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End-capping of the modified melanocortin tetrapeptide (*p*-Cl)Phe-D-Phe-Arg-Trp-NH_2 as a route to hMC4R agonists

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Abstract—Of the 42 R′-X-(*p*-Cl)Phe-D-Phe-Arg-Trp-NH_2 (X = CO, SO_2, PO, PS) tested at the human (h)MC1, hMC3, and hMC4 receptors (R), the most potent MC4R agonists (EC_{50} of 8–20 nM) were obtained by end-capping with R′ = CH_2=$CHCH_2$ (**9**), $NCCH_2$ (**16**), NH_2COCH_2 (**17**), $HCONHCH_2$ (**18**), CH_3NH (**19**), CH_2=$CHCH_2NH$ (**21**), 2-Th (**23**), $PhCH_2$ (**30**) and X = CO. These compounds possess 35–60-fold hMC4 versus hMC1Rs selectivity with urea LK-71 (**19**) being the most potent at hMC4R and MC4/1R selective (EC_{50} = 8.5 nM, MC4/1R = 100). LK-75 (**16**) combines high potency at hMC4R and MC4/3R selectivity (EC_{50} = 10.5 nM, MC4/3R = 290). SAR is discussed.

Melanocortin receptors (MCRs) belong to the superfamily of seven transmembrane G-protein coupled receptors and stimulate the cAMP signal transduction pathway. Five MCRs have been identified (MC1R–MC5R) and are found both peripherally and in the CNS. These receptors are involved in pigmentation and animal coat coloration (e.g. MC1R), feeding behavior, obesity, diabetes, metabolism, and energy homeostasis (e.g. MC3R and MC4R) as well as exocrine gland function (e.g. MC5R).[1,2] The endogenous agonists for the MCRs, α-, β- and γ-melanocyte stimulating hormones (MSH), and adrenocorticotropin (ACTH), share the central message sequence His-Phe-Arg-Trp responsible for stimulation of MCR.[8–10] The investigation of analogs of the natural α-melanocyte stimulating hormone (α-MSH) Ac-Ser-Tyr-Ser-Met-Glu-**His**^{6}-**Phe**^{7}-**Arg**^{8}-**Trp**^{9}-Gly-Lys-Pro-Val-NH_2 showed that the inversion of Phe^7 configuration gave compounds with increased potency and in vivo stability such as NDP-MSH (MT-I) Ac-Ser-Tyr-Ser-Nle-Glu-**His**^{6}-D-**Phe**^{7}-**Arg**^{8}-**Trp**^{9}-Gly-Lys-Pro-Val-NH_2[3] and MT-II Ac-Nle-c[Asp-**His**^{6}-D-**Phe**^{7}-**Arg**^{8}-**Trp**^{9}-Lys]-NH_2.[4]

Recently, using a series of end-capped analogs of the melanocortin core fragment His^6-D-Phe^7-Arg^8-Trp^9 (HfRW), we have mapped human melanocortin receptors (hMCRs) near the His^6 and designed super-potent hMC1R agonists LK-184 [$Ph(CH_2)_3CO$-HfRW-NH_2, EC_{50} of 0.01][5] and LK-312 [$PhCO(CH_2)_3CO$-HfRW-NH_2, EC_{50} of 0.05][6] with high selectivity for MC1R compared to MC3R and MC4R. Based on the SAR obtained in this series, a model of the region of hMC1, hMC3, and hMC4R adjacent to the His^6 was proposed. It contained a large open hydrophobic area common for MC1, MC3, and MC4Rs and a π-binding zone within this area about three carbons from the N-terminal binding site for hMC1R.[5,6] No potent agonists of hMC4R were discovered within this series.

The main goal of this work was a search for hMC4R agonists with hMC4/1R selectivity that could provide potential anti-obesity agents[1,2,7] as well as an attempt to discriminate hMC1/3/4Rs using end-capping of a modified melanocortin core sequence. The literature data on the role of the His^6 for melanocortin activity are a bit controversial, it seems to be important but not crucial. Deletion of the His from AcHfRW-NH_2 resulted in only a 2-fold drop in potency at hMC4R[8] but a dramatic 228-fold drop at the mouse receptor (mMC4R) and 176-fold drop at mMC1R.[9] Mutation of the His^6 into Gly resulted in a 2-fold drop in potency at MC1R in the frog skin assay.[10] Mutation of the His^6 into Glu or Lys in a cyclic peptide MT-II resulted in an 84-fold drop in potency at hMC4R for Glu or in a 2-fold

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increase for Lys.[11] The initial report that a modified analog ala-Gln-Tyr-Phe-Arg-Trp-Gly-NH_2 (BIM-22015) of ACTH(4–10) Met-Glu-Tyr-Arg-Trp-Gly-OH acts through activation of MCRs[12] was not confirmed later by its binding study.[13] When this work was close to completion, it was reported that mutation of the His^6 into Ala or Pro in AcHfRW-NH_2 resulted in a 90/211-fold drop (Ala/Pro) in potency at mMC1R and a 58/87-fold drop in potency at mMC4R, while its mutation into Phe resulted in only a 25/4-fold drop in potency at mMC1/4Rs. The potencies of the Phe analogs—(4-Py)- and (2-Th)ala at mMC1/4Rs were even closer to those of the parent tetrapeptide.[14] Similarly, mutation of the His^6 into Ala in PrCO-HfRWG-NH_2 resulted in a 60/23-fold drop in potency at hMC1 and hMC4Rs, while its substitution for non-natural aromatic amino acids resulted in MC4 selective compounds.[15]

Amides **1**–**11**, **13**–**18**, **22**–**37** were obtained, analyzed and assayed as previously described.[5] Ureas **19**–**21**, **38** and the carbamate **12** were made by end-capping of the resin bound NH_2-tetrapeptide with the corresponding isocyanates or ethylchloroformate followed by the same cleavage-deprotection as for the amides. Sulfonamides **39**, **40** and phosphonamides **41**, **42** were obtained from the corresponding chlorides similar to the amides. Compounds **1-4**, **13**, **15**, **20**, **22**, **24**, **28**, **29**, **39**, **41**, and **42** had HPLC purity (214 nM) > 95%, compounds **5**–**7**, **9**, **14**, **16**–**19**, **21**, **25**, **27**, **37**, and **38** > 96%, compounds **8**, **10**–**12**, **23**, and **30** > 97%, compounds **26** and **40** > 98%, compounds **31**–**36** > 99%. The EC_{50} values for compounds **1**–**42** reported in Table 1 are the average of at least two separate experiments in duplicate. Unless noted (Table 1), all compounds are full agonists (E_{max} 100–80% relative to MT-I).

As we hypothesized initially,[16,17] the substitution of the His in AcHfRW-NH_2 with (*p*-Cl)Phe (**1**) resulted in a compound with potency at hMC4R close to that of the prototype but with considerably increased MC4 versus MC1Rs selectivity (Table 1). This trend can be seen throughout nearly all (*p*-Cl)FfRW series (**1**–**42**) and is in a sharp contrast with HfRW counterparts of the compounds **1**–**8**, **10**–**16**, **22**, **30**–**36**, **39**, reported by us earlier as being more potent at hMC1R versus hMC4R (in rare cases equipotent).[5,6]

The SAR for end-capped (*p*-Cl)FfRW (Table 1) is also dissimilar to that found for the HfRW series.[5,6] The latter showed little response to the bulkiness, length, polarity or polarizability of R′ at hMC1R (except for the superpotent agonists LK-184 and LK-312, discussed above) and hMC4R, suggesting that the end-capping group does not have to fit into a confined pocket. MC3R was more sensitive to these factors. In the case of end-capping of (*p*-Cl)FfRW, MC1R seems to be least sensitive to the nature of the substituent with EC_{50} of ca. 600 nM for most of the compounds (Table 1). Exceptions are urethane **12** (EC_{50} of 210 nM), which is 4 times more potent than its all carbon isostere **3**, and alkylaromatic subseries **32**–**36** with EC_{50} of 23–144 nM. Though the latter derivatives are analogs of LK-184 and LK-312, there is no dramatic increase in potency for **32**, **34** or its drop for **33**, **35**–**36**, observed in the HfRW series.[5,6] This indicates, that we are dealing with a hydrophobic interaction and not localized π–π binding, characteristic of HfRW derivatives. The drop in potency of **32** and **34** compared to LK-184 and LK-312 may be because the former lack a favorable hydrophilic/hydrogen-bonding interaction provided by the His or because (*p*-Cl)Phe and the alkylaromatic tail are competing for the same hydrophobic binding site at hMC1R. At the same time, a 6-fold increase in potency for **34** relative to **33** (both having C_4 spacers) indicates that polar non-hydrogen-bonding interaction or extended conjugated π-system contributes favorably to the interaction with hMC1R. The latter seems to be the case, since **36** is ca. 3-times more potent than **35**, though they share the same $CO(CH_2)_2$ spacer.

The potencies at hMC3R cover a range from 3000 (**12**) to 35–45 nM (**33**, **34**, **30**, **21**). Out of the entire series only compounds **7**, **11**, **12**, **20**, **30**, **32**–**34**, and **36** are full agonists. Sulfonamides **39**, **40** thiophosphonamide **42** and oxamide **15** have efficacies as low as of 20–26%. Phenylurea **38** has an efficacy of only 6%. If, we assume that the (*p*-Cl)Ph ring of (*p*-Cl)Phe occupies the aromatic binding site of MCRs (or, strictly speaking, an aromatic binding site within the same radius from the N-terminus as the imidazole ring of His), then the observed SAR suggests that the space of hMC3R accommodating the end-capping group is more heterogeneous and/or confined, as compared to hMC1R. In the subseries of simple acids **1**–**11** MC3R favors short bulky hydrophobic *t*-Bu (**7**) but ureas **19**–**21**, bearing a hydrophilic hydrogen-bonding nitrogen next to the N-terminal amide fragment, have better potencies albeit with a slightly lower E_{max} of 70% for NH derivatives **19**, **21**. The presence of a hydrophilic hydrogen-bonding imidazole ring is not crucial for ligands of hMC3R. The most potent phenylaliphatic derivatives with C_1 (**30**) and C_4 spacers (**33**, **34**) are full agonists, that is, in contrast to the hMC1R there is no competition for a hydrophobic binding site at hMC3R. At the same time, a strategic placement of just one N in a shorter linear chain (**21**) is almost equivalent to the contribution of hydrophobic/aromatic term of a Ph ring in **33**.

The potencies at hMC4R show a variance from 4500 (**40**) to 10 nM (**16**, **19**) with all compounds except **40**–**42** being full agonists. In the subseries of simple acids **1**–**11** allyl (**9**), *t*-Bu (**7**), *c*-Pr (**5**) and $MeSCH_2$ (**11**) give the most active compounds at MC4R, while Et (**2**), Pr (**3**), and *i*-Pr (**4**) are the least active. The presence of polar groups both at the end (**14**–**18**) or beginning (**19**–**21**) of a short chain increases both potency and MC4/MC1 selectivity. The importance of polarity or polarizability is clearly seen by comparison of Ph derivative **22** with isosteric thienyl (Th) (**23**, **24**) and pyridyl (Py) (**26**–**28**) analogs, the latter being ca. 5 times more potent than **22**. The potency of 2-furoyl derivative **25** falls into the same range as for the former, while the potency of pentafluorophenyl derivative **29** is close to **22**. The drop in activity is not caused by a larger size of C_6F_5 compared to C_6H_5, since the potency of $PhCH_2$ derivative **30** is close to that of **23**–**28**. Phenylali-

Table 1. Functional activity of melanocortins at human MC1, MC3, and MC4Rs (mean ± SEM)

No.	**R′-(*p*-Cl)FfRW-NH_2** R′	MC-1 EC_{50} (nM)	MC-3 EC_{50} (nM)	MC-4 EC_{50} (nM)	MC 4/1	MC 4/3
	MT-I	**0.07 ± 0.01**	**0.36 ± 0.14**	**0.51 ± 0.07**	0.1	0.7
	AcHFfRW-NH_2	13.0 ± 1.7	170 ± 64	59.5 ± 15.5	0.2	3
LK-184	$Ph(CH_2)_3COHFfRW-NH_2$	0.009 ± 0.004	4.7 ± 1.2	4.6 ± 2.8	0.002	0.002
LK-312	$PhCO(CH_2)_3COHFfRW-NH_2$	0.05 ± 0.01	3.0 ± 1.0	5.0 ± 0	0.01	0.017
1	MeCO	867[a] ± 228	553[b] ± 37	88.5 ± 26.3	10	6
2	EtCO	1982 ± 531	2364[a] ± 183	159[a] ± 39	12	15
3	PrCO	803 ± 143	827[a] ± 121	106 ± 2	8	8
4	*i*-PrCO	565 ± 96	497[a] ± 87	123 ± 36	5	4
5	*c*-PrCO	536 ± 215	473 ± 80	38.5 ± 10.7	14	12
6	BuCO	441 ± 2	460[a] ± 54	155 ± 41	3	3
7	*t*-BuCO	381[a] ± 30	145 ± 25	23.7 ± 7.3	16	6
8	CF_3CH_2CO	1214 ± 80	886[a] ± 153	150 ± 45	8	69
9	$CH_2{=}CHCH_2CO$	856 ± 24	863[a] ± 232	13.7 ± 5.8	62	63
10	$MeOCH_2CO$	502 ± 133	961[a] ± 21	68.7 ± 32.2	7	14
11	$MeSCH_2CO$	378 ± 243	1016 ± 308	39.3 ± 12.7	10	26
12	CH_3CH_2OCO	211 ± 66	3083 ± 854	35.0 ± 22.0	6	88
13	CH_3CH_2OCOCO	1028 ± 19	1481[b] ± 27	131 ± 47	8	11
14	$HOCH_2CO$	499 ± 105	1449[a] ± 545	114 ± 29	4	13
15	NH_2COCO	349 ± 85	164 ± 54	31.3 ± 13.3	11	5
16	$NCCH_2CO$ (LK-75)	567 ± 14	3044[a] ± 56	10.5 ± 4.5	54	290
17	NH_2COCH_2CO	597 ± 68	1013[a] ± 265	14.5 ± 4.7	41	70
18	$HCONHCH_2CO$	600 ± 57	757[b] ± 140	14.1 ± 4.0	43	54
19	CH_3NHCO (LK-71)	848 ± 131	86.5[a] ± 15.8	8.5 ± 1.5	100	10
20	Me_2NCO	2162 ± 323	175 ± 10	42.0 ± 2.0	51	4
21	$CH_2{=}CHCH_2NHCO$	505 ± 95	45.0[a] ± 5.0	12.0 ± 2.0	42	4
22	PhCO	700 ± 133	556[b] ± 72	135 ± 19	5	4
23	2-ThCO	892 ± 36	644[b] ± 25	18.7 ± 4.9	48	34
24	3-ThCO	671 ± 69	674[b] ± 60	27.0 ± 13.7	25	25
25	2-Furoyl	1430 ± 234	913[b] ± 25	29.7 ± 9.9	48	31
26	2-PyCO	947 ± 324	452[b] ± 46	31.3[a] ± 18.4	30	14
27	3-PyCO	577 ± 37	313[b] ± 19	21.0 ± 11.0	28	15
28	4-PyCO	541 ± 76	992[b] ± 331	30.0 ± 16.0	18	33
29	C_6F_5CO	555 ± 173	1340[b] ± 49	168 ± 47	3	8
30	$PhCH_2CO$	699 ± 90	43.0 ± 12.0	20.0 ± 5.0	35	2
31	$Ph(CH_2)_2CO$	442 ± 165	170[a] ± 36	121 ± 10	4	1
32	$Ph(CH_2)_3CO$	113 ± 16	207 ± 38	82.0 ± 3.0	1	3
33	$Ph(CH_2)_4CO$	144 ± 11	34.5 ± 1.5	45.0 ± 5.0	3	1
34	$PhCO(CH_2)_3CO$	23.0 ± 12.8	41.0 ± 1.0	58.0 ± 8.0	0.4	0.7
35	$4\text{-}FC_6H_4CO(CH_2)_2CO$	114 ± 39	203[a] ± 38	50.0 ± 4.0	2	4
36	$3,5\text{-}F_2C_6H_3CO(CH_2)_2CO$	41.0 ± 7.0	140 ± 4	96.0 ± 15.0	0.4	1.5
37	$PhOCH_2CO$	841 ± 123	859[a] ± 70	185 ± 22	4	5
38	PhNHCO	985 ± 208	92.5 ± 4.5	146 ± 27	7	0.6
39	CH_3SO_2	534 ± 11	6700[c] ± 1869	419 ± 125	1	16
40	CF_3SO_2	2157 ± 420	1666[c] ± 24	4527[a] ± 1845	0.5	0.4
41	$(CH_3O)_2PO$	785 ± 257	2635[b] ± 1033	137[a] ± 34	6	19
42	$(CH_3O)_2PS$	1894 ± 354	1604[c] ± 37	446[a] ± 217	4	4

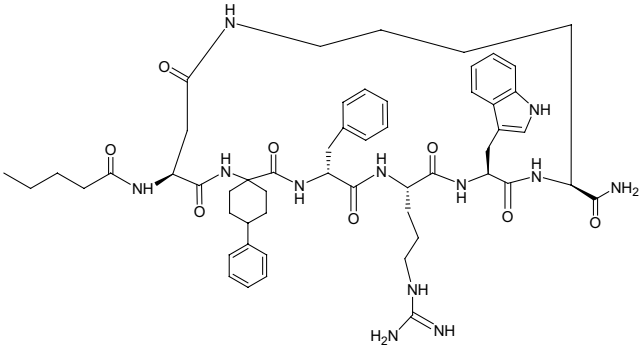
Efficacy (% of MT-I): full agonists (100–80%) unmarked, [a]79–60%, [b]59–30%, [c]29–20%.

phatic derivatives **31**–**35** (especially **34**) with spacers longer than in **30**, showing the best potencies at hMC1R and hMC3R, are 2-3 times less potent at hMC4R compared to **23**–**28** and **30**. Therefore, the end-capping of (4-Cl)Phe-D-Phe-Arg-Trp-NH_2 with short acids bearing polar hydrogen-bonding ($NCCH_2$, NH_2COCH_2, $HCONHCH_2$, CH_3NH, $CH_2{=}CHCH_2NH$) or polarizable hydrophobic ($PhCH_2$, 2-Th, $CH_2{=}CHCH_2$) groups increases the potency at MC4R.

Receptor selectivity of melanocortin agonists is of crucial importance for development of novel drugs, since the natural ligands of MCRs possess sub-nanomolar potency but are essentially nonselective. Table 1 shows that the most potent MC4 agonists **9**, **16**–**19**, **21**, **23**, and **30** are also the most MC4/MC1 selective (MC4/MC1 = 35–100). Out of these, the urea LK-71 (**19**) proved to be not only the most potent (EC_{50} = 8.5 nM) but also the most selective (MC4/1R = 100). LK-75 (**16**) combines high potency at hMC4R with high MC4/3R selectivity (EC_{50} = 10.5 nM, MC4/3R = 290).

Probably, inability of *p*-ClPh in *p*-ClPhe to provide an 'anchoring point' for the 6-position of flexible end-capped tetrapeptides by formation of hydrogen bonds (in contrast to imidazole in His) combined with

heterogeneity (in terms of hydrophobicity/polarity/hydrogen bonding) of the binding site near N-terminus of (*p*-Cl)FfRW-NH_2 is a reason for the absence of very potent ligands in this series, similar to LK-184 and LK-312 in HfRW-NH_2 series. At the same time, due to the ability to adopt a 'right conformation' at the receptor, LK-71 provides the potency and MC1/MC4 selectivity comparable to those of a recently reported extra rigid cyclic peptide (EC_{50} MC1 654 nM, MC4 9 nM) with 1-amino-4-phenylcyclohexane-carboxylic acid in the 6-position of the core sequence mimicking (*p*-Cl)Phe in our linear series.[18]



Thus, the His-D-Phe-Arg-Trp-NH_2 series provides the most MC1 potent and selective compounds when end-capped with a hydrophobic aromatic ring attached through a relatively long C_4 spacer,[5,6] while the (4-Cl)Phe-D-Phe-Arg-Trp-NH_2 series gives the most MC4 potent and selective compounds when the end-capped with a short polar tail (Table 1). Generally speaking, by reversal of the properties of the amino acid in the 6-position and end-capping group, we can engage either hMC1R (polar, hydrophilic, π- and hydrogen bonding His + hydrophobic π-binding tail) or hMC4R (non-polar, hydropoblic (4-Cl)Phe + hydrophilic, hydrogen bonding tail). Taking into account the flexibility of both tetrapeptide scaffolds and degree of conformational freedom of end-capping groups, such sensitivity of MCRs to the nature of substituents in positions 6 and '5' (occupied by the end-capping group) is rather remarkable. Further work is in progress.

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References and notes

1. *The Melanocortin Receptors*; Cone, R. D., Ed.; Humana: Totowa, 2000.
2. Wikberg, J. E. S. *Eur. J. Pharm.* **1999**, *375*, 295.
3. Sawyer, T. K.; Sunfilippo, P. J.; Hruby, V. J.; Engel, V. J.; Heward, C. B.; Burnett, J. B.; Hadley, M. E. *Proc. Natl. Acad. Sci. U.S.A.* **1980**, *77*, 5754.
4. Al-Obeidi, F.; de Castrucci, L.; Hadley, M. E.; Hruby, V. J. *J. Med. Chem.* **1989**, *32*, 2555.
5. Koikov, L. N.; Knittel, J. J.; Solinsky, M. G.; Cross-Doersen D.; Ebetino F. H. *Abstracts of Papers*, 224 National Meeting of the American Chemical Society, Boston, MA; American Chemical Society: Washington, DC, 2002; Abstract 337; Koikov, L. N.; Knittel, J. J.; Solinsky, M. G.; Cross-Doersen, D.; Ebetino, F. H. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 2647; Abdel-Malek, Z. A.; Kavanagh, R. J.; Koikov, L. N.; Todorovic, A.; Haskell-Luevano, C.; McNulty, J. C.; Jackson, P.; Milhauser, G.; Knittel, J. J. *Abstracts of Papers*, 12th Annual Meeting of the Pan-American Society for Pigment Cell Research, Irvine, CA, 2004.
6. Koikov, L. N.; Knittel, J. J.; Solinsky, M. G.; Cross-Doersen, D.; Ebetino, F. H. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 3997.
7. Chaki, S.; Nakazato, A. *Expert Opin. Ther. Patents* **2003**, *11*, 1677.
8. Yang, Y.-K.; Fong, T. M.; Dickinson, C. J.; Mao, C.; Li, J.-Y.; Tota, M. R.; Mosley, R.; Van der Ploeg, L. H. T.; Gantz, I. *Biochemistry* **2000**, *39*, 14900.
9. Haskell-Luevano, C.; Holder, J. R.; Monck, E. K.; Bauzo, R. M. *J. Med. Chem.* **2001**, *44*, 2247.
10. Haskell-Luevano, C.; Hendrata, S.; North, C.; Sawyer, T. K.; Hadley, M. E.; Hruby, V. J.; Dickinson, C.; Gantz, I. *J. Med. Chem.* **1997**, *40*, 2133.
11. Bednarek, M. A.; MacNeil, T.; Kalyani, R. N.; Tang, R.; Lex, H. T.; Van der Ploeg, L. H. T.; Weinberg, D. H. *Biochem. Biophys. Res. Commun.* **2000**, *272*, 23.
12. Strand, F. L.; Saint-Come, C.; Lee, T. S.; Lee, S. J.; Kume, J.; Zuccarelli, L. A. *Peptides* **1993**, *14*, 287.
13. Schioth, H. B.; Muceniece, R.; Wikberg, J. E. S. *Peptides* **1997**, *18*, 761.
14. Holder, J. R.; Bauzo, R.; Xiang, Z.; Haskell-Luevano, C. *J. Med. Chem.* **2002**, *45*, 2801.
15. Cheung, A. W.-H.; Danho, W.; Swistok, J.; Qi, L.; Kurylko, G.; Rowan, K.; Yeon, M.; Franco, L.; Chu, X.-J.; Chen, L.; Yagaloff, K. A. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 133.
16. Mazur, A. W.; Wang, F.; Sheldon, R. J.; Ebetino, F. H. WO Patent 58361, 2000; *Chem. Abstr*. **2000**, *133*, 267160.
17. Ebetino, F. H.; Mazur, A. W.; Hayes, J. C.; Wang, F.; Solinsky, M. G.; Colson, A. O.; Lin, Q. WO Patent 226774, 2001; *Chem. Abstr*. **2002**, *136*, 295093.
18. Sun, H.; Greeley, D. N.; Chu, X.-J.; Cheung, A.; Danho, W.; Swistok, J.; Wang, Y.; Zhao, C.; Chen, L.; Fry, D. C. *Bioorg. Med. Chem.* **2004**, *12*, 2671.