Bioorganic & Medicinal Chemistry Letters 20 (2010) 4812–4815

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

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

Structure and activity relationships of tartrate-based TACE inhibitors

Dansu Li [a,*], Janeta Popovici-Muller [a], David B. Belanger [a], John Caldwell [b], Chaoyang Dai [a], Maria David [b], Vinay M. Girijavallabhan [b], Brian J. Lavey [b], Joe F. Lee [b], Zhidan Liu [b], Rob Mazzola [b], Razia Rizvi [b], Kristin E. Rosner [a], Bandarpalle Shankar [b], Jim Spitler [b], Pauline C. Ting [b], Henry Vaccaro [b], Wensheng Yu [b], Guowei Zhou [b], Zhaoning Zhu [b], Xiaoda Niu [c], Jing Sun [c], Zhuyan Guo [b], Peter Orth [b], Shiying Chen [c], Joseph A. Kozlowski [b], Daniel J. Lundell [c], Vincent Madison [b], Brian McKittrick [b], John J. Piwinski [a], Neng-Yang Shih [a], Gerald W. Shipps Jr. [a], M. Arshad Siddiqui [a], Corey O. Strickland [b]

[a] Department of Medicinal Chemistry, Merck Research Laboratories, Cambridge, 320 Bent Street, Cambridge, MA 02141, United States
[b] Department of Medicinal Chemistry, Merck Research Laboratories, 2015 Galloping Hill Road, Kenilworth, NJ 07033, United States
[c] Department of Inflammation, Merck Research Laboratories, 2015 Galloping Hill Road, Kenilworth, NJ 07033, United States

ARTICLE INFO

Article history:
Received 29 April 2010
Revised 15 June 2010
Accepted 21 June 2010
Available online 25 June 2010

Keywords:
TACE inhibitor
Rheumatoid arthritis
Tartrates

ABSTRACT

The syntheses and structure–activity relationships of the tartrate-based TACE inhibitors are discussed. The optimization of both the prime and non-prime sites led to compounds with picomolar activity. Several analogs demonstrated good rat pharmacokinetics.

Tumor necrosis factor-α (TNF-α) is a proinflammatory cytokine that plays a central role in several autoimmune disorders such as rheumatoid arthritis (RA), Crohn's disease, and psoriasis.[1] The current success of anti-TNF-α biologics such as Enbrel®, Remicade®, and Humira® in the treatment of RA has increased interest in discovering an orally active small molecule inhibitor to modulate the level of TNF-α.

One target of interest is TACE (TNF-α converting enzyme), the metalloprotease that releases the soluble 17-kDa form of TNF-α from membrane-bound pro-TNF-α. A TACE inhibitor would have the potential to treat RA by reducing the level of soluble TNF-α.[2] Our focus was to identify an orally active and highly selective TACE inhibitor.[3]

Recently, we disclosed a novel series of tartrate diamide TACE inhibitors[4] such as **1** (Fig. 1). A unique tridentate zinc binding mode was revealed with the tartrate scaffold. Non-prime site amine exploration identified 2-arylpyrrolidines as preferred moieties. Addition of a benzyl group (compound **2**) provided a substantial potency gain compared to **1** by extending into the S3′ region. Herein, we report our structure–activity relationship (SAR) investigation of the prime and non-prime sites. Our goal was to improve potency as well as to achieve acceptable bioavailability.

Our initial efforts focused on the design of the optimal group in the S3′ region, a binding site on the protein which has been found to confer both selectivity and potency in other reported series.[5] It was discovered that switching the 2,4-disubstituted thiophene ring (compound **2**) to a 2,5-analog afforded similar activity (compound **3**). We decided to focus on compound **3** to further explore the S3′ region with modifications of the phenyl ring and the results are depicted in Table 1. Addition of an *ortho*-chloro substituent (**3a**) improved the potency, while either *meta*- or *para*-chloro substituents (**3b** and **3c**) were not beneficial. Changing to an *ortho*-methoxy group further improved the potency to 3 nM (**3d**). Heterocycles were also tolerated, since both the thiophene (**3e**) and the imidazole (**3f**) analogs maintained similar activities as compound **3**. While the naphthalene compound **3g** lost most of the activity, the fused benzimidazoles (**3h** and **3i**) had similar potencies compared to **3d**. The crystal structure of **3i** is shown in Figure 2.[6] Its binding mode is similar to what was reported[4] earlier. As expected, the methylene linker directs the benzimidazole group toward the S3′ region.

Synthesis of benzyl-linked aromatic thiophenes is exemplified in Scheme 1 for compound **3a**. Negishi[7] coupling of commercially available **4** with 2-chlorobenzylzinc chloride, followed by depro-

* Corresponding author. Tel.: +1 617 499 3533.
E-mail address: dansu.li@spcorp.com (D. Li).

0960-894X/$ - see front matter © 2010 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2010.06.104

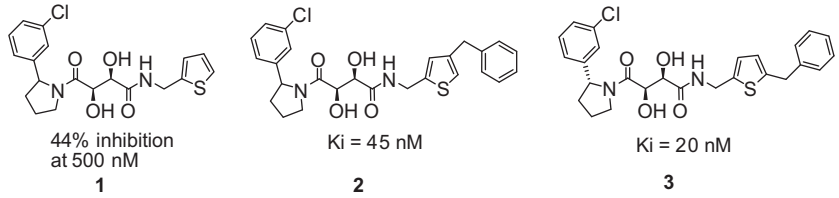
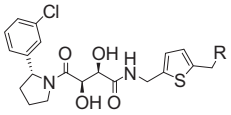
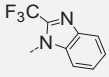


Figure 1.

Table 1
TACE K_i of thienyl-benzyl linked tartrate diamide analogs



Compound	R	TACE K_i (nM)
3		20
3a		9.3
3b		48
3c		37
3d		3
3e		11
3f		37
3g		50% at 500 nM[a]
3h		4
3i		2

[a] The data are reported as % inhibition.

tection gave amine **5**, which reacted with acid **6**[8] to afford compound **3a** after final acetonide deprotection.

The general synthesis of the amines for the nitrogen-linked compounds (**3f**, **3h**, and **3i**) is described in Scheme 2. Protection of 2-thiophenemethanamine (**7**) gave phthalimide **8**, which was converted to thienyl chloride **9** upon treatment with paraformaldehyde/HCl followed by zinc chloride.[9] Alkylation with 2-methylbenzimidazole gave compound **10**. Deprotection with hydrazine hydrate yielded amine **11**, from which **3h** was prepared following the procedures shown in Scheme 1.

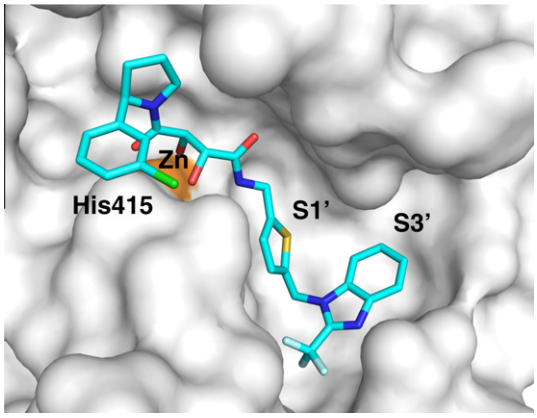
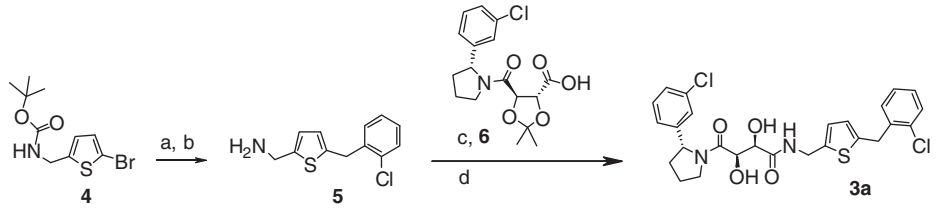


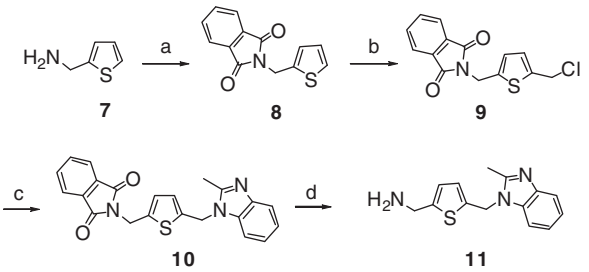
Figure 2. X-ray structure of compound **3i** (stick) bound to TACE catalytic domain (PDB code: 3LGP). The S1′/S3′ loop residues were removed in order to show the inhibitor binding.

Though several S3′ motifs with improved potency were identified (**3d**, **3h**, and **3i**), it was decided to re-examine the SAR for the non-prime amide around compound **3h**. Heterocyclic analogs of 2-phenylpyrrolidines[4] were investigated and the results are shown in Table 2. Of the pyridine compounds (**12a**–**c**), the 4-pyridine (**12b**) was the less favored. While thiophene **12d** was equipotent to **3h**, thiazole analog (**12e**) afforded a slightly less active compound. The introduction of aminothiazole (**12f**) restored the potency. Interestingly, isoindoline analogs **12g** and **12h** provided compounds with excellent enzymatic activity. Although our efforts to find an optimal group in the S3′ region had greatly improved the potency (compound **3** vs **12h**), most of the compounds from this series exhibited poor rat pharmacokinetics (PK) profiles.

In order to maintain good potency and gain oral bioavailability, our efforts were redirected to further explore the SAR of the prime site amide. A series of analogs was prepared to investigate whether the thiophene moiety could be replaced with a six-membered ring (Table 3). Compared to compound **3** (Fig. 1), the phenyl analog **13a** was less potent. To address whether this potency loss was due to a different projection in the S3′ pocket, biphenyl compounds **13b** and **13c** were synthesized and had similar activities as compound **3**. Next replacement of the benzene ring with a heterocycle was explored. Pyridine analog **13d** showed significantly better biochemi-

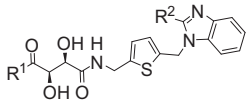


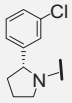
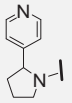
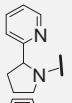
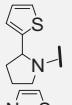
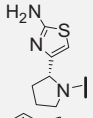
Scheme 1. Reagents and conditions: (a) 2-chlorobenzylzinc chloride, (t-$Bu_3P)_2Pd$, 95%; (b) 4 M HCl/dioxane, 90%; (c) acid **6**, DIEA, DMF, HATU, 64%; (d) 90:10 TFA/water, rt, 87%.



Scheme 2. Reagents and conditions: (a) monomethyl phthalate, EDC, HOBt, Et_3N, DCM, 67%; (b) paraformaldehyde, HCl, 0.5 M $ZnCl_2$/THF, dioxane, 60 °C 78%; (c) 2-methylbenzimidazole, NaI, Cs_2CO_3, DMF, 47%; (d) hydrazine hydrate, EtOH, DCM, Δ, 78%.

Table 2
TACE K_i of thienyl-benzimidazole tartrate diamide analogs



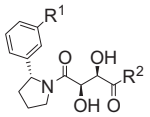
Compound	R^1	R^2	TACE K_i (nM)
3h		CH_3	4
12a[a]		CH_3	12
12b[a]		CH_3	36
12c[a]		CH_3	8
12d[a]		CH_3	4
12e[a]		CH_3	9
12f		CH_3	2
12g		CH_3	0.5
12h		CF_3	0.2

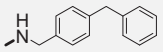
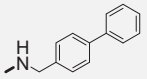
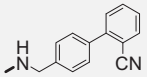
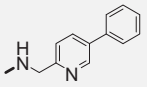
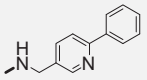
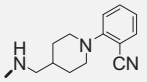
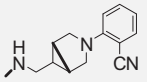
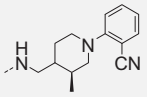
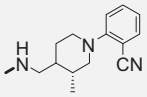
[a] Racemic 2-arylpyrrolidines.

cal potency than its regioisomer **13e**. Changing the biaryl connection to an *N*-arylpiperidine afforded improved K_i (**13f** vs **13c**), while constraining the piperidine into a bicycle gave a slightly less active compound **13g**. Compound **13h** with a (*S*)-methyl piperidine group maintained good enzymatic potency whereas the (*R*)-methyl analog **13i** showed significantly reduced potency. Of the inhibitors examined in Table 3, the biphenyl compound **13b** showed moderate rat PK,[10] with a 6 h AUC of 1500 nM h after a 10 mpk oral dose and it was consequently selected for further SAR profiling.

Based on the observed potency gain with the benzimidazole group in the S3′ pocket (compound **3h**), it was decided to investigate replacement of the distal phenyl group in **13b** with a variety

Table 3
TACE K_i of analogs with different prime site rings



Compound	R^1	R^2	TACE K_i (nM)
13a	Cl[a]		70
13b	H		22
13c	H		17
13d	Cl[a]		6
13e	Cl[a]		307
13f	Cl[a]		4
13g	H		12
13h	H		2
13i	H		315

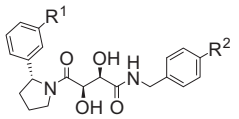
[a] Racemic 3-Cl-phenylpyrrolidine.

of heterocycles (Table 4). The pyridine (**14a** and **14b**) or thiophene (**14d** and **14e**) analogs afforded similar activity. As previously observed, an increase in potency was obtained for compound **14c** (K_i = 5 nM) and **14f** (K_i = 2 nM) with an *ortho*-substitution. The isoxazole (**14g**) and thiadiazole (**14h**) analogs were more active than the thiazoles (**14i** and **14j**). The benzimidazole **14k** and the imidazole **14l** analogs incurred a potency loss, however, switching to the pyrazole **14m** resulted in restoration of the activity to 4 nM. In summary, the prime site amide modification led to compound **14m**, which showed improved enzymatic potency and oral rat PK[10] (AUC = 4400 nM h at 10 mpk).

In an effort to block the potential metabolic oxidation of the benzylic moiety, a methyl substituent was introduced as shown in compound **15** (K_i = 0.86 nM). Only the *R*-orientation of the benzylic methyl group shown in Figure 3 for **15** was tolerated as modeling suggested that this methyl group may make favorable hydrophobic interactions with the side chain of Val402. Both compound **14m** and **15** exhibit good selectivity against a panel of metalloproteases (Table 5). Compound **15** also showed 20% bioavailability in rat (Table 6).

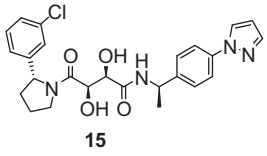
The synthesis of the prime site amine of **15** was accomplished as shown in Scheme 3. Protection of (*R*)-1-phenylethanamine (**16**) as the trifluoroacetamide, followed by iodination[11] afforded the corresponding intermediate **17**, which underwent copper catalyzed cross-coupling with pyrazole. Deprotection with lithium hydroxide gave (*R*)-1-(4-(1H-pyrazol-1-yl)phenyl)-ethanamine (**19**),[8] which was then converted to the desired product **15** via standard transformations.

Table 4
TACE K_i of prime site biaryl tartrate diamide analogs



Compound	R^1	R^2	TACE K_i (nM)
14a	H		36
14b	H		15
14c	H		5
14d	H		28
14e	H		13
14f	H		2
14g	H		8
14h	Cl[a]		3
14i	Cl[a]		18
14j	Cl[a]		74
14k	Cl[a]		296
14l	Cl[a]		270
14m	Cl[a]		4

[a] Racemic 3-Cl-phenylpyrrolidine



15

Figure 3. Compound **15**.

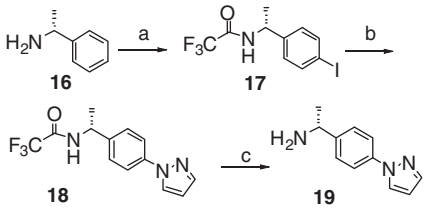
Table 5
Selectivity data for **14m** and **15**

K_i (μM)	**14m**	**15**
TACE	0.004	0.00086
ADAM10	0.602	0.166
MMP1	>100	>100
MMP2	1.9	43.6
MMP3	19	—
MMP7	47	3.8
MMP9	11	88.4
MMP13	3.5	3.3
MMP14	0.12	1.1

We explored the SAR of both the prime and non-prime site amides, which successfully led to potency improvements with two series of TACE inhibitors. Moreover, compound **15** was shown to be potent, selective over a panel of MMPs and orally available in rats. These improvements demonstrate that the tartrate moiety is a viable zinc-coordinating motif. Additional efforts towards further improving potency and oral bioavailability of this series will be discussed in future publications.

Table 6
Rat pharmacokinetic parameters for **15**

PK parameters	
iv	
Dose (mg/kg)	3
Cl (mL/min/kg)	35
AUC (nM h)	3100
po	
Dose (mg/kg)	10
$t_{1/2}$ (h)	0.6
AUC (nM h)	2100
F (%)	20



Scheme 3. Reagents: (a) TFAA, $PhI(O_2CCF_3)_2$, I_2, DCM, 55%; (b) CuI, Cs_2CO_3, 1,10-phenanthroline, DMA, 74%; (c) LiOH, MeOH, 84%.

References and notes

1. (a) Bradley, J. R. *J. Pathol.* **2008**, *214*, 149; (b) Palladino, M. A.; Bahjat, F. R.; Theodorakis, E. A.; Moldawer, L. L. *Nat. Rev. Drug Disc.* **2003**, *2*, 736; (c) Newton, R. C.; Decicco, C. P. *J. Med. Chem.* **1999**, *42*, 2295; (d) Newton, R. C.; Solomon, K. A.; Covington, M. B.; Decicco, C. P.; Haley, P. J.; Friedman, S. M.; Vaddi, K. *Ann. Rheum. Dis.* **2001**, *60*, 25.
2. (a) Black, R. A.; Rauch, C. T.; Kozlosky, C. J.; Peschon, J. J.; Slack, J. L.; Wolfson, M. R.; Castner, B. J.; Stocking, K. L.; Reddy, P.; Srinivasan, S.; Melson, N.; Bioiani, N.; Schooley, K. A.; Gerhart, M.; Davis, R.; Fitzner, J. N.; Johnson, R. S.; Paxton, R. J.; March, C. J.; Cerretti, D. P. *Nature* **1997**, *385*, 729; (b) Moss, M. L.; Jin, S.-L.; Milla, M. E.; Bickett, D. M.; Burkhart, W.; Carter, H. L.; Chem, W. J.; Clay, W. C.; Didsbury, J. R.; Hassler, D.; Hoffman, C. R.; Kost, T. A.; Lambert, M. H.; Leesnitzer, M. A.; McCauley, P.; McGeehan, F.; Mitchell, J.; Moyer, M.; Pahel, G.; Rocque, W.; Overton, L. K.; Schoenen, R.; Seaton, T.; Su, J. L.; Becherer, J. D. *Nature* **1997**, *385*, 733.
3. For recent reviews of TACE inhibitors see: (a) DasGupta, S.; Murumkar, P. R.; Giridhar, R.; Yadav, M. R. *Bioorg. Med. Chem.* **2009**, *17*, 444; (b) Skotnicki, J. S.; Levin, J. I. *Ann. Rep. Med. Chem.* **2003**, *38*, 153; (c) Moss, M. L.; Sklair-Tavron, L.; Nudelman, R. *Nat. Clin. Pract. Rheumatol.* **2008**, *4*, 300.
4. Rosner, K. E.; Guo, Z.; Orth, P.; Shipps, G. W., Jr.; Belanger, D. B.; Chan, T. Y.; Curran, P. J.; Dai, C.; Deng, Y.; Girijavallabhan, V. M.; Hong, L.; Lavey, B. J.; Lee, J. F.; Li, D.; Liu, Z.; Popovici-Muller, J.; Ting, P. C.; Vaccaro, H.; Wang, L.; Wang, T.; Yu, W.; Zhou, G.; Niu, X.; Sun, J.; Kozlowski, J. A.; Lundell, D. J.; Madison, V.; McKittrick, B.; Piwinski, J. J.; Shih, N.-Y.; Siddiqui, M. A.; Strickland, C. O. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 1189.
5. (a) Levin, J. I.; Chen, J. M.; Cheung, K.; Cole, D.; Crago, C.; Santos, E. D.; Du, X.; Khafizova, G.; MacEwan, G.; Niu, C.; Salaski, E. J.; Zask, A.; Cummons, T.; Sung, A.; Xu, J.; Zhang, Y.; Xu, W.; Ayral-Kaloustian, S.; Jin, G.; Cowling, R.; Barone, D.; Mohler, K. M.; Black, R. A.; Skotnicki, J. S. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 2799; (b) Chen, X.-T.; Ghavimi, B.; Corbett, R. L.; Xue, C.-B.; Liu, R.-Q.; Covington, M. B.; Qian, M.; Vaddi, K. G.; Christ, D. D.; Hartman, K. D.; Ribadeneira, M. D.; Trzaskos, J. M.; Newton, R. C.; Decicco, C. P.; Duan, J. J.-W. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 1865.
6. RCSB protein data bank (PDB) deposition number: 3LGP.
7. Negishi, E; Handbook of Organopalladium Chemistry for Organic Synthesis **2002**, *1*, 229.
8. Guo, Z.; Orth, P.; Zhu, Z.; Mazzola, R. D.; Chan, T.-Y.; Vaccaro, H. A.; McKittrick, B.; Kozlowski, J. A.; Lavey, B. J.; Zhou, G.; Paliwal, S.; Wong, S.-C; Shih, N.-Y; Ting, P. C.; Rosner, K. E.; Shipps, G. W., Jr.; Siddiqui, M. A.; Belanger, D. B.; Dai, C.; Li, D.; Girijavallabhan, V. M.; Popovici-Müller, J.; Yu, W.; Zhao, L. WO 2005/121130 (US 2006/0252778).
9. Feigel, M.; Lugert, G.; Heichert, C. *Liebigs Ann. Chem.* **1987**, *4*, 367.
10. Description of the rat PK studies: Following an overnight fast, two male Sprague–Dawley rats (Charles River, Co.) were dosed orally at a dose of 10 mg/kg. Blood was collected into heparin-containing tubes serially from each animal at 0.5, 1, 2, 3, 4, and 6 h post-dosing and centrifuged to generate plasma. Samples at each time point collected from two rats were pooled for LC/MS/MS analysis.
11. Tagat, J. R.; Steensma, R. W.; McCombie, S. W.; Nazareno, D. V.; Lin, S.-I.; Neustadt, B. R.; Cox, K.; Xu, S.; Wojcik, L.; Murray, M. G.; Vantuno, N.; Baroudy, B. M.; Strizki, J. M. *J. Med. Chem.* **2001**, *44*, 3343.