NIH Public Access
Author Manuscript
Bioorg Med Chem Lett. Author manuscript; available in PMC 2014 November 01.

Published in final edited form as:
Bioorg Med Chem Lett. 2013 November 1; 23(21): . doi:10.1016/j.bmcl.2013.08.057.

Synthesis and assessment of catechol diether compounds as inhibitors of trypanosomal phosphodiesterase B1 (TbrPDEB1)

Jennifer L. Woodring[a], **Nicholas D. Bland**[b], **Stefan O. Ochiana**[a], **Robert K. Campbell**[b], and **Michael P. Pollastri**[a]

[a]Northeastern University Department of Chemistry and Chemical Biology, 417 Egan Research Center, 360 Huntington Avenue, Boston, MA 02115, USA

[b]Marine Biological Laboratory, Josephine Bay Paul Center for Comparative Molecular Biology and Evolution, 7 MBL Street, Woods Hole, MA 02543, USA

Abstract

Human African trypanosomiasis (HAT) is a parasitic neglected tropical disease that affects 10,000 patients each year. Current treatments are sub-optimal, and the disease is fatal if not treated. Herein, we report our continuing efforts to repurpose the human phosphodiesterase 4 (hPDE4) inhibitor piclamilast to target trypanosomal phosphodiesterase TbrPDEB1. We prepared a range of substituted heterocyclic replacements for the 4-amino-3,5-dichloro-pyridine head group of piclamilast, and found that these compounds exhibited weak inhibitory activity of TbrPDEB1.

Human African trypanosomiasis (HAT) is a neglected tropical disease caused by the parasites *Trypanosoma brucei gambiense* and *T. b. rhodesiense*. Together, over 60 million people in 36 countries in sub-Saharan Africa are at risk, with approximately 10,000 infections annually.[1] HAT is fatal unless treated and the four drugs approved for this indication: pentamidine, suramin, eflornithine, and melarsoprol, are inadequate for a variety of reasons, including cost, toxicity, and lack of oral bioavailability. For instance, melarsoprol is especially toxic as it induces reactive encepalophathy in 5–10% of patients, killing approximately half of them.[2] As such, new medicines are desperately needed but pharmaceutical companies tend to deprioritize diseases such as HAT due to an inability to recover research costs from the extremely poor who are the most affected by the disease.

In order to speed up the drug discovery process, a drug repurposing approach[3] has been taken against two *T. brucei* phosphodiesterases (PDEs), TbrPDEB1 and TbrPDEB2.[4–6] Simultaneous RNAi knockdown of both is fatal,[7] suggesting that small molecule inhibitors of these enzymes could be useful interventions.[4, 8] Humans have 11 PDEs that have been well explored, producing numerous clinical drug candidates.[9] The catalytic domains of human PDEs are 30–35% homologous to those of the parasite enzymes TbrPDEB1 and TbrPDEB2. Recent crystallographic evidence confirms that there are key regions of the trypanosomal protein that may allow for selective inhibition over human PDEs.[10] We previously reported that human PDE4 inhibitor piclamilast (**1**) represents a promising lead

© 2013 Elsevier Ltd. All rights reserved.

Supporting information. Supporting information includes synthetic preparations, compound characterization, biological assay conditions, and a recapitulation of the tables from the manuscript that include compound registry numbers. All data is freely available on the Collaborative Drug Discovery database (www.collaborativedrug.com).

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

NIH-PA Author Manuscript
NIH-PA Author Manuscript
NIH-PA Author Manuscript

NIH-PA Author Manuscript

NIH-PA Author Manuscript

NIH-PA Author Manuscript

series for optimization towards selective TbrPDEB inhibitors,[4] and others have shown analogous catechol-derived inhibitors to have high potency against the trypanosomal enzyme.[8, 11] In this report, we describe our efforts to explore replacements for the 2,6-dichloro-4-pyridylamide headgroup of **1** by assessing their potency against TbrPDEB1.

Our rationale for focusing first on the headgroup region of **1** is shown in Figure 1. The optimization of phthalazinones as human PDE4 (hPDE4) inhibitors has been described, leading to the discovery of **2** with an IC_{50} of 0.6 nM against hPDE4.[12] This compound was later discovered in a screening campaign to be a potent inhibitor of TbrPDEB1 (3.98 nM),[8] and we found that this compound comparably inhibits TbrPDEB2 (6 nM). A related set of compounds (**3** and **4**, Figure 1) has recently been disclosed as TbrPDEB1 inhibitors.[11] There is obvious structural similarity between compounds **1–4**, though **1** inhibits TbrPDEB1 with an IC_{50}=4.7 μM, whereas **2–4** inhibit TbrPDEB1 with sub-micromolar potency. We prepared **5**,[12–15] an analog of **1** and **2**, to more directly compare the effect of the headgroup region of the inhibitor, and confirmed its activity against TbrPDEB1 and B2 (IC_{50}=278 and 544 nM, respectively). Thus, in observing the 15-fold increase in potency with **5** versus **1**, we hypothesized that the headgroup region must be a major driver of potency against TbrPDEB1. This became our rationale in future compound design and the resulting structure-activity-relationship (SAR) exploration.

We prepared putative heterocyclic aromatic headgroup replacements for the phthalizinone moiety as shown in Scheme 1. Electrophilic bromination of guaiacol by a previously published method afforded 5-bromo-2-methoxyphenol, intermediate **6**.[16] This intermediate reacted with cyclopentanol under Mitsunobu conditions to produce the dialkoxy catechol intermediate **7a**. A small library of boronic acids was used in Suzuki reactions with intermediate **7a** to yield analogs **8a–c**, which were found to be weak inhibitors of TbrPDEB1 (Table 1).

We also prepared a series of pyrimidine and pyridine-derived inhibitors, as shown in Scheme 2. Compound **8a** could be converted to the chloropyridine intermediate **9** by a sequence of N-oxide formation and chlorination with $POCl_3$. The analogous pyrimidine intermediate **10** was made via borylation of intermediate **7a**, followed by a Suzuki reaction with 2,4-dichloropyrimidine. Chloride displacement with small aliphatic and benzylic amines was achieved to provide the pyrimidine and pyridine-derived analogs **11** and **12** (Table 2).

Figure 1 shows a related TbrPDEB1 inhibitor chemotype (**4**) that showed equal enzymatic potency (though improved cellular potency) where the cyclopentyl catechol ether was replaced with a benzyl ether.[11] In light of this, we opted to also make benzyl-substituted analogs of the compounds in Scheme 2, which was achieved by the route shown in Scheme 3. The brominated intermediate **7b** was converted to the 2-chloropyridyl intermediate **14a** under Suzuki conditions in modest yields. We prepared the pyrimidine template **16** via the boronate; we elected to utilize palladium catalysis for the preparation of **13**, as the lithium-halogen exchange route applied in Scheme 2 led to undesirable side reactions. The crude boronate **13** was reacted with 2,4-dichloropyrimidine under Suzuki conditions to obtain intermediate **14b**. As before, the pyrimidine and pyridine libraries were produced from halide displacement with various amines. We note that the dimethyl-substituted analogs (**11a, 12a, 15a, 16a**) were obtained as side-products when performing the displacement reactions in DMF, the chloride having reacted with dimethyl amine generated by thermal decomposition of the solvent.

All analogs generated were tested for inhibition of TbrPDEB1 at a concentration of 10 μM (Table 2). Regrettably, we observed that all analogs showed weak (< 50%) inhibition of

NIH-PA Author Manuscript

NIH-PA Author Manuscript

NIH-PA Author Manuscript

TbrPDEB1. We conclude that, though the headgroup of **5** is responsible for significant improvement of potency over **1**, the substituted aromatic headgroup replacements we prepared do not appropriately occupy the space that the headgroup of **5** does. We hypothesize that larger, nonplanar headgroups may be needed.

Figure 2 shows an overlay of **5** (green) against the cyclopentyl substituted analogs in **8c** (black) and **11a** (purple). This figure was generated by creating a conformer database of the analogs (using OMEGA version 2.4.3 OpenEye Scientific Software, Santa Fe, NM.),[17] and generating optimal shape and electrostatic overlays against a minimized conformation of **5** (using ROCS version 3.0.0 and EON version 2.1.0). It is clear from this analysis that there is poor overlap between these new analogs and the fused ring of the N-alkylphthalizinone core, which most likely attributes to the observed differences in potency. Further, though the pyridyl, pyrimidinyl, or quinolinyl nitrogen is positioned close to the carbonyl of **5**, these lone pairs may not reach far enough into the metal binding pocket to create good binding interactions suggested by previous crystallography results.[10, 18–21] With this in mind, our optimization efforts continue in this class of compounds, with an eye towards a better recapitulation of the headgroup-metal interactions.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

We thank Dr. Geert-Jan Sterk (MercaChem) for helpful discussions. We acknowledge funding from the National Institutes of Health (R01AI082577), and are grateful for a free academic license from OpenEye Scientific Software for ROCS and EON.

References

1. Savioli, L.; Daumerle, D. World Health Organization. 2013. http://apps.who.int/iris/bitstream/10665/77950/1/9789241564540_eng.pdf
2. Chappuis F, Udayraj N, Stietenroth K, Meussen A, Bovier PA. Clin Infect Dis. 2005; 41:748. [PubMed: 16080099]
3. Pollastri MP, Campbell RK. Future Med Chem. 2011; 3:1307. [PubMed: 21859304]
4. Bland ND, Wang C, Tallman C, Gustafson AE, Wang Z, Ashton TD, Ochiana SO, McAllister G, Cotter K, Fang AP, Gechijian L, Garceau N, Gangurde R, Ortenberg R, Ondrechen MJ, Campbell RK, Pollastri MP. J Med Chem. 2011; 54:8188. [PubMed: 22023548]
5. Ochiana SO, Gustafson A, Bland ND, Wang C, Russo MJ, Campbell RK, Pollastri MP. Bioorg Med Chem Lett. 2012; 22:2582. [PubMed: 22377518]
6. Wang C, Ashton TD, Gustafson A, Bland ND, Ochiana SO, Campbell RK, Pollastri MP. Bioorg Med Chem Lett. 2012; 22:2579. [PubMed: 22370268]
7. Oberholzer M, Marti G, Baresic M, Kunz S, Hemphill A, Seebeck T. FASEB J. 2007; 21:720. [PubMed: 17167070]
8. de Koning HP, Gould MK, Sterk GJ, Tenor H, Kunz S, Luginbuehl E, Seebeck T. J Infect Dis. 2012; 206:229. [PubMed: 22291195]
9. Bender AT, Beavo JA. Pharmacol Rev. 2006; 58:488. [PubMed: 16968949]
10. Jansen C, Wang H, Kooistra AJ, de Graaf C, Orrling KM, Tenor H, Seebeck T, Bailey D, de Esch IJ, Ke H, Leurs R. J Med Chem. 2013; 56:2087. [PubMed: 23409553]
11. Orrling KM, Jansen C, Vu XL, Balmer V, Bregy P, Shanmugham A, England P, Bailey D, Cos P, Maes L, Adams E, van den Bogaart E, Chatelain E, Ioset JR, van de Stolpe A, Zorg S, Veerman J, Seebeck T, Sterk GJ, de Esch IJ, Leurs R. J Med Chem. 2012; 55:8745. [PubMed: 22963052]
12. Van der Mey M, Hatzelmann A, Van Klink GP, Van der Laan IJ, Sterk GJ, Thibaut U, Ulrich WR, Timmerman H. J Med Chem. 2001; 44:2523. [PubMed: 11472206]

NIH-PA Author Manuscript

NIH-PA Author Manuscript

NIH-PA Author Manuscript

13. Sterk, G-J. personal communication.
14. Hatzelmann, A.; Boss, H.; Hafner, D.; Beume, R.; Kley, H-P.; Van, DMM.; Van, DLIJ.; Timmerman, H.; Sterk, GJ. WO 9931071A1. 1999. p. 40
15. Van, DMM.; Van, DLIJ.; Timmerman, H.; Hatzelmann, A.; Boss, H.; Hafner, D.; Beume, R.; Kley, H-P.; Sterk, GJ. WO 9831674A1. 1998. p. 59
16. Conde, JJ. WO 2001019785A1. 2001. p. 16
17. Hawkins PCD, Skillman AG, Warren GL, Ellingson BA, Stahl MT. J Chem Inf Model. 2010; 50:572. [PubMed: 20235588]
18. Card GL, England BP, Suzuki Y, Fong D, Powell B, Lee B, Luu C, Tabrizizad M, Gillette S, Ibrahim PN, Artis DR, Bollag G, Milburn MV, Kim SH, Schlessinger J, Zhang KY. Structure. 2004; 12:2233. [PubMed: 15576036]
19. Wang H, Yan Z, Geng J, Kunz S, Seebeck T, Ke H. Mol Microbiol. 2007; 66:1029. [PubMed: 17944832]
20. Xu RX, Hassell AM, Vanderwall D, Lambert MH, Holmes WD, Luther MA, Rocque WJ, Milburn MV, Zhao Y, Ke H, Nolte RT. Science. 2000; 288:1822. [PubMed: 10846163]
21. Huai Q, Liu Y, Francis SH, Corbin JD, Ke H. J Biol Chem. 2004; 279:13095. [PubMed: 14668322]

NIH-PA Author Manuscript

NIH-PA Author Manuscript

NIH-PA Author Manuscript

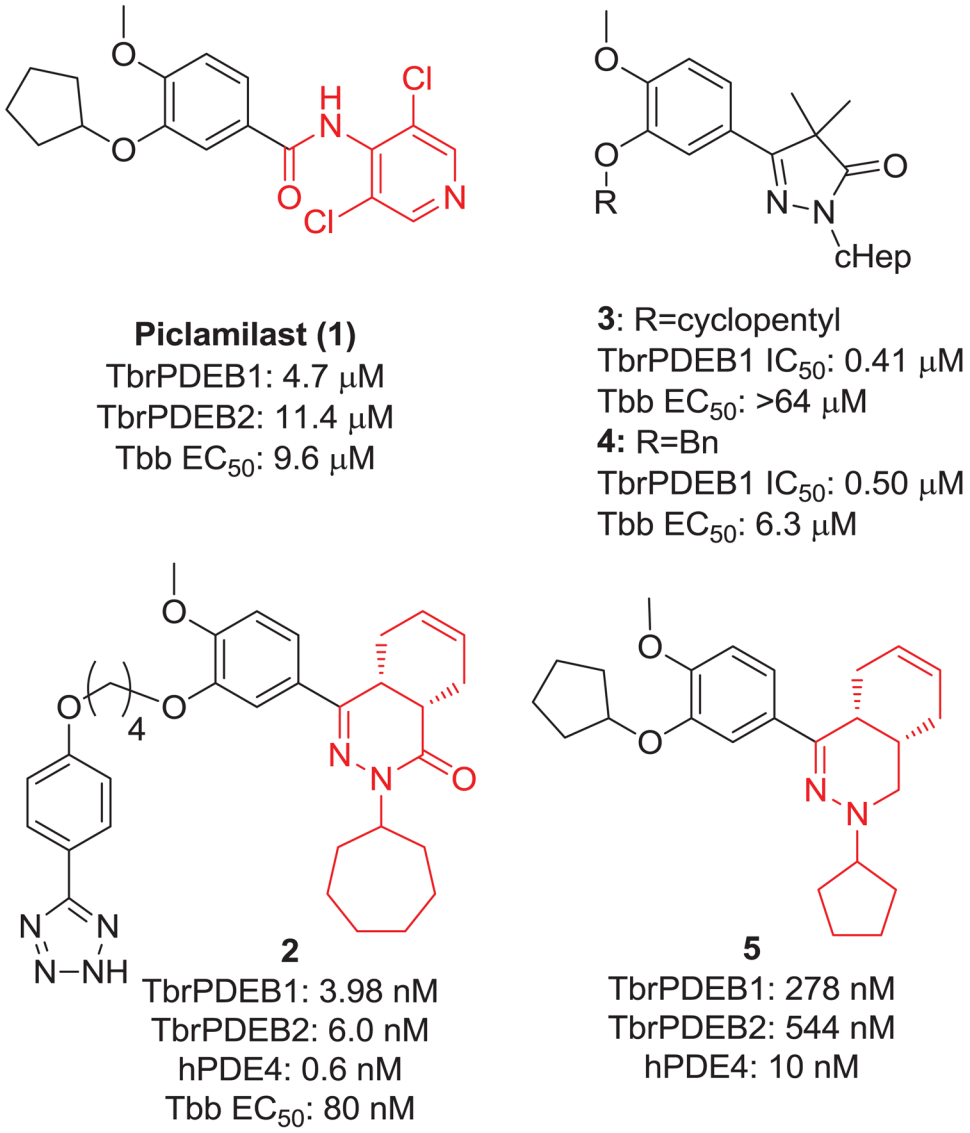


Figure 1.
Headgroup replacement rationale based upon related TbrPDEB1 inhibitor chemotypes.

NIH-PA Author Manuscript

NIH-PA Author Manuscript

NIH-PA Author Manuscript

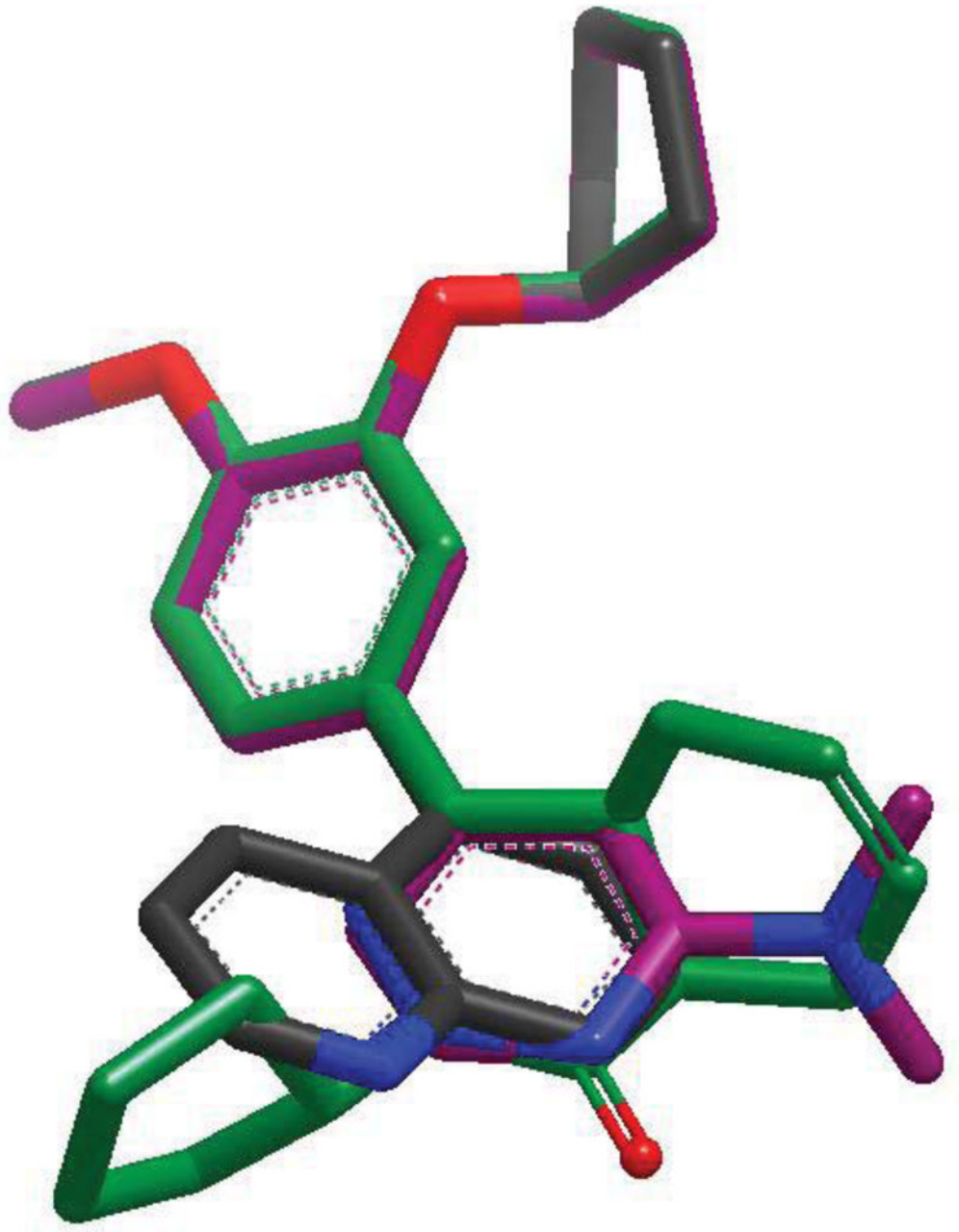
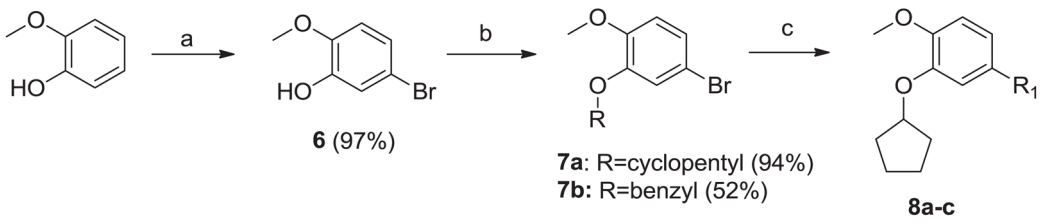


Figure 2.
Overlay of **5**(green) with compounds **8c** (black)and **11a** (purple).



Scheme 1.

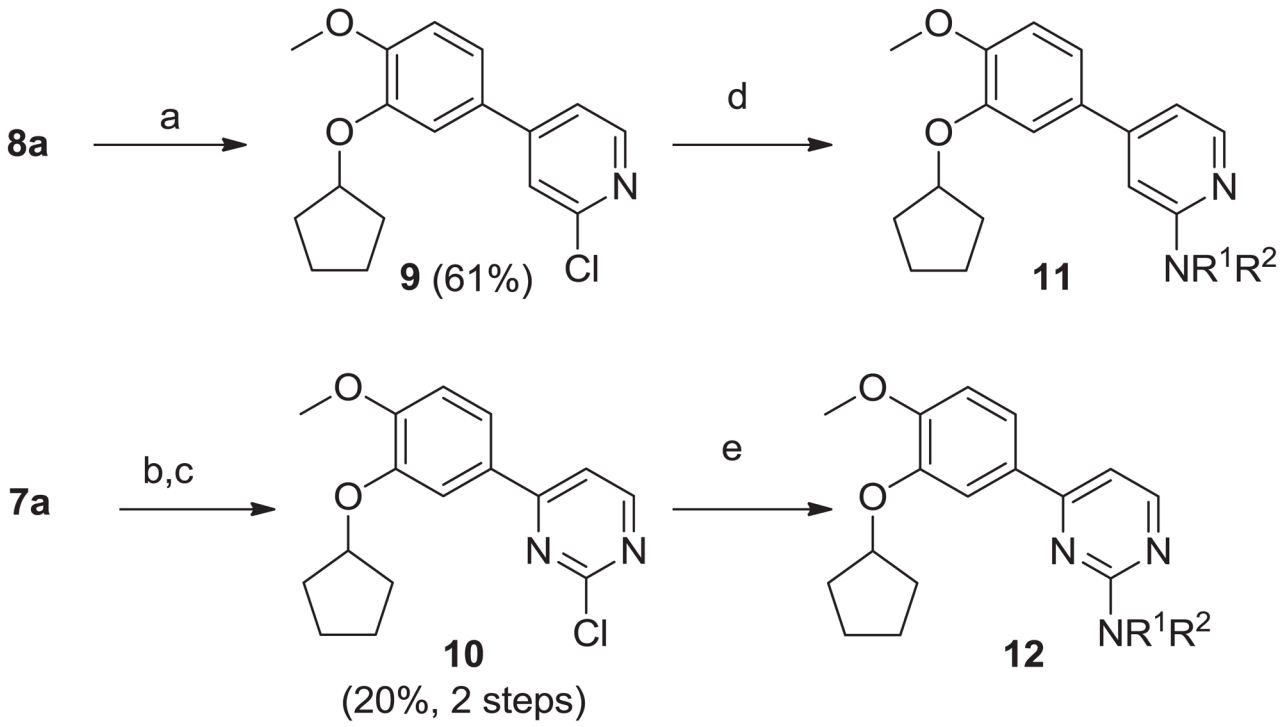
Synthesis of **8a–c**. Reagents and conditions: (a) (i) TFAA, t-BuOK, MeCN, rt, 45 min; (ii) NBS, MeCN, rt, o/n; (b) ROH, PPh_3, DEAD, toluene, rt, 2h; (c) R-$B(OH)_2$, Na_2CO_3, $Pd(PPh_3)_4$, toluene/EtOH/H_2O (4:1:1),105°C, o/n.

NIH-PA Author Manuscript NIH-PA Author Manuscript NIH-PA Author Manuscript

NIH-PA Author Manuscript

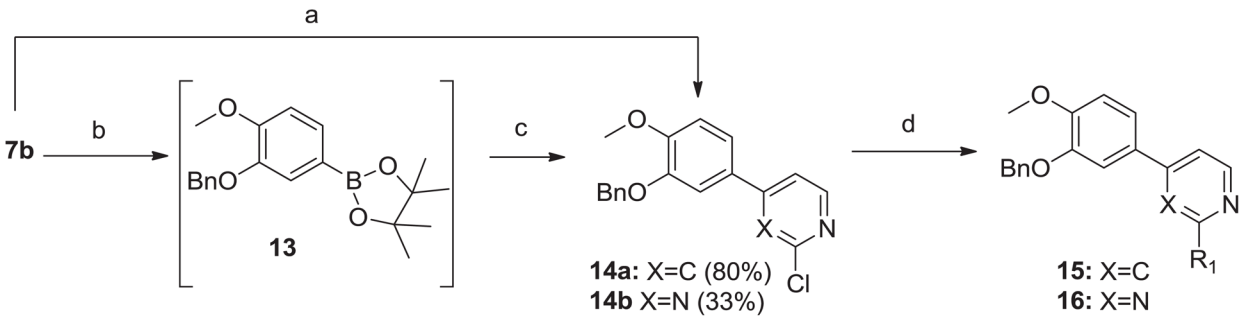
NIH-PA Author Manuscript

NIH-PA Author Manuscript



Scheme 2.

Synthesis of **11** and **12**. Reagents and conditions: (a) (i) m-CPBA, $CHCl_3$, 0°C to RT, o/n; (ii) $POCl_3$, TEA, $CHCl_3$, MW, 100°C, 1 h; (b) n-BuLi, then bis-(pinacolato)diborane, −78°C to rt, o/n; (c) 2,4-dichloropyrimidine, Na_2CO_3, PPh_3, $Pd_2(dba)_3$, tol/EtOH/H_2O (4:1:1), 105°C, o/n; (d) R^1R^2NH, DIEA, NMP or DMF, MW, 250°C, 1 h; (e) R^1R^2NH, DIEA, NMP or DMF, 80°C, o/n.



Scheme 3.

Synthesis of **15** and **16**. Reagents and conditions: (a) 2-chloro-4-pyridylboronic acid, Na_2CO_3, PPh_3, $Pd_2(dba)_3$, tol/EtOH/H_2O (4:1:1), 105°C, o/n; (b) bis-(pinacolato)diborane, KOAc, $PdCl_2(dppf)$, dioxane, MW, 45 min, 145°C; (c) 2,4-dichloropyrimidine, Na_2CO_3, $Pd_2(dba)_3$, tol/EtOH/H_2O (4:1:1), 105°C, o/n; (d) R^1R^2NH, DMF or NMP, heat.

NIH-PA Author Manuscript NIH-PA Author Manuscript NIH-PA Author Manuscript

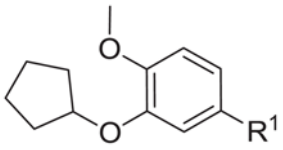
NIH-PA Author Manuscript

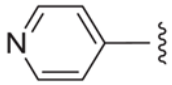
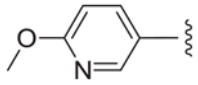
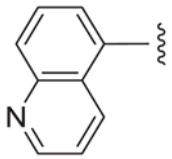
NIH-PA Author Manuscript

NIH-PA Author Manuscript

Table 1

Aryl analogs tested against TbrPDEB1



Compound	R^1	TbrPDEB1 (% inh)[a]
8a		18.7 ± 10.7[b]
8b		7.6 ± 2.6
8c		69.1 ± 13.9

[a] Data shown are average of 3 replicate independent experiments.

[b] Replicate of 5 independent experiments.

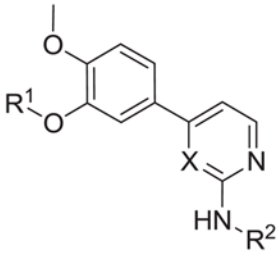
NIH-PA Author Manuscript

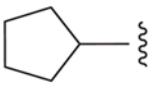
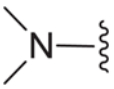
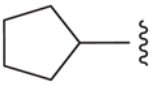
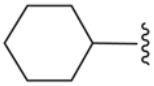
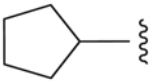
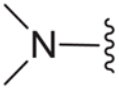
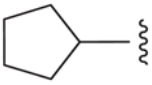
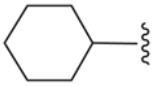
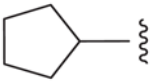
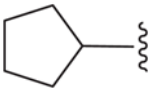
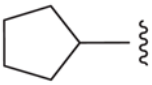
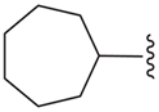
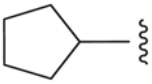
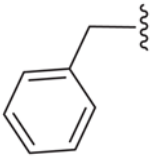
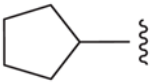
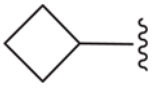
NIH-PA Author Manuscript

NIH-PA Author Manuscript

Table 2

Pyridine and Pyrimidine analogs tested against TbrPDEB1

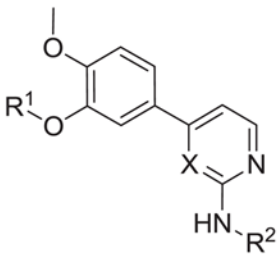


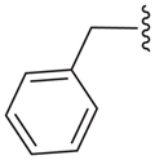
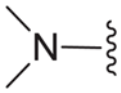
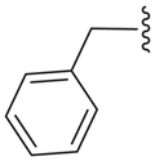
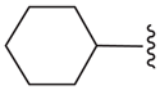
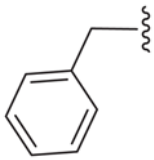
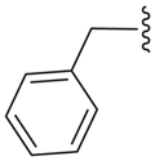
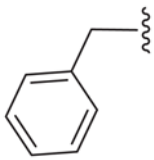
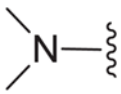
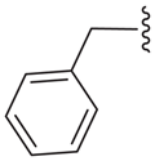
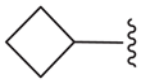
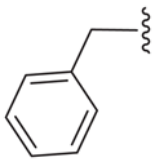
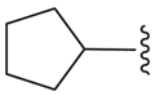
Compound	R^1	R^2	X	TbrPDEB1 (% inh)[a]
11a			C	34.0± 22.8
11b			C	27.1 ± 6.1
12a			N	15.8 ±21.2
12b			N	29.8 ± 0.3
12c			N	12.7 ± 17.9
12d			N	20.6 ± 8.8
12e			N	32.9 ± 6.0
12f			N	3.9 ± 0.5

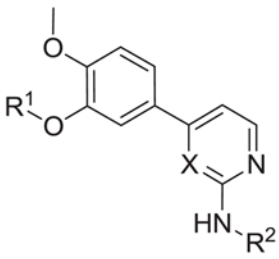
NIH-PA Author Manuscript

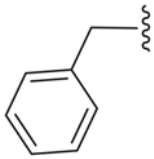
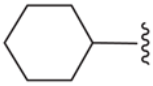
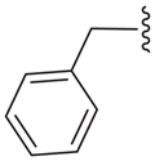
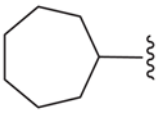
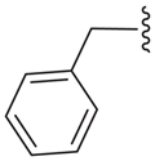
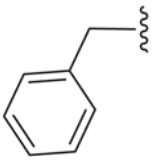
NIH-PA Author Manuscript

NIH-PA Author Manuscript



Compound	R^1	R^2	X	TbrPDEB1 (% inh)[a]
15a			C	48.0 ± 3.1
15b			C	28.4 ± 2.8
15c			C	7.2 ± 33
16a			N	14.5 ±20.5
16b			N	16.1 ± 22.8
16c			N	21.8 ± 4.6[b]



Compound	R^1	R^2	X	TbrPDEB1 (% inh)[a]
16d			N	23.2 ± 10.9
16e			N	22.3 ± 11.2
16f			N	18.5 ± 5.14

[a]Data shown are average of 2 replicate independent experiments.

[b]Replicate of 4 independent experiments.

NIH-PA Author Manuscript NIH-PA Author Manuscript NIH-PA Author Manuscript