Accepted Manuscript

Challenges in the development of an M_4 PAM preclinical candidate: The discovery, SAR, and biological characterization of a series of azetidine-derived tertiary amides

James C. Tarr, Michael R. Wood, Meredith J. Noetzel, Bruce J. Melancon, Atin Lamsal, Vincent B. Luscombe, Alice L. Rodriguez, Frank W. Byers, Sichen Chang, Hyekyung P. Cho, Darren W. Engers, Carrie K. Jones, Colleen M. Niswender, Michael W. Wood, Nicholas J. Brandon, Mark E. Duggan, P. Jeffrey Conn, Thomas M. Bridges, Craig W. Lindsley

PII: S0960-894X(17)31052-1
DOI: https://doi.org/10.1016/j.bmcl.2017.10.053
Reference: BMCL 25382

To appear in: *Bioorganic & Medicinal Chemistry Letters*

Received Date: 5 June 2017
Revised Date: 18 October 2017
Accepted Date: 22 October 2017

Please cite this article as: Tarr, J.C., Wood, M.R., Noetzel, M.J., Melancon, B.J., Lamsal, A., Luscombe, V.B., Rodriguez, A.L., Byers, F.W., Chang, S., Cho, H.P., Engers, D.W., Jones, C.K., Niswender, C.M., Wood, M.W., Brandon, N.J., Duggan, M.E., Jeffrey Conn, P., Bridges, T.M., Lindsley, C.W., Challenges in the development of an M_4 PAM preclinical candidate: The discovery, SAR, and biological characterization of a series of azetidine-derived tertiary amides, *Bioorganic & Medicinal Chemistry Letters* (2017), doi: https://doi.org/10.1016/j.bmcl.2017.10.053

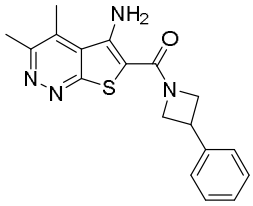
Graphical Abstract

To create your abstract, type over the instructions in the template box below.
Fonts or abstract dimensions should not be changed or altered.

Challenges in the development of an M_4 PAM preclinical candidate: The discovery, SAR, and biological characterization of a series of azetidine-derived tertiary amides

Leave this area blank for abstract info.

James C. Tarr, Michael R. Wood, Meredith J. Noetzel, Bruce J. Melancon, Atin Lamsal, Vincent B. Luscombe, Alice L. Rodriguez, Frank W. Byers, Sichen Chang, Hyekyung P. Cho, Darren W. Engers, Carrie K. Jones Colleen M. Niswender, Michael W. Wood, Nicholas J. Brandon, Mark E. Duggan, P. Jeffrey Conn, Thomas M. Bridges, and Craig W. Lindsley



10a
hM_4 EC_{50} = 94 nM
ACh_{Max} = 65%
rat brain:plasma $K_{p,uu}$ = 2.10
rat AHL = 32% reversal

Bioorganic & Medicinal Chemistry Letters
journal homepage: www.elsevier.com

Challenges in the development of an M_4 PAM preclinical candidate: The discovery, SAR, and biological characterization of a series of azetidine-derived tertiary amides

James C. Tarr,[a] Michael R. Wood,[a,c] Meredith J. Noetzel,[a,b] Bruce J. Melancon,[a] Atin Lamsal,[a,b] Vincent B. Luscombe,[a] Alice L. Rodriguez,[a] Frank W. Byers,[a] Sichen Chang,[a] Hyekyung P. Cho,[a] Darren W. Engers,[a] Carrie K. Jones [a,b] Colleen M. Niswender,[a,b,e] Michael W. Wood,[d] Nicholas J. Brandon,[d] Mark E. Duggan,[d] P. Jeffrey Conn,[a,b,e] Thomas M. Bridges [a,b*] and Craig W. Lindsley [a,b,c*]

[a]*Vanderbilt Center for Neuroscience Drug Discovery, Vanderbilt University School of Medicine, Nashville, TN 37232, USA*
[b]*Department of Pharmacology, Vanderbilt University School of Medicine, Nashville, TN 37232, USA*
[c]*Department of Chemistry, Vanderbilt University, Nashville, TN 37232, USA*
[d]*Neuroscience Innovative Medicines, Astra Zeneca, 141 Portland Street, Cambridge, MA 02139, USA*
[e]*Vanderbilt Kennedy Center, Vanderbilt University School of Medicine, Nashville, TN 37232, USA*

ARTICLE INFO

Article history:
Received
Revised
Accepted
Available online

Keywords:
M4
Muscarinic acetylcholine receptor
Positive Allosteric modulator (PAM)
Schizophrenia
Azetidine

ABSTRACT

Herein we describe the continued optimization of M_4 positive allosteric modulators (PAMs) within the 5-amino-thieno[2,3-*c*]pyridazine series of compounds. In this letter, we disclose our studies on tertiary amides derived from substituted azetidines. This series provided excellent CNS penetration, which had been challenging to consistently achieve in other amide series. Efforts to mitigate high clearance, aided by metabolic softspot analysis, were unsuccessful and precluded this series from further consideration as a preclinical candidate. In the course of this study, we found that potassium tetrafluoroborate salts could be engaged in a tosyl hydrazone reductive cross coupling reaction, a previously unreported transformation, which expands the synthetic utility of the methodology.

Following demonstration of clinical efficacy by xanomeline, a pan muscarinic acetylcholine receptor (mAChR) agonist, to treat multiple symptom clusters of schizophrenia,[1-3] selective activation of the mAChR subtype M_4 has emerged as a promising approach for the treatment of numerous CNS disorders.[4-26] While

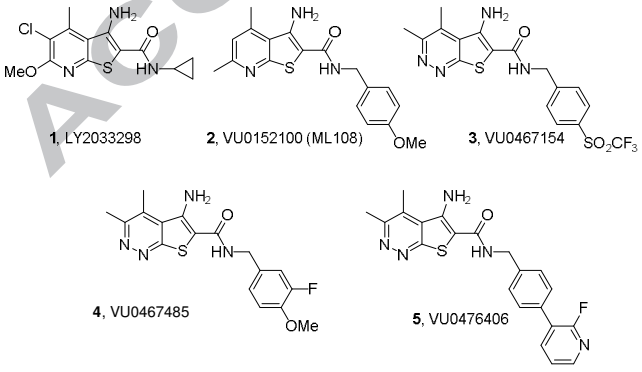


Figure 1. Structures of previously reported M_4 PAMs

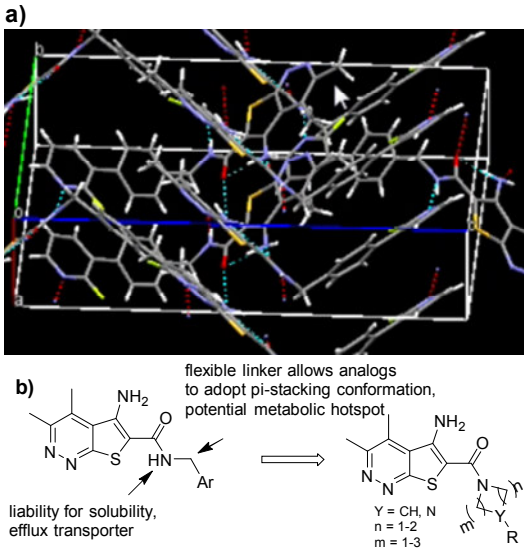


Figure 2. a) X-ray diffraction of **5** b) rationale for tertiary amide design

achieving mAChR subtype selectivity at the highly conserved orthosteric acetylcholine binding site has remained challenging, an alternative strategy of targeting an allosteric site with less evolutionary pressure towards conservation via positive allosteric modulators (PAMs) has met with success. This allosteric approach has not only proven to be a mechanism to potentiate the response of the M_4 receptor to acetylcholine, but also to obtain excellent selectivity across the mAChR subtypes. Our group and others have reported a number of potent, selective M_4 PAMs which have shown efficacy in preclinical animal models of schizophrenia (Figure 1).[4-7,11,14,22-26]

Recently, our group reported the discovery of VU0476406 (**5**),[25] which displayed excellent potency (hM_4 EC_{50} = 91 nM, ACh_{Max} = 74±3%), clean ancillary pharmacology (including high mAChR subtype selectivity), and an early DMPK profile suitable for evaluation as a preclinical candidate (CL_p = 1.6 mL/min/kg, $t_{½}$ = 2.7 h, K_p = 0.11, $K_{p,uu}$ = 1.2). One of the key setbacks found during the characterization of VU0476406, which precluded its advancement into IND-enabling studies, was its low solubility (<0.5 μM at pH 7.4) and solubility-limited oral absorption in dog (%*F* = 4.7 as HCl salt). An x-ray crystal structure of **5** showed extensive hydrogen bonding and π-stacking between neighboring

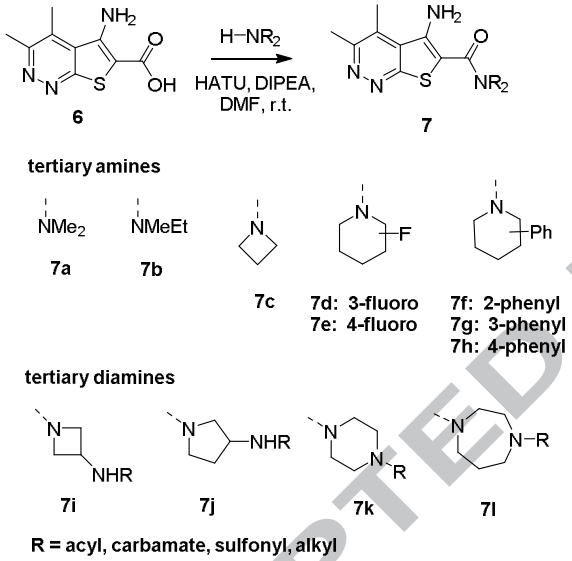
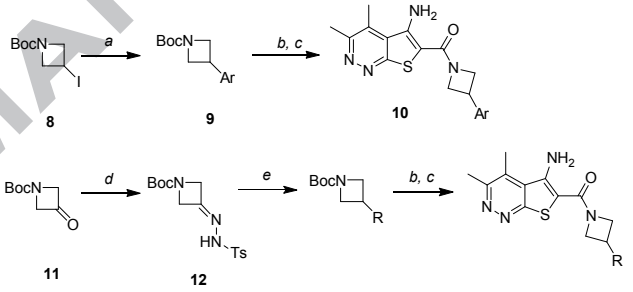


Figure 3. Cyclic tertiary amides generated on thieno-[2,3-*c*]pyridazine core. Majority of examples hM_4 EC_{50} > 2 μM.

molecules in the lattice, which likely contributed to the compound's poor aqueous solubility. We hypothesized that disruption of both the hydrogen bonding and π-stacking would be beneficial. Replacement of the benzyl linker with a saturated cyclic moiety would impart additional sp^3 character and could disrupt the planarity of the compounds which may impede π-stacking. Additionally, removal of the amide N-H could also disrupt the hydrogen bonding network, which may improve the solubility and absorption, as well as possibly mitigate P-gp efflux which had limited CNS exposure in some 5-amino-thieno[2,3-*c*]pyridazine analogs. Unfortunately, in examples across several different amide series, replacement of the amide NH with a methyl group resulted in a dramatic loss of human M_4 (hM_4) potency (data not shown). However, given our success in employing small, cyclic diamine linkers at the amide portion of our compounds,[24] we turned our attention to examining cyclic amines linked through an endocyclic amine to the thieno[2,3-*c*]pyridazine core to furnish a library of tertiary amides.

We initially examined a variety of 4, 5, and 6-membered cyclic amines and diamines to assess the feasibility of this strategy. Despite finding numerous potent and efficacious secondary exocyclic amides representing a variety of ring sizes and substitution patterns, the majority of endocyclic tertiary amides examined were either inactive or only weakly active (Figure 3). Fortunately, in our screen we found that substitution at the 3-position of the azetidine amides provided a substantial boost in potency. In order to synthesize 3-aryl substituted azetidines, we relied on either a Negishi reaction[28] starting from commercially available 1-Boc-3-iodoazetidine or a metal-free reductive cross coupling between a tosyl hydrazone and boronic acid (Scheme 1).[29,30] The operational simplicity (air and moisture stable intermediates amenable to isolation, purification, and storage) and substrate tolerance of the reductive cross coupling allowed us to rapidly and efficiently explore the SAR of this series. In the course of coupling tosyl hydrazone azetidine with boronic acid partners, we sought to investigate the ability of the azetidine tosyl hydrazones to engage BF_3K salts (Table 1). In a small screen of BF_3K salts, we found that four out of five examined provided the desired coupled product. While the simple allyl trifluoroborate salt failed to couple, substituted vinyl, aryl, and heteroatom containing BF_3K salts afforded the desired product. Although the yields were modest, we were able to obtain them with no modification or optimization of the original protocol,

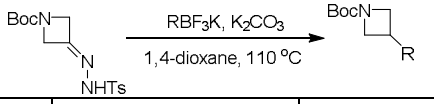
Scheme 1. Synthesis of tertiary azetidine analogs

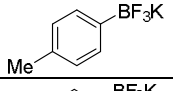
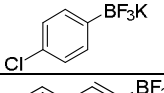
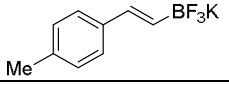


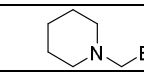
Conditions: a) Zn, TMSCl, 1,2-dibromoethane, ArX, $Pd(PPh_3)_4$ b) TFA, DCM, r.t c) 5-amino-3,4-dimethylthieno[2,3-*c*]pyridazine-6-carboxylic acid, HATU, DIPEA, DMF, r.t d) $TsNHNH_2$, 1,4-dioxane, 80 °C e) $RB(OH)_2$, K_2CO_3, 1,4-dioxane, 110 °C.

demonstrating the general utility of this reaction. To the best of our knowledge, this represents the first reported example whereby a BF_3K salt has been used to engage the tosyl hydrazone in a reductive cross coupling, allowing for the preparation of analogs for which the boronic acid may not be stable.

Table 1. Reductive cross coupling with trifluoroboronate salts

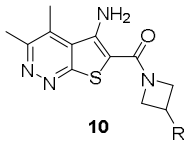


Entry	RBF_3K	Yield (%)
1		NDP
2		31
3		29
4		25

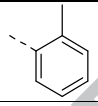
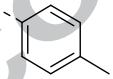
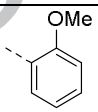
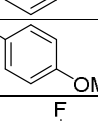
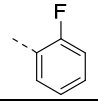
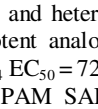
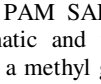
5		28

The 3-position of the azetidine tolerated aryl (**10a**, **10d**-**10l**), aliphatic (**10b**), and heteraryl (**10c**) substitution (Table 2). One of the most potent analogs prepared was unsubstituted phenyl group **10a** (hM_4 EC_{50} = 72 nM, ACh_{Max} = 65±3%). As frequently observed with PAM SAR, substitution on the aryl ring could result in dramatic and unpredictable differences in potency. Introduction of a methyl group at either the 2-, 3-, or 4-position (**10d**-**10f**) resulted in a fairly uniform loss of potency (4-7-fold); however, a methoxy substituent at the 4-postion (**10i**) resulted in a modest ~3.5-fold loss in potency while the meta-substituted analog gave >100-fold loss in potency (**10h**). As we have previously observed in M_4 PAM SAR, fluoro substituents were well tolerated, especially the 3- and 4-position (**10k** and **10l**). Replacement of the fluorine with other halogens resulted in a complete loss of hM_4 activity (**10m**, **10n**). Saturation of the phenyl ring (**10b**) led to a modest decrease in potency (hM_4 EC_{50} = 178 nM, ACh_{Max} = 54 ± 12), as did heteroaryl substitution (**10c**) (hM_4 EC_{50} = 200 nM, ACh_{Max} = 78 ± 7).

Table 2. Analogs derived from 3-substituted azetidines



10

Cmpd	R	hM_4 EC_{50} (nM)[a] [% ACh Max ±SEM]	hM_4 pEC_{50} (±SEM)
10a		72 [65±3]	7.14±0.18
10b		178 [54±12]	6.75±0.16
10c		200 [78±7]	6.70+0.13
10d		457 [72±7]	6.34±0.04
10e		851 [83±6]	6.07±0.06
10f		1096 [82±4]	5.96±0.11
10g		2290 [54±7]	5.64±0.15
10h		>10,000 [N.D.]	N.D.
10i		331 [94±7]	6.48±0.12
10j		407 [75±8]	6.39±0.03
10k		57 [83±9]	7.24±0.20
10l		151 [80±7]	6.82±0.13
10m		>10,000 [N.D.]	N.D.
10n		>10,000 [N.D.]	N.D.

[a]Calcium mobilization assays with $hM_{4/Gqi5}$-CHO cells performed in the presence of an EC_{20} fixed concentration of acetylcholine; values represent means from three (*n*=3) independent experiments performed in triplicate.[31]

In order to further evaluate this new amide series, **10a**, **10b**, **10k** and **10l** were evaluated for mAChR subtype selectivity and *in vitro* DMPK assays. Compound **10a** was subjected to full subtype selectivity screening and found to possess excellent selectivity for hM_4 over other subtypes ($hM_{1\text{-}3,5}$ EC_{50} > 10 μM). Compound **10k** and **10l** were counter screened against hM_2, and **10l** was found to be inactive. While **10k** was active at hM_2 (EC_{50} = 2.10 μM), it only weakly potentiated the ACh response (ACh_{Max} = 38%). In contrast to a number of other series with the thieno[2,3-*c*]pyridazine core, **10a** was ~2-fold more potent at hM_4 compared to rat M_4 (rM_4; data not shown). When assessed for CNS penetration, all four compounds were found to have high brain distribution in rat (**10a** K_p = 2.6, $K_{p,uu}$ = 2.1; **10b** K_p = 8.46, $K_{p,uu}$ = 0.71; **10k** K_p = 2.1, $K_{p,uu}$ = 0.92, **10l** K_p = 2.3, $K_{p,uu}$ = 1.3). As a class, the tertiary azetidines appear to address one of the major challenges associated with the previously reported secondary azetidine series, which suffered from P-gp efflux and low CNS exposure, requiring an extensive SAR campaign to address these parameters. Unfortunately, all four compounds exhibited high predicted clearance in both rat and human (**10a** CL_{hep} = 64 (r), 20 (h) mL/min/kg; **10b** CL_{hep} = 65 (r), 20 (h) mL/min/kg; **10k** CL_{hep} = 56 (r), 19 (h) mL/min/kg; **10l** CL_{hep} = 56 (r), 19 (h) mL/min/kg) based on hepatic microsomal CL_{int} data using the well-stirred model (binding terms excluded).

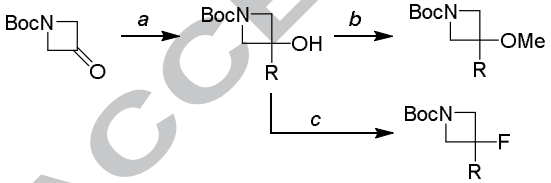
To address the putative high clearance in this series of compounds, metabolic softspot analysis was performed using rat hepatic microsomal incubations, which identified NADPH-dependent hydroxylation of the azetidine ring as the principle pathway of metabolism (data not shown). Based on this finding, we sought to improve the metabolic stability by blocking the benzylic site on the azetidine ring. Synthesis of these compounds was accomplished by addition of the appropriate Grignard or organolithium reagent into 1-Boc azetidinone, followed by alkylation with MeI or fluorination of the resultant alcohol with DAST (**Scheme 2**). The Boc group was then cleaved with TFA and the azetidine connected to the core via a HATU coupling, analogous to the route described in **Scheme 1**. In general, the introduction of geminal substituents at the 3-position resulted in a loss of potency, with -OH (**11a**) and -OMe (**11b**) resulting in a ~10-fold and ~6-fold loss of potency, respectively. Both of these substitutions also resulted in a marked decline in the compounds' efficacy in potentiation of the ACh response. Replacement of the benzylic hydrogen with fluorine was better tolerated (**11c-f**), especially with regards to ACh_{Max}; however, the EC_{50}s obtained from these compounds were still ~5-fold weaker than the best analogs bearing a hydrogen at the benzylic position. Additionally, all compounds tested in which the benzyl site was blocked were still found to possess predicted rat and human clearance near or at hepatic blood flow rates. Ultimately,

metabolite identification efforts found that the primary pathway for metabolism was oxidation on the azetidine ring irrespective of the fluorine atom at the benzylic site.

With these data in hand, additional strategies were also examined for minimizing the clearance of the 3-aryl azetidine analogs. We examined introduction of a methyl group at the 2-position of the azetidine, which we hoped may reduce oxidation on the azetidine ring. Starting from commercially available tert-butyl 2-methyl-3-oxoazetidine-1-carboxylate and following the synthetic route described in **Scheme 1**, analogs **12a** and **12b** were prepared. Both compounds suffered from weaker potency (EC_{50} = 1510, 1455 nM, respectively), but also an impaired ability to reach full potentiation of the hM_4 receptor (ACh_{Max} = ~45%). Introduction of a spiro-pyrroldine ring to the azetidine amide afforded analogs **13a** and **13b**. This substitution also led to a decrease in hM_4 potency relative to **10a** (EC_{50} = 324 and 500 nM, respectively). Furthermore, both analogs still suffered from high predicted clearance (CL_{hep} = 49 and 56 mL/min/kg, respectively). After being unable to identify substitutions to either the 3-position substituent or the azetidine ring that would allow us to address the high clearance associated with this class of compounds yet still maintain reasonable potency, **10a** was evaluated in a rat amphetamine-induced hyperlocomotion (AHL) model of antipsychotic efficacy. Despite its high predicted clearance, at a 30 mg/kg oral dose **10a** was still able to achieve 32% reversal of AHL.

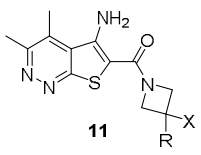
In summary, we have described the synthesis and biological evaluation of a novel series of tertiary azetidine amides within the 5-amino-thieno[2,3-*c*]pyridazine class of M_4 PAMs. In the synthesis of these compounds, we were able to utilize a reductive cross coupling between azetidinyl tosyl hydrazones and boronic acids, and found that this methodology could be extended to trifluoroboronate salts as well. This new series of azetidine amides was promising because its members generally provided excellent CNS exposure and possessed a preference for activity at hM_4 over rM_4, two trends that had been challenging to overcome in other series studied. Despite suffering from high predicted clearance, which we were unable to alleviate with substitutions at either the 3-position substituent or the azetidine ring itself, compound **10a** was able to achieve 32% reversal in a rodent AHL model. Future studies aimed at achieving potent molecules with a suitable disposition are currently underway and will be reported in due course.

Scheme 2. Synthesis of azetidines with benzylic hydrogen removed

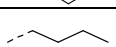


Conditions: a) RMgBr or RLi, THF, 0 °C b) MeI, K_2CO_3, DMF c) DAST, DCE

Table 3. Analogs of 3-substituted azetidines with benzylic position blocked

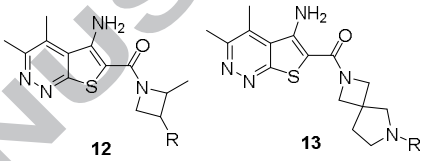


11

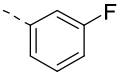
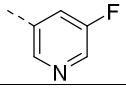
Cmpd	R	X	hM_4 EC_{50} (nM)[a] [% ACh Max ±SEM]	hM_4 pEC_{50} (±SEM)
11a		OH	1047 [30±4]	5.98±0.04
11b		OMe	417 [36±2]	6.38±0.08
11c		F	447 [67±3]	6.35±0.18
11d		F	2240 [78±1]	5.65±0.23
11e		F	316 [85±12]	6.50±0.23
11f	-Me	F	363 [98±12]	6.44±0.07

[a]Calcium mobilization assays with $hM_{4/Gqi5}$-CHO cells performed in the presence of an EC_{20} fixed concentration of acetylcholine; values represent means from three (*n*=3) independent experiments performed in triplicate.

Table 4. Azetidine analogs to address high clearance



12 **13**

Cmpd	R	hM_4 EC_{50} (nM)[a] [% ACh Max ±SEM]	hM_4 pEC_{50} (±SEM)
12a		1510 [45±3]	5.82±0.24
12b		1455 [45±2]	5.84±0.15
13a		324 [57±0]	6.49±0.07
13b		500 [53±1]	6.39±0.22

[a]Calcium mobilization assays with $hM_{4/Gqi5}$-CHO cells performed in the presence of an EC_{20} fixed concentration of acetylcholine; values represent means from three (*n*=3) independent experiments performed in triplicate.

Acknowledgments

The authors would like to thank NIH (U01MH087965, Vanderbilt, NCDDG). We also thank William K. Warren, Jr. and the William K. Warren Foundation who funded the William K Warren, Jr. Chair in Medicine (to C.W.L.).

References

1. Bodick, N. C.; Offen, W. W.; Levey, A. I.; Cutler, N. R.; Gauthier S. G.; Satlin, A.; Shannon, H. E.; Tollefson, G. D.; Rasmussen, K.; Bymaster, F. P.; Hurley, D. J.; Potter, W. Z.; Paul, S. M. *Arch. Neurol*. **1997**, *54*, 465-473.
2. Shekhar, A.; Potter, W. Z.; Lightfoot, J.; Lienemann, J.; Dube, S.; Mallinckrodt, C.; Bymaster, F. P.; McKiinzie, D. L.; Felder, C. C. *Am. J. Psychiatry* **2008**, *165*, 1033-1039.
3. Lieberman, J. A.; Javitch, J. A.; Moore, H. *Am. J. Psychiatry* **2008**, *165*, 931-936.
4. Chan, W.Y.; McKinize, D.L.; Bose, S.; Mitchell, S.N.; Witkins, J.M.; Thompson, R.C.; Christopoulos, A.; Birdsall, N.J.;

Bymaster, F.P.; Felder, C.C. *Proc. Natl. Acad. Sci. USA* **2008**, *105*, 10978-10983.
5. Leach, K.; Loiancono, R.E.; Felder, C.C.; McKinize, D.L.; Mogg, A.; Shaw, D.B.; Sexton, P.M.; Christopoulos, A. *Neuropsychopharmacology* **2010**, *35*, 855-869.
6. Brady, A.; Jones, C.K.; Bridges, T.M.; Kennedy, P.J.; Thompson, A.D.; Breininger, M.L.; Gentry, P.R.; Yin, H.; Jadhav, S.B.; Shirey, J.; Conn, P.J.; Lindsley, C.W. *J. Pharm. & Exp. Ther.* **2008**, *327*, 941-953.
7. Byun, N.E.; Grannan, M.; Bubser, M.; Barry, R.L.; Thompson, A.; Rosanelli, J.; Gowrishnakar, R.; Kelm, N.D.; Damon, S.; Bridges, T.M.; Melancon, B.J.; Tarr, J.C.; Brogan, J.T.; Avison, M.J.; Deutch, A.Y.; Wess, J.; Wood, M.R.; Lindsley, C.W.; Gore, J.C.; Conn, P.J.; Jones C.K. *Neuropsychopharmacology* **2014**, *39*, 1578-1593.
8. Farrell, M.; Roth, B.L. *Neuropsychopharmacology* **2010**, *35*, 851-852.
9. Jones, C.K.; Byun, N.; Bubser, M. *Neuropsychopharmacology* **2012**, *37*, 16-42.
10. Shirey, J.K.; Xiang, Z.; Orton, D.; Brady, A.E.; Johnson, K.A.; Williams, R.; Ayala, J.E.; Rodriguez, A.L.; Wess, J.; Weaver, D.; Niswender, C.M.; Conn, P.J. *Nat. Chem. Bio.* **2008**, *4*, 42-50.
11. Le, U.; Melancon, B.J.; Bridges, T.M.; Utley, T.J.; Lamsal, A.; Vinson, P.N.; Sheffler, D.J.; Jones, C.K.; Morrison, R.; Wood, M.R.; Daniels, J.S.; Conn, P.J.; Niswender, C.M.; Lindsley, C.W. Hopkins, C.R. *Bioorg. Med. Chem. Lett*. **2013**, *23*, 346-350.
12. Kennedy, J.P.; Bridges, T.M.; Gentry, P.R.; Brogan, J.T.; Brady, A.E.; Shirey, J.K.; Jones, C.K.; Conn, P.J.; Lindsley, C.W. *ChemMedChem.* **2009**, *4*, 1600-1607.
13. Salovich, J.M.; Sheffler, D.J.; Vinson, P.N.; Lamsal, A.; Utley, T.J.; Blobaum, A.L.; Bridges, T.M.; Le, U.; Jones, C.K.; Wood, M.R.; Daniels, J.S.; Conn, P.J.; Niswender, C.M.; Lindsley, C.W.; Hopkins, C.R. *Bioorg. Med. Chem. Lett.* **2012**, *22*, 5084-5088.
14. Bubser, M.; Bridges, T.M.; Thorbeck, D.D.; Gould. R.W.; Grannan, M.; Noetzel, M.J.; Niswender, C.M.; Daniels, J.S.; Melancon, B.J.; Tarr, J.C.; Wess, J.; Duggan, M.E.; Brandon, N.J.; Dunlop, J.; Wood, M.W.; Wood, M.R.; Lindsley, C.W.; Conn, P.J.; Jones C.K. *ACS Chem. Neurosci.* **2014**, *5*, 920-942.
15. Smith, E.; Chase, P.; Niswender, C.M.; Conn, P.J.; Lindsley, C.W.; Madoux, F.; Acosta, M.; Scampavia, L.; Spicer, T.; Hodder, P. *J. Biomol. Screening* **2015**, *20*, 858-868.
16. Wood, M.R.; Noetzel, M.J.; Tarr, J.C.; Rodriguez, A.L.; Lamsal, A.; Chang, S.; Foster, J.J.; Smith, E.; Hodder, P.S.; Engers, D.W.; Niswender, C.M.; Brandon, N.J.; Wood, M.W.; Duggan, M.E.; Conn, P.J.; Bridges, T.M.; Lindsley, C.W. *Bioorg. Med. Chem. Lett.* **2016**, *26*, 4282-4286.
17. Wood, M.R.; Noetzel, M.J.; Engers, J.L.; Bollinger, K.A.; Melancon, B.J.; Tarr, J.C.; Han, C.; West, M.; Gregro, A.R.; Lamsal, A.; Chang, S.; Ajmera, S.; Smith, E.; Chase, P.; Hodder, P.S.; Bubser, M.; Jones, C.K.; Hopkins, C.R.; Emmitte, K.A.; Niswender, C.M.; Wood, M.W.; Duggan, M.E.; Conn, P.J..; Bridges, T.M.; Lindsley, C.W. *Bioorg. Med. Chem. Lett.* **2016**, *26*, 3029-3033.
18. Szabo, M.; Huynh, T.; Valant, C.; Lane, J.R.; Sexton, P.M.; Christopoulos, A.; Capuano, B. *MedChemComm.* **2015**, *6*, 1998-2003.
19. Huynh, T.; Valant, C.; Crosby, I.T.; Sexton, P.M.; Christopoulos, A.; Capuano, B. *ACS Chem. Neurosci.* **2015**, *6*, 1592-1599.
20. Croy, C.H.; Schober, D.A.; Xiao, H.; Quets, A.; Christopoulos, A.; Felder, C.C. *Mol. Pharmacol.* **2014**, *86*, 106-115.
21. Huynh, T.; Valant, C.; Crosby, I.T.; Sexton, P.M.; Christopoulos, A.; Capuano, B. *J. Med. Chem.* **2013**, *56*, 1592-1599.
22. Wood, M. R.; Noetzel, M. J.; Poslusey, M. S.; Melancon, B. J.; Tarr, J. C.; Lamsal, A.; Chang, S.; Luscombe, V. B.; Weiner, R. L.; Cho, H. P.; Bubser, M.; Jones, C. K.; Niswender, C. M.; Wood, M. W.; Engers, D. W.; Brandon, N. J.; Duggan, M. E.; Conn, P. J.; Bridges, T. M; Lindsley, C. W. *Bioor. Med. Chem. Lett*. **2017**, *27*, 171-175.
23. Wood, M. R.; Noetzel, J; Melancon, B. J.; Poslusney, M. S.; Nance, K. D.; Hurtade, M. A.; Luscombe, V. B.; Weiner, R. L.; Rodriguez, A L.; Lamsal, A.; Chang, S.; Bubser, M.; Blobaum, A. L.; Engers, D. W.; Niswender, C. M.; Jones, C. K.; Brandon, N. J.; Wood, M. W.; Duggan, M. E.; Conn, P. J.; Bridges, T. M.; Lindsley, C. W. *ACS Med. Chem. Lett*. **2017**, *8*, 233-238.
24. Tarr, J. C.; Wood, M. R.; Noetzel, M. R.; Bertron, J.; Weiner, R. L.; Rodriguez, A. L.; Lamsal, A.; Byers, F. W.; Chang, S.; Cho, H. P.; Jones, C. K.; Niswender, C. M.; Wood, M. W.; Brandon, N. J.; Duggan, M. E.; Conn, P. J.; Bridges, T. M.; Lindsley, C. W. *Bioorg. Med. Chem. Lett*. **2017,** In press.
25. Melancon, B. J; Wood, M. R.; Noetzel, M. J; Nance, K. D.; Engelberg, E. M.; Han, C.; Lamsal, A.; Chang, S.; Cho, H. P.; Byers, F. W.; Bubser, M.; Jones, C. K.; Niswender, C. M.; Wood, M. W.; Engers, D. W.; Wu, D.; Brandon, N. J.; Duggan, M. E.; Conn, P. J.; Bridges, T. M.; Lindsley, C. W. *Bioorg. Med. Chem. Lett*. **2017**, *11*, 2296-2301.
26. Pancani, T.; Foster, D.J.; Bichell, T.; Bradley, E.; Bridges, T.M.; Klar, R.; Daniels, J.S.; Jones, C.K.; Bowman, A.B.; Lindsley, C.W.; Xiang, Z.; Conn, P.J. *Proc. Natl. Acad. Sci. USA,* **2015**, *112*, 14078-14083.
27. Shen, W.; Plotkin, J.L.; Francardo, V.; Ko, W.K.D.; Xie, Z.; Li, Q.; Fieblinger, T.; Wess, J.; Neubig, R.R.; Lindsley, C.W.; Conn, P.J.; Greengrad, P.; Bezard, E.; Cenci, M.A.; Surmeier, D.J. *Neuron* **2015**, *88*, 762-773.
28. Haas, D.; Hammann, J. M.; Greiner, R.; Knochel, P. *ACS Catal*. **2016**, *6*, 1540-1552.
29. Barluenga, J.; Tomas-Gamasa, M.; Aznar, F.; Valdes C. *Nature Chemistry* **2009**, *1*, 494-499.
30. Allwood, D. M.; Blakemore, D. C.; Brown, A. D.; Ley, S. V. *J. Org. Chem.* **2014**, *79*, 328-338.
31. VU0467485 (Ref. 23) was included as a positive control during each experiment.