. A.; Boyd, S. A. *Organic Letters* **2000**, *2*, 3345.
15. Guckian, K.; Jacobson, I.; Scott, D.; Sowell, C. G. W.O. Patent 2005105766, 2005.; Guckian, K.; Jacobson, I.; Scott, D.; Sowell, C. G. *Chem. Abstr.* **2005**, *143*, 460164.
16. Guckian, K. M.; Lin, E. Y.-S.; Silvian, L.; Friedman, J. E.; Chin, D.; Scott, D. M. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 5249.