Available online at www.sciencedirect.com
SCIENCE DIRECT®
Bioorganic & Medicinal Chemistry 12 (2004) 2691–2708
Bioorganic & Medicinal Chemistry

Synthesis and structure activity relationship of guanidines as NPY Y5 antagonists

Christopher J. Aquino,[a,*] Joshi M. Ramanjulu,[b] Dennis Heyer,[a] Alejandro J. Daniels,[c] Fabio Palazzo[a] and Milana Dezube[a]

[a]*Department of Medicinal Chemistry, GlaxoSmithKline, RTP, NC 27709, USA*
[b]*Department of Chemistry, GlaxoSmithKline, Collegeville, PA 19426, USA*
[c]*Department of Metabolic Diseases, GlaxoSmithKline, RTP, NC 27709, USA*

Received 18 August 2003; revised 19 February 2004; accepted 8 March 2004

Available online 15 April 2004

Abstract—A series of bis-aryl substituted guanidines have been discovered as potent NPY Y5 antagonists. The SAR and in vitro metabolic stability of these compounds are discussed.

1. Introduction

Neuropeptide Y (NPY), a 36-residue polypeptide has been linked to regulation of food intake. Continuous administration of NPY leads to hyperphagic condition in rodents and consequently, obesity. Abundantly expressed in the hypothalamus, NPY carries out a variety of neuroendocrine and autonomic functions through several G-protein coupled receptors. To date, five receptor subtypes, Y1–Y5, have been characterized and the existence of several others has been postulated. Prior to the characterization of the Y5 receptor, the Y1 receptor was suggested to be involved in food regulation using peptide antagonists such as 1229U91[1,2] and BIBP3226.[3] Since its discovery, however, numerous studies have implicated the Y5 receptor as an active component in the regulation of food consumption. The following are supportive of this hypothesis: (a) an ICV infusion of a Y5 selective peptide agonist induced feeding in rats;[4] (b) in vivo and in vitro affinities of various NPY agonists upon intracerebroventricular administration supported a role for the Y5 receptor subtype in feeding;[5] and (c) an intracerebroventricular injection of Y5 antisense oligodeoxynucleotides was shown to decrease food consumption in fasted rats.[6] In addition, the role of Y1 in regulating food intake has been brought into question by a study in which a potent, selective, Y1 agonist [Cys7,21,Pro34]NPY was found to be inactive at stimulating food intake.[7]

Traditionally, small molecules that bind selectively to a receptor have been used to validate biological targets. Therefore, small molecules that bind selectively to the Y5 receptor would be useful in elucidating the mechanism of action of NPY. Realizing this opportunity, many pharmaceutical companies have developed a diverse collection of small molecule Y5 antagonists. Investigators at Novartis reported a sub-nanomolar, highly selective cyclohexyl based arylsulfonamide that when administered intraperitoneally reduced food intake in animal models.[8] Similarly, we reported a selective benzimidazole Y5 antagonist that inhibited food intake and reduced body weight in rodents after intraperitoneal administration.[9] Other reports include quinazoline,[10] aminopyrimidine,[11] pyrazole,[12–14] imidazole,[15] thiazole,[16] and tetrahydrodiazabenzazulene[17] derivatives. More recently, carbamoyl derivatives,[18] aminotetralins,[19] and phenyl urea derivatives[20] have been claimed to be active as Y5 antagonists.

In this paper, we wish to disclose our research efforts directed toward the discovery of potent Y5 selective antagonists based on a bisheteroarylguanidine template. A high throughput screen of our in-house compound collection using a hY5 receptor binding assay revealed our initial lead, **9**. This compound was found to be

* Corresponding author. Tel.: +1-9194831232; fax: +1-9194836053; e-mail: cja9377@gsk.com

0968-0896/$ - see front matter © 2004 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmc.2004.03.012

highly potent (12 nM) on the Y5 receptor subtype and showed >50,000-fold selectivity against subtypes Y1–Y4. An in vitro rat liver S9 assay, however, indicated poor metabolic stability for this compound, which correlated in vivo with pharmacokinetic studies showing high clearance and low bioavailability. Accordingly, our objective was to identify other compounds in this class that maintained potency and selectivity on Y5 while improving in vitro stability in support of our efforts to identify orally efficacious Y5 antagonists.

2. Chemistry

In conducting the structure–activity relationship studies, we chose to concentrate on three regions of the molecule (Fig. 1): the 1,2,3,6-tetrahydropyridine (R^1), the guanidine moiety (R^3), and the phenyl ring (R^2). Our efforts were facilitated by a reliable synthetic scheme beginning with condensation of dicyandiamide **1** and ethylacetoacetate (Scheme 1).[21] The resulting intermediate, **2**, was reacted with amine nucleophiles to generate the guanidine functionality. The hydroxyl group was converted to the chloride by treatment with $POCl_3$, which was subsequently displaced with nucleophiles to furnish the desired analogs, **4**. This route was used extensively to explore the SAR of R^1 and R^2 regions.

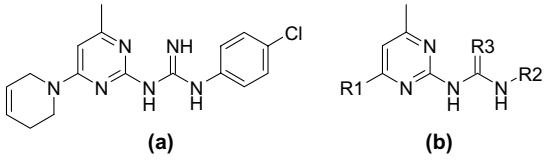
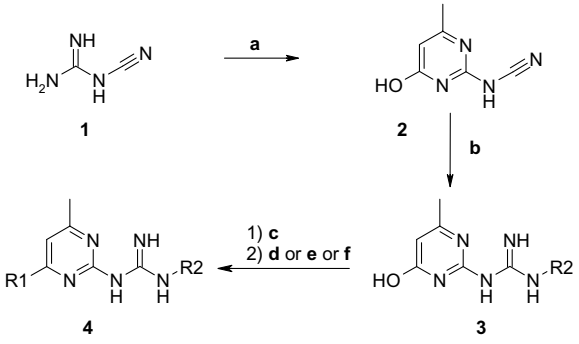


Figure 1. (a) **9**, initial lead; (b) generalized structure.

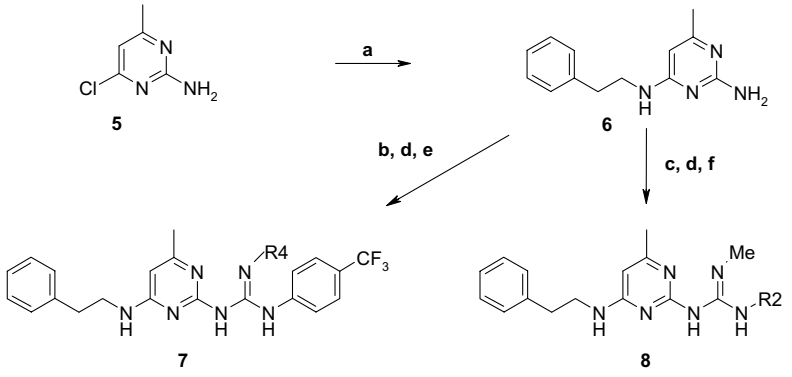


Scheme 1. General synthesis of bisarylguanidines. Reagents and conditions: (a) ethylacetoacetate, NaOMe, MeOH, reflux; (b) 2-ethoxyethanol, R2NH2; (c) $POCl_3$; (d) Amine, K_2CO_3, CH_3CN, reflux; (e) $Ph(CH_2)_2OH$; (f) $Ph(CH_2)_2SH$.

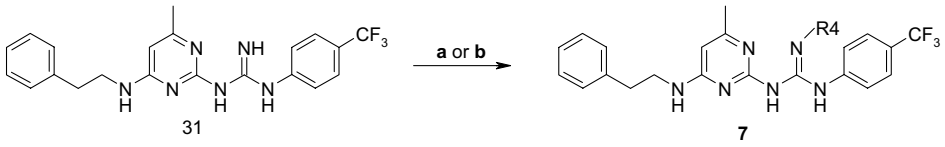
We have relied on two different approaches for the synthesis of trisubstituted guanidines. In the 'electrophile approach,' chloropyrimidine **5** was treated with phenethylamine to give **6**, which was subsequently reacted with isothiocyanates to generate intermediate thioureas.[22] The thioureas were activated by alkylation and reacted with amines and anilines to produce the desired analogs of type **7** and **8**[23] (Scheme 2). The 'nucleophile approach' was used to synthesize analogs that were not conveniently prepared by the electrophile approach. The disubstituted guanidine **31** was reacted with a variety of electrophiles to afford a novel set of analogs of type **7** (Scheme 3).

3. Results and discussion

Early work focused on finding more metabolically stable R^1 groups than the 1,2,3,6-tetrahydropyridine contained in the screening hit, **9** (Table 1). Varying ring size showed five- and six-membered rings to be approximately equipotent (**12** vs. **14**) whereas a four-membered ring was markedly less active (**11** vs. **13**). Incorporation of heteroatoms into the R^1 moiety typically resulted in decreased potency and provided evidence for the need for this group to be largely lipophilic in nature. While S was well tolerated (**15**, **16**), both hydrogen bond donating groups such as alcohol, **17**, and carboxylic acid, **18**, and hydrogen bond accepting groups such as ether, **19**, and amine, **41**, resulted in sharply diminished potency. Importantly, we consistently found that we could not optimize R^5/R^6 for both potency and metabolic stability simultaneously. While the more potent compounds were substituted with $R^5 = Cl$ (**9**, **12**, **15**), they lacked sufficient metabolic stability. By contrast, their respective counterparts substituted with $R^6 = CF_3$

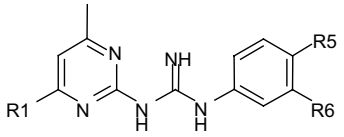


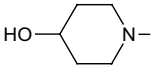
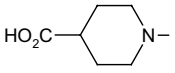
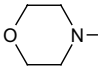
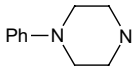
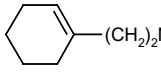
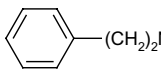
Scheme 2. Synthesis of tertiary guanidines—electrophile approach. Reagents and conditions: (a) phenethylamine, ethanol, NaOAc, reflux; (b) *p*-trifluoromethylphenylisothiocyanate, 1,4-dioxane, reflux; (c) CH_3NCS, 1,4-dioxane, reflux; (d) MeI, acetone; (e) $R4NH_2$, ethanol, reflux; (f) $R2NH_2$, ethanol, reflux.



Scheme 3. Synthesis of tertiary guanidines—nucleophile approach. Reagents and conditions: (a) R4X, CH_2Cl_2; (b) R4X, 1,4-dioxane, 60 °C.

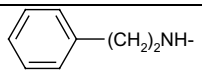
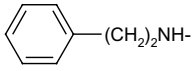
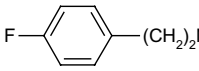
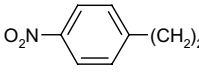
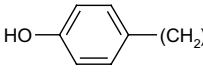
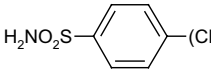
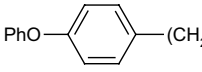
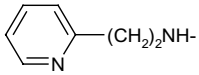
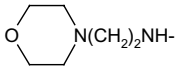
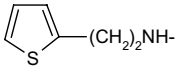
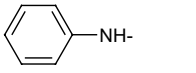
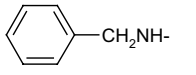
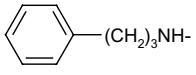
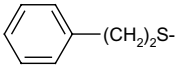
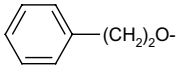
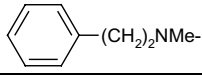
Table 1. Binding affinity and in vitro metabolic stability data for bisheteroarylguanidines



Compound	R1	R5	R6	IC_{50} (nM) ± SD	% Metabolized in RAT Liver S9[a]
9		Cl	H	12 ± 16.0 ($n = 5$)	89
10		H	CF_3	88 ± 1.4 ($n = 2$)	35
11		H	CF_3	476 ± 69.5 ($n = 2$)	—
12		Cl	H	16 ± 0.3 ($n = 2$)	97
13		H	CF_3	126 ± 16.4 ($n = 2$)	68
14		Cl	H	22 ± 0.3 ($n = 2$)	79
15		Cl	H	32 ± 0.0 ($n = 1$)	100
16		H	CF_3	135 ± 0 ($n = 2$)	21
17		Cl	H	⩾3139 ± 79 ($n = 2$)	—
18		Cl	H	⩾1950 ± 63.5 ($n = 2$)	—
19		Cl	H	446	—
20		CF_3	H	1503 ± 319.7 ($n = 6$)	—
21	CH_3NH–	H	CF_3	933 ± 0.0 ($n = 2$)	—
22	CH_3CH_2NH–	H	CF_3	421 ± 81.7 ($n = 2$)	—
23	$CH_3(CH_2)_3NH$–	H	CF_3	150 ± 38.8 ($n = 2$)	30
24	$CH_3(CH_2)_4NH$–	H	CF_3	77 ± 0.0 ($n = 2$)	19
25	$CH_3(CH_2)_5NH$–	H	CF_3	69 ± 4.5 ($n = 2$)	25
26	$CH_3(CH_2)_7NH$–	CF_3	H	667 ± 129.5 ($n = 2$)	—
27	$(CH_3)_2CH(CH_2)_2NH$–	H	CF_3	75 ± 0.0 ($n = 2$)	93
28		H	CF_3	15 ± 7.7 ($n = 2$)	68
29		H	CF_3	28 ± 6.4 ($n = 9$)	14

(*continued on next page*)

Table 1 (*continued*)

Compound	R1	R5	R6	IC_{50} (nM) ± SD	% Metabolized in RAT Liver S9[a]
30		Cl	H	17 ± 20.7 ($n = 5$)	24
31		CF_3	H	6 ± 1.0 ($n = 18$)	15
32		H	CF_3	39 ± 6.2 ($n = 4$)	21
33		H	CF_3	49 ± 4.7 ($n = 4$)	23
34		H	CF_3	⩾1905 ± 0 ($n = 2$)	—
35		H	CF_3	553 ± 89.7 ($n = 2$)	—
36		H	CF_3	180 ± 8.8 ($n = 2$)	33
37		H	CF_3	155 ± 0 ($n = 2$)	30
38		Cl	H	⩾1995 ($n = 1$)	—
39		H	CF_3	29 ± 3.3 ($n = 2$)	9
40		Cl	H	54 ± 0.0 ($n = 2$)	47
41		Cl	H	208 ($n = 1$)	36
42		H	CF_3	74 ± 13.2 ($n = 2$)	8
43		CF_3	H	⩾1905 ± 0 ($n = 2$)	—
44		H	CF_3	⩾1905 ± 0 ($n = 2$)	—
45	$(CH_3CH_2)_2NH$–	Cl	H	386 ($n = 1$)	—
46		Cl	H	388 ($n = 1$)	—

[a] 60-min incubation (as described in Biological Methods and Materials).

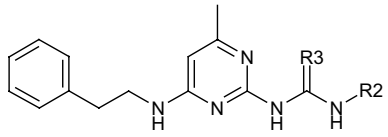
(**10**, **13**, **16**) demonstrated sufficient metabolic stability but significantly decreased potency. As such, we chose to maintain the trifluoromethyl group at either R^5 or R^6 for metabolic stability while seeking to improve potency with other R^1 ligands.

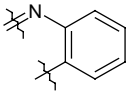
In general, primary amines of reasonable size and lipophilicity (e.g., phenethylamine and substituted analogs) were found to possess optimal potency, with an evident upper limit on size/lipophilicity. A progression of linear alkyl substituents from methylamine through octylamine (**21**–**26**) revealed steadily increasing potency as chain length/lipophilicity was increased to hexylamine, **25**, after which a sharp decrease in potency was seen. Branched primary amines further demonstrated the ability to incorporate large aliphatic groups to good

effect at this position as demonstrated by compounds **27** and **28**. This finding was capitalized on with phenethylamine, which yielded both highly potent and metabolically stable analogs (**29**–**31**). The phenyl ring could be substituted with electron-withdrawing groups with only modest effect (**32**, **33**). Consistent with results of the cyclic secondary amines, we found that incorporation of hydrogen bond donating groups (**34**, **35**) resulted in a marked decrease in potency. Likewise, incorporation of hydrogen bond acceptors either as pendant ether, **36**, or as a phenyl ring replacement such as pyridyl, **37**, or morpholino, **38**, resulted in a significant loss or elimination of potency. Also in line with earlier results, an electroneutral thiophene, **39**, yielded an equipotent phenyl ring replacement. Varying the spacer between the phenyl ring and the amine nitrogen (**40**–**42**) confirmed phenethyl to be optimal. Replacement of the amine N with alternate heteroatoms, S and O, eliminated activity as demonstrated by compounds **43** and **44**, respectively. Linear secondary amines were found to be significantly less potent than their cyclic secondary amine (**45** vs. **12**) or linear primary amine (**46** vs. **30**) counterparts.

With R^1 thus set as phenethylamine, the SAR of R^2 was explored (Table 2). The intent here was to maintain potency while improving the in vitro metabolic stability profile. Numerous analogs were synthesized to probe the influence of electronic effects. This set was generated by 'walking' chloro, methoxy, methyl, trifluoromethyl, fluoro, and di-fluoro groups around the phenyl ring. Surprisingly, there was only a modest difference in potency among these compounds that could not be attributed to electronics. There was, however, some loss in potency attributable to sterics as demonstrated by substitution of the ortho-position with bulky groups (**56**, **58**). Replacement of the phenyl ring with the heterocycles 2-pyridyl, **68**, or 3-pyridyl, **69**, resulted in a marked decrease in potency, whereas replacement with a benzyl group, **66**, was tolerated. Cyclization of the guanidino moiety onto the phenyl ring to give a benzimidazole, **70**, also resulted in a decrease in potency. What distinguished these compounds from each other, ultimately, was their metabolic stability. Here, a clear preference for electron-withdrawing groups was noted and compounds such as **29**, **31**, and **64**, containing fluoro or

Table 2. Binding affinity and in vitro metabolic stability data for bisheteroarylguanidines and tertiary guanidines



Compound	R2	R3	$hIC_{50} \pm$ (nM)SD	% Metabolized in RAT Liver S9[a]
47	Ph	NH	45 ± 4.2 ($n = 3$)	32
48	Ph(2-F)	NH	46 ± 22.4 ($n = 7$)	29
49	Ph(3-F)	NH	12 ($n = 1$)	35
50	Ph(4-F)	NH	76 ($n = 1$)	31
51	Ph(2-Cl)	NH	53 ($n = 1$)	33
52	Ph(3-Cl)	NH	9 ($n = 1$)	30
30	Ph(4-Cl)	NH	17 ± 20.7 ($n = 5$)	24
53	Ph(2-OCH_3)	NH	15 ($n = 1$)	51
54	Ph(3-OCH_3)	NH	11 ($n = 1$)	59
55	Ph(4-OCH_3)	NH	154 ± 84.4 ($n = 3$)	45
56	Ph(2-CH_3)	NH	141 ($n = 1$)	27
57	Ph(3-CH_3)	NH	14 ($n = 1$)	41
58	Ph(2-CF_3)	NH	589 ± 101.5 ($n = 4$)	—
29	Ph(3-CF_3)	NH	28 ± 6.3 ($n = 9$)	14
31	Ph(4-CF_3)	NH	6 ± 1.0 ($n = 18$)	15
59	Ph(2,3-F_2)	NH	51 ± 7.4 ($n = 2$)	24
60	Ph(2,4-F_2)	NH	95 ± 6.2 ($n = 2$)	20
61	Ph(2,5-F_2)	NH	38 ± 0.6 ($n = 2$)	35
62	Ph(2,6-F_2)	NH	158 ± 41.5 ($n = 2$)	16
63	Ph(3,4-F_2)	NH	38 ± 14.5 ($n = 2$)	12
64	Ph(3,5-F_2)	NH	18 ± 1.5 ($n = 2$)	0
65	Ph(3-NO_2)	NH	16 ($n = 1$)	33
66	Bn	NH	74 ($n = 1$)	65
67	Ph(4-CF_3)	NCH_3	4 ± 0.5 ($n = 6$)	10
68	2-Pyridyl	NCH_3	436 ± 42.6 ($n = 2$)	—
69	3-Pyridyl	NCH_3	812 ± 66.9 ($n = 2$)	—
70			851 ± 70.1 ($n = 2$)	—

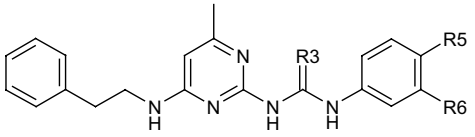
[a] 60-min incubation (as described in Biological Methods and Materials).

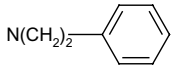
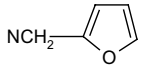
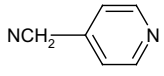
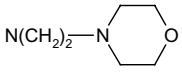
trifluoromethyl substituted phenyl groups, surfaced as having the best composite potency/stability profile.

With R^1 and R^2 group thus set, we turned our attention to the SAR of the R^3 position (Table 3). While this position can tolerate a variety of electronic features, it does show a strict limitation on size and bulk. R^3 groups built up from small, aliphatic, primary amines (**67**, **71**–**73**, **75**) uniformly demonstrated increased potency at this position as did their oxygen containing analogs, **74** and **76**. Importantly, these compounds maintained the high metabolic stability of their 'parent' compound, **31**. The closely related analogs built up from secondary amines (**77**, **78**), however, showed a significant drop in potency due, presumably, to their increased bulk. Larger aliphatic groups such as aniline, **79**, and phenethylamine, **81**, showed markedly decreased potency that could be rescued to a degree by introduction of polarity in the form of an ether (**80**) or heterocycles such as furan (**82**), pyridyl (**83**), or morpholine (**84**). Electron-withdrawing functionalities were reasonably well tolerated when they were smaller groups (**85**, **86**) but lost potency as their size increased (**87**, **88**). Replacement of the central guanidino functionality with urea, **89**, or thiourea, **90**, yielded inactive compounds.

Optimized compounds from these investigations were subsequently evaluated for favorable drug-like properties according to our progression criteria. Compounds with good equilibrium solubility in simulated intestinal fluid and permeability in a MDCK cell line were progressed into rodent models for in vivo studies. Several of these guanidine-based NPY5 ligands demonstrated favorable whole brain exposure (concentrations >100-fold hIC_{50}) following oral dosing. For example, the concentration of compounds **67** and **71** in whole brain

Table 3. Binding affinity and in vitro metabolic stability data for bisheteroarylguanidines and tertiary guanidines



Compound	R3	R5	R6	IC_{50} (nM) ± SD	% Metabolized in RAT Liver S9[a]
29	NH	H	CF_3	28 ± 6.3 ($n = 9$)	14
31	NH	CF_3	H	6 ± 1.0 ($n = 18$)	15
67	NCH_3	CF_3	H	4 ± 0.5 ($n = 6$)	10
71	NCH_2CH_3	CF_3	H	1 ± 0.3 ($n = 4$)	9
72	$NCH(CH_3)_2$	CF_3	H	2 ± 0.7 ($n = 2$)	—
73	$N(CH_2)_2CH_3$	CF_3	H	2 ± 0.4 ($n = 2$)	26
74	NCH_2CH_2OH	CF_3	H	1 ± 0.3 ($n = 2$)	—
75	$N(CH_2)_3CH_3$	CF_3	H	6 ± 1.9 ($n = 2$)	13
76	$N(CH_2)_2OCH_3$	CF_3	H	3 ± 0.1 ($n = 2$)	23
77	$N(CH_3)_2$	CF_3	H	⩾1905 ± 0 ($n = 2$)	—
78		CF_3	H	⩾1905 ± 0 ($n = 2$)	—
79	NPh	CF_3	H	630 ± 199.0 ($n = 2$)	—
80	$NCH_2Ph(4\text{-}OCH_3)$	CF_3	H	41 ± 7.2 ($n = 2$)	26
81		CF_3	H	158 ± 25.8 ($n = 2$)	—
82		CF_3	H	11 ± 2.6 ($n = 2$)	32
83		CF_3	H	20 ± 3.3 ($n = 2$)	23
84		CF_3	H	34 ± 0.55 ($n = 2$)	51
85	NCH_2CF_3	CF_3	H	17 ± 0 ($n = 2$)	19
86	$NCOCH_3$	CF_3	H	28 ± 2.3 ($n = 2$)	0
87	NCO_2CH_2Ph	CF_3	H	⩾1905 ± 0 ($n = 2$)	—
88	$NCONHCH_2CH_3$	CF_3	H	⩾1905 ± 0 ($n = 2$)	—
89	O	Cl	H	⩾1905 ± 0 ($n = 2$)	—
90	S	CF_3	H	⩾1905 ± 0 ($n = 2$)	—

[a] 60-min incubation (as described in Biological Methods and Materials).

following oral administration (25 mg/kg as a suspension in methyl cellulose) were 0.45 and 0.68 μM (336- and 170-fold IC_{50}, at the 2.5 h time point), respectively.[24] Accordingly, these compounds may be useful in determining the effect on feeding and body weight of NPY5 antagonists in rodents. Further in vivo experiments with these compounds could enable us to elucidate the role of NPY Y5 in appetite regulation.

4. Conclusion

We have discovered a novel series of potent, small molecule NPY antagonists with high selectivity toward the Y5 receptor. SAR investigations resulted in several highly stable compounds with favorable in vitro permeability, solubility properties, and in vivo brain penetrability. Our investigations centered on three portions of the molecule. Electron-withdrawing groups were necessary on the aryl ring (R^2) for improved metabolic stability. A trifluoromethyl group on this ring provided the optimal in vitro profile. Modifications of the R^1 region exhibited a high degree of tolerance toward lipophilic groups with an upper limit on size. The nitrogen atom attached to the pyrimidine ring in this moiety was required for affinity to the receptor. Changes to the guanidine moiety (R^3) generally resulted in more potent analogs when they were restricted to smaller groups. Compounds with smaller lipophilic groups exhibited improved pharmacokinetics in the rat. Further experiments in animal models would indicate the potential use of these entities as pharmacological agents for obesity.

5. Experimental section

5.1. General

All commercial chemicals and solvents are reagent grade and were used without further purification unless otherwise specified. The yields for some reactions are unoptimized. Analytical thin-layer chromatography (TLC) was performed on Merck silica gel (60 F-254) plates (0.25 mm) precoated with a fluorescence indicator. Visualization was affected using standard procedures unless otherwise stated. Flash column chromatography was carried out on EM silica gel 60 particle size (0.04–0.063 mm). Melting points (mp) were determined with a Mel-Temp melting point apparatus. Proton and carbon magnetic resonance spectra (^1H, ^{13}C NMR) were recorded on a Varian 300 or 400 MHz instrument, and chemical shifts were expressed in parts per million (ppm, δ units). Coupling constants are reported in hertz (Hz). Splitting patterns are designated as s, singlet; br s, broad singlet; d, doublet; t, triplet; q, quartet; m, multiplet. Low-resonance mass spectra (MS) were obtained using Micromass platform and acquired in the positive ion mode under ESI^+ method and reported in the form of m/z. High-resolution mass spectral (HRMS) data were recorded using a Micromass LCT time-of-flight mass spectrometer using the positive-ion electrospray (ESI^+) mode. Accurate mass data was obtained by using reserpine as the lock mass ($m/z = 609.2812$) reference. Elemental analysis were performed by Atlantic Mircrolabs, Inc., Norcross, GA. The following solvents and reagents have been abbreviated: dichloromethane (DCM, CH_2Cl_2), dimethyl sulfoxide (DMSO), tetrahydrofuran (THF), ethyl acetate (EtOAc), diisopropyl ethylamine (DIEA), phosphorous oxychloride ($POCl_3$).

5.2. Biological methods and materials

5.2.1. Cell culture. CHO cells were transfected to stably express the cloned NPY-Y1, Y2, Y4, and Y5 receptors as previously described[25] and were grown in Kaighn's Nutrient Mixture F-12 media supplemented with 500 units/mL Penicillin–Streptomycin, 0.5 mg/mL Geneticin, and 10% fetal calf serum. Cells were maintained at 37 °C with 95% air and 5% CO_2.

5.2.2. Radioreceptor binding assays. Binding of [125I]-PYY (0.08 nM; ~50,000 cpm) to membranes from CHO cells expressing the human Y1, Y2, Y4, or Y5 receptors was determined using scintillation proximity assay methodology (Amersham, Arlington, IL) as previously described.[25] Briefly, membranes (Y1 = 3 μg/tube, Y2 = 0.2 μg/tube, Y4 = 0.5 μg/tube, and Y5 = 7.5 μg/tube) were incubated overnight under constant shaking at room temperature with wheat germ agglutinin beads in the presence of increasing concentrations of test compounds in 50 mM HEPES, pH 7.4 containing 2.5 mM $CaCl_2$, 1 mM $MgCl_2$, 2.5% bacitracin, and 0.1% BSA. Specific binding (85%) was determined by incubation in the presence of 1 μM PYY. Experiments were performed in triplicate and repeated at least three times. Competitive binding curves were fit to models of ligand binding to single and multiple receptor sites using a proprietary GlaxoSmithKline curve fitting software. K_i values were calculated utilizing the Cheng and Prusoff method.[26]

5.2.3. In vitro metabolic stability—rat liver S9 assay. Each compound (10 μM final concentration) was incubated at 37 °C in pooled S9 post-mitochondrial homogenates prepared from rat livers (Xenotech, LLC, Kansas City, KS). Stock solutions of the test compounds were prepared in DMSO such that the final DMSO concentration in the reaction mixture (0.5 mL) was <1%. All reactions were initiated by addition of an NADPH regeneration system supplemented with UDPGA as described by Wring et al.[27] At time 0 (preincubation) and 60 min, the reaction (200-μL aliquot) was stopped by addition of cold acetonitrile (400 μL). The resulting samples were vortexed and centrifuged (2600 rcf, 15 min), and the supernatants were analyzed for parent compound by LC/MS/MS. Metabolic stability, expressed as the amount (%) of compound

metabolized, was determined by comparison of peak areas in the pre- and post-incubation samples.

5.3. Chemical synthesis

5.3.1. General procedure A, for the synthesis of analogs of type 4 (9–66, Scheme 1)

5.3.1.1. 4-Hydroxy-6-methyl-2-pyrimidinylcyanamide (2). Dissolve dicyandiamide (**1**, 67.26 g, 800 mmol) and ethyl acetoacetate (124.9 g, 960 mmol) in MeOH (500 mL) followed by dropwise addition of NaOMe (182 mL of a 4.4 M solution in MeOH, 800 mmol). Heat at reflux for 2 h during which a precipitate forms. Cool, filter, and wash with cold MeOH to obtain ~125 g of the sodium salt. This material was dissolved in water (2 L) and glacial acetic acid added until solution reached pH 5. A white precipitate formed and was filtered, washed with water and dried in vacuum oven to obtain 81 g of the desired product. Combining the amounts obtained from second and third crops produced the overall yield of 74% of the title compound, **2**, as a white solid.[21]

5.3.1.2. *N*-(4-Hydroxy-6-methyl-2-pyrimidinyl)-*N*′-arylguanidines (3). The selected anilines (2 equiv) and **2** (1 equiv) were dissolved in ethoxyethanol (0.5 M) and stirred at 140 °C overnight. Cool to room temperature and place in an ice bath. The resulting precipitate was filtered and washed with cold ethanol (minimum amount) followed by diethyl ether. The products were air dried to give the desired products, **3**, in moderate yields.

5.3.1.3. *N*-(4-Chloro-6-methyl-2-pyrimidinyl)-*N*′-arylguanidines. The intermediates **3** (15 mmol) were dissolved in $POCl_3$ (40 mL) and stirred at room temperature overnight. The $POCl_3$ was removed in vacuo and crushed ice added to the residue. After the remaining $POCl_3$ was consumed, a white solid formed, which was broken up with a spatula. A stir bar was added and the chunky solid was converted to a fine crystal with vigorous stirring. Filter and wash with water to remove any remaining acid. Upon drying, the desired chlorides were obtained in good yields.

5.3.1.4. General procedure for the chlorine displacement reaction. Amine (2 equiv) and chloride (1 equiv) were dissolved in ethanol (0.2 M). K_2CO_3 (2 equiv) was added and the reaction was heated to reflux overnight. While hot, water was added until all K_2CO_3 was dissolved. The solution was then allowed to come to room temperature and placed in an ice bath. Cold water was added to the reaction mixture and a white precipitate formed, which was filtered and washed with cold appropriate solvent. In the event the product did not precipitate, usual reaction work up was implemented. Purification methods were tailored for each reaction and are outlined below to give compounds of type **4**.

5.3.1.5. *N*-(4-Chlorophenyl)-*N*′-[4-(3,6-dihydro-1(2*H*)-pyridinyl)-6-methyl-2-pyrimidinyl]guanidine (9). The crude product was washed with water and air dried to produce the desired product in 45% (262 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz, 100 °C) δ 8.30 (br s, 1H), 7.51 (d, 2H, $J = 8.8$ Hz), 7.40 (d, 2H, $J = 8.8$ Hz), 6.45 (s, 1H), 5.93 (m, 1H), 5.79 (m, 1H), 4.10 (m, 2H), 3.76 (t, 2H, $J = 5.7$ Hz), 2.30 (s, 3H), 2.24 (m, 2H); MS (ESI$^+$) m/e 343.2 (M+H); HRMS calcd for $C_{17}H_{19}N_6Cl$ (M+H) 343.1438, found 343.1454.

5.3.1.6. *N*-[4-(3,6-Dihydro-1(2*H*)-pyridinyl)-6-methyl-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (10). The crude product was washed with water and air dried to produce the desired product in 77% (220 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.76 (br s, 1H), 7.65 (br s, 1H), 7.58–7.20 (m, 2H), 7.44 (t, 1H, $J = 7.6$ Hz), 7.20 (d, 1H, $J = 7.6$ Hz), 6.25 (s, 1H), 5.88 (d, 1H, $J = 10.2$ Hz), 5.76 (d, 1H, $J = 10.2$ Hz), 2.19 (s, 3H), 2.18–2.14 (m, 2H); MS (ESI$^+$) m/e 377.43 (M+H); MS (ESI$^-$) m/e 375.27 (M−1); Anal. Calcd for $C_{18}H_{19}F_3N_6$: C, 57.44; H, 5.09; N, 22.33. Found: C, 57.50; H, 5.08; N, 22.42.

5.3.1.7. *N*-[4-(1-Azetidinyl)-6-methyl-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (11). The crude product was washed with water and air dried to produce the desired product in 11% (30 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.60 (br s, 1H), 7.90–7.00 (m, 6H), 7.44 (t, 1H, $J = 7.6$ Hz), 7.20 (d, 1H, $J = 7.6$ Hz), 5.78 (s, 1H), 3.97 (t, 4H, $J = 7.5$ Hz), 2.32 (pent, 2H, $J = 7.6$ Hz), 2.15 (s, 3H); MS (ESI$^+$) m/e 350.93 (M+H); Anal. Calcd for $C_{16}H_{17}F_3N_6$: C, 54.85; H, 4.89; N, 23.99. Found: C, 54.92; H, 4.97; N, 23.61.

5.3.1.8. *N*-(4-Chlorophenyl)-*N*′-[4-methyl-6-(1-pyrrolidinyl)-2-pyrimidinyl]guanidine (12). The crude product was washed with water and air dried to produce the desired product in 73% (410 mg) yield. ^{1}H NMR (CD_3OD, 300 MHz) δ 7.30 (d, 2H, $J = 8.6$ Hz), 7.12 (d, 2H, $J = 8.6$ Hz), 5.97 (s, 1H), 3.58–3.42 (m, 4H), 2.24 (s, 3H), 2.08–1.95 (m, 4H); MS (ESI$^+$) m/e 331.1, 333.1 (M+H); Anal. Calcd for $C_{16}H_{19}ClN_6$ $0.4H_2O$: C, 56.85; H, 5.90; N, 24.86. Found: C, 56.89; H, 5.60; N, 25.06.

5.3.1.9. *N*-[4-Methyl-6-(1-pyrrolidinyl)-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (13). The crude product was washed with water and air dried to produce the desired product in 64% (176 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.60 (br s, 1H), 7.70 (br s, 1H), 7.50–7.15 (m, 5H), 5.93 (s, 1H), 3.45–3.30 (m, 4H), 2.16 (s, 3H), 1.98–1.80 (m, 4H); MS (ESI$^+$) m/e 365.45 (M+H); MS (ESI$^-$) m/e 363.32 (M−1); HRMS calcd for $C_{17}H_{19}F_3N_6$ (M+H) 365.1702, found 365.1703.

5.3.1.10. *N*-(4-Chlorophenyl)-*N*′-[4-methyl-6-(1-piperidinyl)-2-pyrimidinyl]guanidine (14). The crude product was washed with water and air dried to produce the desired product in 65% (383 mg) yield. ^{1}H NMR (CD_3OD, 300 MHz) δ 7.35–7.25 (m, 2H), 7.15–7.05 (m, 2H), 6.24 (s, 1H), 3.68–3.55 (m, 4H), 2.24 (s, 3H), 1.75–1.68 (m, 2H), 1.68–1.55 (m, 4H); MS (ESI$^+$) m/e 345.1, 347.1 (M+H); Anal. Calcd for $C_{17}H_{21}ClN_6$ $0.25H_2O$: C, 58.45; H, 6.20; N, 24.06. Found: C, 58.41; H, 5.96; N, 24.28.

5.3.1.11. *N*-(4-Chlorophenyl)-*N*′-[4-methyl-6-(1,3-thiazolidin-3-yl)-2-pyrimidinyl]guanidine (15). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, chased with EtOH, and pumped dry to produce the desired product in 41% (247 mg) yield. ^1H NMR ($CDCl_3$, 300 MHz) δ 7.26 (d, 2H, $J = 8.7$ Hz), 6.95 (d, 2H, $J = 8.7$ Hz), 5.85 (s, 1H), 5.30 (s, 1H), 4.55 (s, 2H), 3.74 (t, 2H, $J = 6.2$ Hz), 3.12 (t, 2H, $J = 6.2$ Hz), 2.26 (s, 3H); MS (ESI^+) m/e 349.0, 351.0 (M+H); Anal. Calcd for $C_{15}H_{17}ClN_6S$ $0.25C_2H_5OH$: C, 51.25; H, 5.22; N, 23.52. Found: C, 51.56; H, 5.09; N, 23.50.

5.3.1.12. *N*-[4-Methyl-6-(1,3-thiazolidin-3-yl)-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (16). Dioxane was used in place of CH_3OH as the reaction solvent and heated 16 h at 100 °C. The crude product was washed with water and air dried to produce the desired product in 90% (260 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.90 (br s, 1H), 7.70 (br s, 1H), 7.47–7.42 (m, 3H), 7.22–7.18 (m, 1H), 6.13 (s, 1H), 4.56 (s, 2H), 3.70 (t, 2H, $J = 6.3$ Hz), 3.11 (t, 2H, $J = 6.3$ Hz), 2.21 (s, 3H); MS (ESI^+) m/e 383.35 (M+H); MS (ESI^-) m/e 381.22 (M−1); Anal. Calcd for $C_{16}H_{17}F_3N_6S$ 0.10 dioxane: C, 50.35; H, 4.59; N, 21.48. Found: C, 50.44; H, 4.60; N, 21.37.

5.3.1.13. *N*-(4-Chlorophenyl)-*N*′-[4-(4-hydroxy-1-piperidinyl)-6-methyl-2-pyrimidinyl]guanidine (17). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, and pumped dry to produce the desired product in 48% (295 mg) yield. ^1H NMR ($CDCl_3$, 300 MHz) δ 7.45 (d, 2H, $J = 8.9$ Hz), 7.35 (d, 2H, $J = 8.7$ Hz), 6.08 (s, 1H), 4.06–3.92 (m, 3H), 3.35–3.22 (m, 2H), 2.25 (s, 3H), 2.00–1.90 (m, 2H), 1.62–1.47 (m, 2H); MS (ESI^+) m/e 361.1, 363.1 (M+H); HRMS calcd for $C_{17}H_{21}N_6OCl$ (M+H) 361.1544, found 361.1557.

5.3.1.14. 1-(2-{[(4-Chloroanilino)(imino)methyl]amino}-6-methyl-4-pyrimidinyl)-4-piperidinecarboxylic acid (18). The crude product was washed with water and air dried to produce the desired product in 73% (480 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 7.32–7.10 (m, 4H), 6.25 (s, 1H), 4.20–4.08 (m, 2H), 3.10–2.95 (m, 2H), 2.51–2.48 (m, 1H), 2.17 (s, 3H), 1.87–1.77 (m, 2H), 1.53–1.38 (m, 2H); MS (ESI^+) m/e 389.1, 391.1 (M+H); Anal. Calcd for $C_{18}H_{21}ClN_6O_2$ $1.25H_2O$: C, 52.55; H, 5.76; N, 20.43. Found: C, 52.77; H, 5.59; N, 20.78.

5.3.1.15. *N*-(4-Chlorophenyl)-*N*′-[4-methyl-6-(4-morpholinyl)-2-pyrimidinyl]guanidine (19). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, and pumped dry to produce the desired product in 52% (309 mg) yield. ^1H NMR ($CDCl_3$, 300 MHz) δ 7.27 (d, 2H, $J = 8.7$ Hz), 6.98 (d, 2H, $J = 8.2$ Hz), 5.97 (s, 1H), 3.80–3.73 (m, 4H), 3.56–3.53 (m, 4H), 2.26 (s, 3H); MS (ESI^+) m/e 347.1, 349.1 (M+H); HRMS calcd for $C_{16}H_{19}N_6OCl$ (M+H) 347.1387, found 347.1381.

5.3.1.16. *N*-[4-Methyl-6-(4-phenyl-1-piperazinyl)-2-pyrimidinyl]-*N*′-[4-(trifluoromethyl)phenyl]guanidine (20). The crude product was washed with water and air dried to produce the desired product in 73% yield. ^1H NMR (DMSO-d_6, 400 MHz) δ 8.90 (br s, 1H), 7.34–7.22 (m, 3H), 6.97 and 6.58 (AB, 4H, $J = 9$ Hz), 6.81–6.77 (m, 2H), 6.34 (s, 1H), 3.72–3.70 (m, 4H), 3.20–3.18 (m, 4H), 2.20 (s, 3H); HRMS calcd for $C_{23}H_{24}F_3N_7$ (M+H) 455.4861, found 456.2104; HRMS calcd for $C_{21}H_{20}F_3N_5S$ (M+H) 431.4840, found 432.1454. Anal. Calcd for $C_{23}H_{24}F_3N_7$ $(0.01H_2O)$: C, 60.63; H, 5.31; N, 21.52. Found: C, 60.62; H, 5.25; N, 21.34.

5.3.1.17. *N*-[4-Methyl-6-(methylamino)-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (21). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 39% (95 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.50 (br s, 1H), 7.70 (br s, 1H), 7.45–7.17 (m, 5H), 5.86 (s, 1H), 2.73 (d, 3H, $J = 4.4$ Hz), 2.12 (s, 3H); MS (ESI^-) m/e 323.24 (M−1); HRMS calcd for $C_{14}H_{15}N_6F_3$ (M+H) 325.1389, found 325.1381.

5.3.1.18. *N*-[4-(Ethylamino)-6-methyl-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (22). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 42% (107 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.50 (br s, 1H), 7.70 (br s, 1H), 7.46–7.17 (m, 5H), 5.86 (s, 1H), 3.20 (m, 2H), 2.11 (s, 3H), 1.09 (t, 3H, $J = 7.1$ Hz); MS (ESI^-) m/e 337.26 (M−1); Anal. Calcd for $C_{15}H_{17}F_3N_6$ $0.1CH_3CN$: C, 53.64; H, 5.06; N, 24.78. Found: C, 53.66; H, 5.08; N, 24.66.

5.3.1.19. *N*-[4-(Butylamino)-6-methyl-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (23). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 43% (119 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.50 (br s, 1H), 7.70 (br s, 1H), 7.45–7.17 (m, 5H), 5.86 (s, 1H), 3.31 (m, 2H) 2.10 (s, 3H), 1.48–1.41 (m, 2H), 1.36–1.24 (m, 2H), 0.87 (t, 3H, $J = 7.2$ Hz); MS (ESI^+) m/e 367.39 (M+H); MS (ESI^-) m/e 365.33 (M−1); HRMS calcd for $C_{17}H_{21}N_6F_3$ (M+H) 367.1858, found 367.1848.

5.3.1.20. *N*-[4-Methyl-6-(pentylamino)-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (24). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 46% (135 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.55 (br s, 1H), 7.70 (br s, 1H), 7.45–7.17 (m, 5H), 5.86 (s, 1H), 3.16 (m, 2H), 2.10 (s, 3H), 1.47 (m, 2H), 1.29–1.24 (m, 4H), 0.85 (t, 3H, $J = 6.5$ Hz); MS (ESI^+) m/e 381.38 (M+H); MS (ESI^-) m/e 379.27 (M−1);

HRMS calcd for $C_{18}H_{23}N_6F_3$ (M+H) 381.2015, found 381.2028.

5.3.1.21. *N*-[4-(Hexylamino)-6-methyl-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (25). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 20% (61 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.45 (br s, 1H), 7.65 (br s, 1H), 7.45–7.17 (m, 5H), 5.86 (s, 1H), 3.18 (m, 2H), 2.10 (s, 3H), 1.53–1.42 (m, 2H), 1.38–1.23 (m, 6H), 0.84 (t, 3H, $J = 7.0$ Hz); MS (ESI^+) m/e 395.38 (M+H); MS (ESI^-) m/e 393.24 (M−1); HRMS calcd for $C_{19}H_{25}N_6F_3$ (M+H) 395.2171, found 395.2175.

5.3.1.22. *N*-[4-Methyl-6-(octylamino)-2-pyrimidinyl]-*N*′-[4-(trifluoromethyl)phenyl]guanidine (26). The crude product was washed with water and air dried to produce the desired product in 73% (0.50 g) yield. ^1H NMR (DMSO-d_6, 400 MHz) δ 8.40 (s, 2H), 7.45 (d, 2H, $J = 8$ Hz), 7.20 (br s, 1H), 5.81 (s, 1H), 5.86 (s, 1H), 2.09 (s, 3H), 1.45–1.40 (m, 2H), 1.22–1.20 (m, 12H), 0.80 (t, 3H, $J = 8$ Hz); HRMS calcd for $C_{21}H_{29}F_3N_6$ (M+H) 423.2484, found 423.2480.

5.3.1.23. *N*-[4-(Isopentylamino)-6-methyl-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (27). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 66% (190 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.50 (br s, 1H), 7.70 (br s, 1H), 7.45–7.17 (m, 5H), 5.86 (s, 1H), 3.20 (m, 2H), 2.10 (s, 3H), 1.59 (sept, 1H, $J = 6.6$ Hz), 1.41–1.34 (m, 2H), 0.87 (d, 6H, $J = 6.6$ Hz); MS (ESI^+) m/e 381.46 (M+H); MS (ESI^-) m/e 379.37 (M−1); Anal. Calcd for $C_{18}H_{23}F_3N_6$: C, 56.83; H, 6.09; N, 22.09. Found: C, 56.80; H, 6.00; N, 22.12.

5.3.1.24. *N*-(4-{[2-(1-Cyclohexen-1-yl)ethyl]amino}-6-methyl-2-pyrimidinyl)-*N*′-[3-(trifluoromethyl)phenyl]guanidine (28). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 63% (200 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.50 (br s, 1H), 7.70 (br s, 1H), 7.45–7.17 (m, 5H), 5.87 (s, 1H), 5.40 (s, 1H), 3.25 (m, 2H), 2.11 (s, 3H), 2.11–2.09 (m, 2H), 1.95–1.87 (m, 4H), 1.55–1.46 (m, 4H); MS (ESI^+) m/e 419.43 (M+H); MS (ESI^-) m/e 417.30 (M−1); Anal. Calcd for $C_{21}H_{25}F_3N_6$: C, 60.28; H, 6.02; N, 20.08. Found: C, 60.23; H, 5.99; N, 20.10.

5.3.1.25. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N*′-[3-(trifluoromethyl)phenyl]guanidine (29). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 164–166 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.45–7.17 (m, 9H), 5.88 (s, 1H), 3.41 (br s, 2H), 2.80 (t, 2H, $J = 8$ Hz), 2.11 (s, 3H); MS (ESI^+) m/e 415 (M+H); Anal. Calcd for $C_{21}H_{21}F_3N_6$: C, 60.86; H, 5.11; N, 20.28. Found: C, 60.93; H, 5.06; N, 20.18.

5.3.1.26. *N*-(4-Chlorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (30). The resulted precipitate was washed with cold water and air dried to produce the desired product. ^1H NMR (DMSO-d_6, 300 MHz) δ 7.40–7.10 (m, 9H), 5.88 (s, 1H), 3.41 (br s, 2H), 2.80 (t, 2H, $J = 7.8$ Hz), 2.12 (s, 3H); HRMS calcd for $C_{20}H_{21}ClN_6$ (M+H) 381.1594, found 381.1614.

5.3.1.27. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N*′-[4-(trifluoromethyl)phenyl]guanidine (31). The resulted precipitate was washed with cold water and air dried and it was further recrystalized from isopropanol to produce the desired product in 71% yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.60 (br s, 1H), 7.80 (br s, 1H), 7.53 (d, 2H, $J = 9$ Hz), 7.39–6.98 (m, 9H), 5.89 (s, 1H), 3.41 (s, 2H), 2.80 (t, 2H, $J = 8$ Hz), 2.12 (s, 3H); HRMS calcd for $C_{21}H_{21}F_3N_6$ (M+H) 414.4329, found 415.1856; Anal. Calcd for $C_{21}H_{21}F_3N_6$ ($0.1H_2O$): C, 60.60; H, 5.13; N, 20.19. Found: C, 60.65; H, 5.18; N, 20.01.

5.3.1.28. *N*-(4-{[2-(4-Fluorophenyl)ethyl]amino}-6-methyl-2-pyrimidinyl)-*N*′-[4-(trifluoromethyl)phenyl]guanidine (32). Mp 164–166 °C; ^1H NMR (DMSO-d_6, 400 MHz) δ 7.45–7.06 (m, 8H), 5.87 (s, 1H), 3.39 (br s, 2H), 2.78 (t, 2H, $J = 8$ Hz), 2.10 (s, 3H); HRMS calcd for $C_{21}H_{20}F_4N_6$ (M+H) 433.1764, found 433.1768.

5.3.1.29. *N*-(4-{[2-(4-Nitrophenyl)ethyl]amino}-6-methyl-2-pyrimidinyl)-*N*′-[3-(trifluoromethyl)phenyl]guanidine (33). The crude product was washed with water and air dried to produce the desired product in 73% yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.75 (br s, 1H), 8.13 and 7.54 (AB, 4H, $J = 4$, 9 Hz), 5.88 (s, 1H), 3.48 (br s, 2H), 2.95 (t, 2H, $J = 7.5$ Hz), 2.11 (s, 3H); MS (ESI^+) m/e 460 (M+H). HRMS calcd for $C_{21}H_{20}F_3N_7O_2$ (M+H) 460.1709, found 460.1719. Anal. Calcd for $C_{21}H_{20}F_3N_7O_2$: C, 60.63; H, 5.31; N, 21.52. Found: C, 54.82; H, 4.37; N, 21.12.

5.3.1.30. *N*-(4-{[2-(4-Hydroxyphenyl)ethyl]amino}-6-methyl-2-pyrimidinyl)-*N*′-[3-(trifluoromethyl)phenyl]guanidine (34). The reaction mixture was cooled and H_2O was added to the reaction mixture. The reaction mixture was extracted with ethyl acetate and the combined organic layer were dried over Na_2SO_4 and concentrated to give pale yellow syrup. The crude reaction mixture was purified by liquid chromatography to produce the desired product in 37% yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 11.00 (br s, 1H), 10.57 (br s, 1H), 8.59 (br s, 1H), 7.81–7.61 (m, 6H), 7.29 and 7.22 (AB, $J = 4$, 9 Hz), 6.79 (s, 1H), 2.99 (br s, 2H), 2.77 (t, 2H, $J = 9$ Hz), 2.45 (s, 3H); HRMS calcd for $C_{21}H_{21}F_3N_6O$ (M+H) 430.4319, found 431.1809; Anal. Calcd for $C_{27}H_{25}F_3N_6O{\cdot}2TFA$ ($1.25H_2O$): C, 44.09; H, 3.77; N, 12.34. Found: C, 44.07; H, 3.62; N, 12.55.

5.3.1.31. *N*-4-(2-{[2-({Imino[3-(trifluoromethyl)anilino]methyl}amino)-6-methyl-4-pyrimidinyl]amino}ethyl)-benzenesulfonamide (35). The crude product was washed with water and air dried to produce the desired product. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 7.75 and 7.44 (AB, 4H, $J = 12$, 6 Hz), 7.41–7.18 (m, 6H), 5.88 (s, 1H), 3.31 (br s, 2H), 2.88 (t, 2H, $J = 6$ Hz), 2.11 (s, 3H); MS (ESI$^+$) m/e 494 (M+H); HRMS calcd for C21H22F3N7O2S (M+H) 494.1586, found 494.1592. Anal. Calcd for $C_{21}H_{22}F_3N_7O_2S$: C, 54.90; H, 4.39; N, 21.34. Found: C, 51.14; H, 4.48; N, 19.85.

5.3.1.32. *N*-(4-Methyl-6-{[2-(4-phenoxyphenyl)ethyl]-amino}-2-pyrimidinyl)-*N*′-[3-(trifluoromethyl)phenyl]guanidine (36). The resulted precipitate was washed with cold water and air dried to produce the desired product in 66% yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.20 (br s, 1H), 8.00 (br s, 1H), 7.46–6.83 (m, 14H), 5.89 (s, 1H), 3.42–3.32 (m, 2H), 2.78 (t, 2H, $J = 6$ Hz), 2.11 (s, 3H); HRMS calcd for $C_{27}H_{25}F_3N_6O$ (M+H) 506.5295, found 507.2018; Anal. Calcd for $C_{27}H_{25}F_3N_6O$ (0.1H_2O): C, 63.80; H, 5.00; N, 16.53. Found: C, 63.85; H, 5.02; N, 16.42.

5.3.1.33. *N*-(4-Methyl-6-{[2-(2-pyridinyl)ethyl]amino}-2-pyrimidinyl)-*N*′-[3-(trifluoromethyl)phenyl]guanidine (37). The resulted precipitate was washed with cold water and air dried to produce the desired product in 53% yield. ^{1}H NMR (CD_3OD, 400 MHz) δ 7.70–7.02 (m, 8H), 5.89 (s, 1H), 3.53 (br s, 2H), 3.28 (s, 1H), 2.84 (t, 2H, $J = 8$ MHz), 2.21 (s, 3H); HRMS calcd for $C_{20}H_{20}F_3N_7$ (M+H) 416.1811, found 416.1799.

5.3.1.34. *N*-(4-Chlorophenyl)-*N*′-(4-methyl-6-{[2-(4-morpholinyl)ethyl]amino}-2-pyrimidinyl)guanidine (38). The crude product was washed with water and air dried to produce an amorphous solid. This material was triturated with hot EtOH, cooled, and the supernatant decanted off to give the desired product in 5% (35 mg) yield. ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.27 (d, 2H, $J = 8.1$ Hz), 6.95 (d, 2H, $J = 8.1$ Hz), 5.81 (s, 1H), 3.71 (t, 4H, $J = 4.8$ Hz), 3.35–3.25 (m, 2H), 2.53–2.45 (m, 2H), 2.45–2.37 (m, 4H), 2.15 (s, 3H); MS (ESI$^+$) m/e 390.1 (M+H); HRMS calcd for $C_{18}H_{24}N_7OCl$ (M+H) 390.1809, found 390.1815.

5.3.1.35. *N*-(4-Methyl-6-{[2-(2-thienyl)ethyl]amino}-2-pyrimidinyl)-*N*′-[3-(trifluoromethyl)phenyl]guanidine (39). The crude product was washed with water and air dried to produce the desired product in 72% (230 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.60 (br s, 1H), 7.70 (br s, 1H), 7.46–7.18 (m, 6H), 6.95–5.89 (m, 2H), 3.44–3.38 (m, 2H), 3.02 (t, 2H, $J = 7.0$ Hz), 2.12 (s, 3H); MS (ESI$^+$) m/e 421.34 (M+H); MS (ESI$^-$) m/e 419.18 (M−1); Anal. Calcd for $C_{19}H_{19}F_3N_6S$: C, 54.28; H, 4.55; N, 19.99. Found: C, 54.41; H, 4.56; N, 20.06.

5.3.1.36. *N*-(4-Anilino-6-methyl-2-pyrimidinyl)-*N*′-(4-chlorophenyl)guanidine (40). The crude product was washed with water and air dried to produce the desired product in 98% (590 mg) yield. ^{1}H NMR (CD_3OD, 300 MHz) δ 7.42–7.38 (m, 2H), 7.35–7.26 (m, 5H), 7.14–7.09 (m, 2H), 6.18 (s, 1H), 2.25 (s, 3H); MS (ESI$^+$) m/e 353.0, 355.0 (M+H); HRMS calcd for $C_{18}H_{17}N_6Cl$ (M+H) 353.1301, found 353.1281.

5.3.1.37. *N*-[4-(Benzylamino)-6-methyl-2-pyrimidinyl]-*N*′-(4-chlorophenyl)guanidine (41). The crude product was washed with water and air dried to produce the desired product in 56% (350 mg) yield. ^{1}H NMR (CD_3OD, 300 MHz) δ 7.35–7.23 (m, 7H), 7.11–7.07 (d, 2H, $J = 8.7$ Hz), 5.99 (s, 1H), 4.50 (s, 2H), 2.19 (s, 3H); MS (ESI$^+$) m/e 367.1, 369.1 (M+H); Anal. Calcd for $C_{19}H_{19}ClN_6$ 0.5 H_2O: C, 60.72; H, 5.36; N, 22.36. Found: C, 60.85; H, 5.18; N, 22.71.

5.3.1.38. *N*-{4-Methyl-6-[(3-phenylpropyl)amino]-2-pyrimidinyl}-*N*′-[3-(trifluoromethyl)phenyl]guanidine (42). The resulted precipitate was washed with cold water and air dried to produce the desired product in 65% yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.40 (s, 2H), 7.45 (d, 2H, $J = 8$ Hz), 7.20 (br s, 1H), 5.81 (s, 1H), 5.86 (s, 1H), 2.09 (s, 3H), 1.45–1.40 (m, 2H), 1.22–1.20 (m, 12H), 0.80 (t, 3H, $J = 8$ Hz); HRMS calcd for $C_{22}H_{23}F_3N_6$ (M+H) 429.2014, found 429.2011; Anal. Calcd for $C_{27}H_{25}F_3N_6O$ (0.05H_2O): C, 61.54; H, 5.42; N, 19.57. Found: C, 61.54; H, 5.35; N, 19.58.

5.3.1.39. *N*-{4-Methyl-6-[(2-phenylethyl)sulfanyl]-2-pyrimidinyl}-*N*′-[4-(trifluoromethyl)phenyl]guanidine (43). The resulted precipitate was washed with cold water and air dried to produce the desired product in 42% yield. ^{1}H NMR (DMSO-d_6, 400 MHz) δ 9.20 (br s, 1H), 7.90 (br s, 1H), 7.64–7.15 (m, 9H), 6.71 (s, 1H), 3.31 (t, 3H, $J = 8$ Hz), 2.91 (t, 2H, $J = 8$ Hz), 2.24 (s, 3H); HRMS calcd for $C_{21}H_{20}F_3N_5S$ (M+H) 432.1469, found 432.1454. Anal. Calcd for $C_{21}H_{20}F_3N_5S$: C, 58.46; H, 4.67; N, 16.23. Found: C, 58.74; H, 4.90; N, 16.02.

5.3.1.40. *N*-[4-Methyl-6-(2-phenylethoxy)-2-pyrimidinyl]-*N*′-[3-(trifluoromethyl)phenyl]guanidine (44). The appropriate chloride of intermediate 3 (250 mg, 0.76 mmol) was treated with 7 mL phenethyl alcohol and K_2CO_3 (210 mg, 1.51 mmol) at 75 °C for 16 h. The reaction mixture was concentrated and purified by flash chromatography on silica gel with gradient elution from 20% to 50% EtOAc in hexanes. Appropriate fractions were combined and concentrated to a white solid. The solid was triturated with hexanes, filtered, and the filtrate concentrated to give the desired product as a yellow foam in 19% (61 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 9.15 (br s, 1H), 7.80 (br s, 1H), 7.48–7.42 (m, 1H), 7.27–7.16 (m, 8H), 6.17 (s, 1H), 4.42 (t, 2H, $J = 7.0$ Hz), 2.97 (t, 2H, $J = 7.0$ Hz), 2.22 (s, 3H); MS (ESI$^+$) m/e 415.89 (M+H); HRMS calcd for $C_{21}H_{20}N_5OF_3$ (M+H) 416.1698, found 416.1709.

5.3.1.41. *N*-(4-Chlorophenyl)-*N*′-[4-(diethylamino)-6-methyl-2-pyrimidinyl]guanidine (45). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, and pumped dry to produce the desired product in 48% (274 mg) yield. ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.34 (d, 2H, $J = 8.7$ Hz),

7.10 (d, 2H, $J = 8.7$ Hz), 5.91 (s, 1H), 3.44 (q, 4H, $J = 6.8$ Hz), 2.24 (s, 3H), 1.23–1.07 (t, 6H, $J = 7.1$ Hz); MS (ESI^+) m/e 333.1, 335.1 (M+H); HRMS calcd for $C_{16}H_{21}N_6Cl$ (M+H) 333.1594, found 333.1596.

5.3.1.42. *N*-(4-Chlorophenyl)-*N′*-{4-methyl-6-[methyl-(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (46). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, and pumped dry to produce the desired product in 47% (316 mg) yield. ^1H NMR ($CDCl_3$, 300 MHz) δ 7.35–7.20 (m, 5H), 7.18 (d, 2H, $J = 8.4$ Hz), 6.96 (d, 2H, $J = 8.4$ Hz), 5.85 (s, 1H), 3.72–3.64 (m, 2H), 2.91 (s, 3H), 2.85 (t, 2H, $J = 7.4$ Hz), 2.24 (s, 3H); MS (ESI^+) m/e 395.1, 397.1 (M+H); Anal. Calcd for $C_{21}H_{23}ClN_6$: C, 63.87; H, 5.87; N, 20.87. Found: C, 63.50; H, 5.88; N, 20.92.

5.3.1.43. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-phenylguanidine (47). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, and pumped dry to produce the desired product in 88% (520 mg) yield. ^1H NMR ($CDCl_3$, 300 MHz) δ 8.65 (br s, 1H), 8.10 (br s, 1H), 7.45–7.00 (m, 10H), 5.95 (s, 1H), 3.48 (m, 2H), 2.82 (t, 2H, $J = 7.4$ Hz), 2.16 (s, 3H); MS (ESI^+) m/e 347.1 (M+H); HRMS calcd for $C_{20}H_{22}N_6$ (M+H) 347.1984, found 347.1974.

5.3.1.44. *N*-(2-Fluorophenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (48). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 173–175 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.38–6.86 (m, 9H), 5.88 (s, 1H), 3.39 (br s, 2H), 2.78 (t, 2H, $J = 8$ Hz), 2.24 (s, 3H); MS (ESI^+) m/e 365 (M+H); MS (ESI^+) m/e 456 (M+H); Anal. Calcd for $C_{20}H_{21}FN_6$ ($0.05H_2O$): C, 65.75; H, 5.82; N, 23.00. Found: C, 65.72; H, 5.89; N, 22.89.

5.3.1.45. *N*-(3-Fluorophenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (49). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 186–188 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.30–7.16 (m, 8H), 6.67 (m, 1H), 5.87 (s, 1H), 3.40 (br s, 2H), 2.79 (t, 2H, $J = 7$ Hz), 2.11 (s, 3H); MS (ESI^+) m/e 365 (M+H); Anal. Calcd for $C_{20}H_{21}FN_6$: C, 65.92; H, 5.81; N, 23.06. Found: C, 65.78; H, 5.93; N, 22.83.

5.3.1.46. *N*-(4-Fluorophenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (50). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, and pumped dry to produce the desired product in 56% (350 mg) yield. ^1H NMR ($CDCl_3$, 300 MHz) δ 8.10 (br s, 1H), 7.45–7.10 (m, 9H), 5.85 (s, 1H), 3.48 (m, 2H), 2.82 (t, 2H, $J = 7.4$ Hz), 2.16 (s, 3H); MS (ESI^+) m/e 365.1 (M+H); HRMS calcd for $C_{20}H_{21}N_6F$ (M+H) 365.1890, found 365.1878.

5.3.1.47. *N*-(2-Chlorophenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (51). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 181–183 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.44–6.88 (m, 9H), 5.88 (s, 1H), 3.39 (s, 2H), 2.78 (t, 2H, $J = 7$ Hz), 2.10 (s, 3H); HRMS calcd for $C_{20}H_{21}N_6Cl$ (M+H) 381.1594, found 381.1591.

5.3.1.48. *N*-(3-Chlorophenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (52). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 205–207 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.30–7.16 (m, 9H), 6.90 (m, 1H), 5.87 (s, 1H), 3.40 (br s, 2H), 2.79 (t, 2H, $J = 7$ Hz), 2.10 (s, 3H); HRMS calcd for $C_{21}H_{21}N_6Cl$ (M+H) 381.1594, found 381.1584.

5.3.1.49. *N*-(2-Methoxyphenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (53). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 144–146 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.27–7.18 (m, 5H), 6.96–6.85 (m, 4H), 5.87 (s, 1H), 3.77 (s, 3H), 3.41 (br s, 2H), 2.79 (t, 2H, $J = 7$ Hz), 2.11 (s, 3H); HRMS calcd for $C_{21}H_{24}N_6O$ (M+H) 377.2090, found 377.2090.

5.3.1.50. *N*-(3-Methoxyphenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (54). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 205–206 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.30–7.09 (m, 8H), 6.48 (dd, 1H, $J = 2$, 8 Hz), 5.86 (s, 1H), 3.69 (s, 3H), 3.40 (br s, 2H), 2.79 (t, 2H, $J = 8$ Hz), 2.10 (s, 3H); MS (ESI^+) m/e 377 (M+H). HRMS calcd for $C_{21}H_{24}N_6O$ (M+H) 377.2090, found 377.2105.

5.3.1.51. *N*-(4-Fluorophenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (55). The crude product was purified by flash chromatography on silica gel with gradient elution of 10–40% CH_3OH in EtOAc. Isolated fractions were combined, concentrated, and pumped dry to produce the desired product in 20% (130 mg) yield. ^1H NMR (CD3OD, 300 MHz) δ 7.30–7.17 (m, 5H), 7.05 (d, 2H, $J = 8.7$ Hz), 6.90 (d, 2H, $J = 8.7$ Hz), 5.87 (s, 1H), 3.78 (s, 3H), 3.51 (t, 2H, $J = 7.2$ Hz), 2.86 (t, 2H, $J = 7.2$ Hz), 2.16 (s, 3H); MS (ESI^+) m/e 377.1 (M+H); Anal. Calcd for $C_{21}H_{24}N_6O$: C, 67.00; H, 6.43; N, 22.43. Found: C, 66.81; H, 6.44; N, 22.49.

5.3.1.52. *N*-(2-Methylphenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (56). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 185–187 °C; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.32–7.05 (m, 9H), 6.88–6.83 (m, 1H), 5.85 (s, 1H), 3.38 (br s, 2H), 2.77 (t, 3H, $J = 8$ Hz), 2.10–2.08 (m, 6H); MS (ESI^+) m/e 361 (M+H); Anal. Calcd for $C_{23}H_{26}N_2O_6$: C, 66.01; H, 6.26; N, 20.08. Found: C, 65.74; H, 6.26; N, 19.88.

5.3.1.53. *N*-(3-Methylphenyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (57). The resulted

precipitate was washed with cold water and air dried to produce the desired product. Mp 184–185 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 7.30–7.08 (m, 8H), 6.95 (br s, 1H), 6.72 (d, 1H, $J = 7$ Hz), 5.85 (s, 1H), 3.30 (br s, 2H), 2.79 (t, 2H $J = 8$ Hz), 2.24 (s, 3H,), 2.10 (s, 3H); MS (ESI$^+$) m/e 361 (M+H); Anal. Calcd for $C_{21}H_{24}N_6$ (0.4H_2O): C, 68.84; H, 6.66; N, 22.84. Found: C, 68.84; H, 6.78; N, 22.94.

5.3.1.54. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N*′-[2-(trifluoromethyl)phenyl]guanidine (58). The resulted precipitate was washed with cold water and air dried to produce the desired product. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.30 (br s, 1H), 7.56–6.98 (m, 10H), 5.88 (s, 1H), 3.40 (br s, 2H), 2.78 (t, 2H, $J = 7.5$ Hz), 2.09 (s, 3H); MS (ESI$^+$) m/e 415 (M+H); Anal. Calcd for $C_{21}H_{21}F_3N_6$ (0.75H_2O): C, 58.94; H, 5.30; N, 19.64. Found: C, 58.79; H, 5.01; N, 19.42.

5.3.1.55. *N*-(2,3-Difluorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (59). Dioxane was use in place of CH_3OH as reaction solvent. DIEA was used in place of K_2CO_3 as base. Reaction mix heated at 75 °C for 16 h. The crude product was washed with water and air dried to produce the desired product in 68% (216 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.70 (br s, 1H), 7.42 (br s, 1H), 7.27–7.20 (m, 5H), 7.15 (m, 1H), 6.96 (m, 1H), 6.83 (m, 1H), 6.70 (br s, 1H), 5.86 (s, 1H), 3.45–3.32 (m, 2H), 2.76 (t, 2H, $J = 7.4$ Hz), 2.08 (s, 3H); MS (ESI$^+$) m/e 382.9 (M+H); Anal. Calcd for $C_{20}H_{20}F_2N_6$: C, 62.82; H, 5.25; N, 21.89. Found: C, 62.45; H, 5.34; N, 22.02.

5.3.1.56. *N*-(2,4-Difluorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (60). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 83% (270 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.55 (br s, 1H), 7.40 (br s, 1H), 7.32–6.85 (m, 8H), 5.88 (s, 1H), 3.47–3.30 (m, 2H), 2.78 (t, 2H, $J = 7.3$ Hz), 2.10 (s, 3H); MS (ESI$^+$) m/e 382.9 (M+H); Anal. Calcd for $C_{20}H_{20}F_2N_6$: C, 62.82; H, 5.27; N, 21.98. Found: C, 62.81; H, 5.28; N, 22.00.

5.3.1.57. *N*-(2,5-Difluorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (61). Dioxane was use in place of CH_3OH as reaction solvent. DIEA was used in place of K_2CO_3 as base. Reaction mix heated at 75 °C for 16 h. The crude product was washed with water and air dried to produce the desired product in 68% (219 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.67 (br s, 1H), 7.42 (br s, 1H), 7.25–6.65 (m, 8H), 5.86 (s, 1H), 3.42–3.30 (m, 2H), 2.77 (t, 2H, $J = 7.4$ Hz), 2.08 (s, 3H); MS (ESI$^+$) m/e 382.9 (M+H); HRMS calcd for $C_{20}H_{20}N_6F_2$ (M+H) 383.1796, found 383.1788.

5.3.1.58. *N*-(2,6-Difluorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (62). CH_3CN was used in place of CH_3OH as reaction solvent and heated 16 h at 75 °C. The crude product was washed with water and air dried to produce the desired product in 59% (190 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.70 (br s, 1H), 7.45 (br s, 1H), 7.33–7.12 (m, 5H), 7.00–6.84 (m, 3H), 5.89 (s, 1H), 3.48–3.33 (m, 2H), 2.79 (t, 2H, $J = 7.1$ Hz), 2.11 (s, 3H); MS (ESI$^+$) m/e 382.9 (M+H); Anal. Calcd for $C_{20}H_{20}F_2N_6$ 0.65HCl: C, 59.15; H, 5.13; N, 20.69. Found: C, 59.26; H, 5.00; N, 20.79.

5.3.1.59. *N*-(3,4-Difluorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (63). Dioxane was use in place of CH_3OH as reaction solvent. DIEA was used in place of K_2CO_3 as base. Reaction mix heated at 75 °C for 16 h. The crude product was washed with water and air dried to produce the desired product in 64% (208 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.50 (br s, 1H), 7.35 (br s, 1H), 7.28–7.12 (m, 8H), 6.70 (br s, 1H), 5.84 (s, 1H), 3.42–3.32 (m, 2H), 2.76 (t, 2H, $J = 7.4$ Hz), 2.08 (s, 3H); MS (ESI$^+$) m/e 382.9 (M+H); HRMS calcd for $C_{20}H_{20}N_6F_2$ (M+H) 383.1796, found 383.1790.

5.3.1.60. *N*-(3,5-Difluorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (64). Dioxane was use in place of CH_3OH as reaction solvent. DIEA was used in place of K_2CO_3 as base. Reaction mix heated at 75 °C for 16 h. The crude product was washed with water and air dried to produce the desired product in 39% (126 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.60 (br s, 1H), 7.40 (br s, 1H), 7.28–7.18 (m, 5H), 7.18–7.10 (m, 2H), 6.65–6.57 (m, 1H), 6.45 (br s, 1H), 5.85 (s, 1H), 3.45–3.30 (m, 2H), 2.77 (t, 2H, $J = 7.4$ Hz), 2.09 (s, 3H); MS (ESI$^+$) m/e 382.9 (M+H); HRMS calcd for $C_{20}H_{20}N_6F_2$ (M+H) 383.1796, found 383.1795.

5.3.1.61. *N*-(3-Nitrophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (65). The resulted precipitate was washed with cold water and air dried to produce the desired product. Mp 180–181 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 7.72 (d, 1H, $J = 2$ Hz), 7.68 (t, 1H, $J = 2$ Hz), 7.50–7.15 (m, 7H), 5.89 (s, 1H), 3.31 (br s, 2H), 2.80 (t, 2H, $J = 8$ Hz), 2.12 (s, 3H); MS (ESI$^+$) m/e 392 (M+H); Anal. Calcd for $C_{21}H_{24}N_6$: C, 61.37; H, 5.41; N, 25.05. Found: C, 61.65; H, 5.45; N, 24.78.

5.3.1.62. *N*-Benzyl-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}guanidine (66). The resulted precipitate was washed with cold water and air dried to produce the desired product. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 9.62 (br s, 1H), 8.42 (br s, 1H), 7.96 (br s, 1H), 7.40–7.16 (m, 10H), 6.20 (br s, 1H), 5.99 (s, 1H), 4.53 (d, 2H, $J = 5.4$ H), 3.39 (br s, 2H), 2.75 (br s, 2H), 2.13 (s, 3H); MS (ESI$^+$) m/e 361 (M+H); Anal. Calcd for $C_{21}H_{24}N_6$·2TFA (1.4H_2O): C, 48.93; H, 4.73; N, 13.69. Found: C, 48.90; H, 4.43; N, 13.61.

5.3.2. General procedure B, for synthesis of compounds of type 8 (68–69, Scheme 2). To a solution of compound **6** (11.6 g, 51.9 mmol) in dioxane (100 mL) was added methylisothiocyanate (8.9 g, 122 mmol) and heated at reflux for 48 h. The solvent was evaporated in vacuo, the residue triturated with Et_2O (250 mL), filtered, and air

dried to give the corresponding thiourea in 79% (41.1 mmol) yield.

The thiourea (12 g, 40.0 mmol) was dissolved in acetone (200 mL) and treated with iodomethane (17 g, 120 mmol) at ambient temperature for 16 h. The reaction mixture was diluted with EtOAc (100 mL), cooled in ice, and filtered to give a hygroscopic white solid. This material was partitioned between EtOAc/satd $NaHCO_3$, the organic phase separated, dried over $MgSO_4$, filtered, and filtrate concentrated to give the intermediate isothiourea as a white foam in 42% (5.29 g) yield. The isothiourea (500 mg, 1.59 mmol) was dissolved in dioxane (5 mL) and treated with the corresponding aminopyridine (225 mg, 2.38 mmol) at 100 °C for 16 h. The reaction mixtures were concentrated and title compounds purified by flash chromatography on silica gel with gradient elution from 0% to 7% CH_3OH in EtOAc with 1% TEA.

5.3.2.1. *N*″-[(*E*)-Methyl]-*N*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N*′-(2-pyridinyl)guanidine (68). Desired product obtained in 57% (332 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 9.42 (s, 1H), 8.12 (s, 1H), 7.62–7.52 (m, 2H), 7.32–7.15 (m, 6H), 6.82–6.70 (m, 2H), 5.93 (s, 1H), 3.50–3.30 (m, 2H), 2.88 (s, 3H), 2.85–2.75 (m, 2H), 2.14 (s, 3H); MS (ESI$^+$) m/e 361.98 (M+H); Anal. Calcd for $C_{20}H_{23}N_7$ (0.1EtOAc): C, 66.18; H, 6.48; N, 26.48. Found: C, 66.41; H, 6.41; N, 26.66.

5.3.2.2. *N*″-[(*E*)-Methyl]-*N*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N*′-(3-pyridinyl)guanidine (69). Desired product obtained in 37% (214 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.80 (s, 1H), 8.11 (s, 1H), 8.03 (s, 1H), 7.55–7.40 (m, 1H), 7.30–7.10 (m, 8H), 6.00–5.85 (m, 1H), 3.42–3.30 (m, 2H), 2.81 (s, 3H), 2.80–2.72 (m, 2H), 2.07 (s, 3H); MS (ESI$^+$) m/e 361.91 (M+H); Anal. Calcd for $C_{20}H_{23}N_7$ (0.4CH_3OH): C, 65.47; H, 6.63; N, 26.20. Found: C, 65.37; H, 6.44; N, 26.50.

5.3.2.3. N^2-(1*H*-Benzimidazol-2-yl)-6-methyl-N^4-(2-phenylethyl)-2,4-pyrimidinediamine (70). 2-Guanidino benzimidazole (4.16 g, 23.7 mmol) and methyl acetoacetate (3.08 mL, 28.6 mmol) were combined in MeOH (15 mL) followed by dropwise addition of NaOMe (1.28 g in 15 mL MeOH, 23.8 mmol). The reaction mixture was heated at reflux for 2 h during which time a precipitate forms. The reaction was cooled, filtered, and washed with cold MeOH to give the sodium salt. This material was dissolved in water and glacial acetic acid added until solution reached pH 5. A white precipitate formed and was filtered, washed with water, and dried in vacuum oven to obtain the desired product in 80% (4.56 g) yield. MS (ESI$^+$) m/e 242.1 (M+H). This intermediate (1.15 g, 4.44 mmol) was dissolved in $POC1_3$ (10 mL) and stirred at room temperature overnight. The $POC1_3$ was removed in vacuo and crushed ice added to the residue. After the remaining $POC1_3$ was consumed, a white solid formed, which was broken up with a spatula. A stir bar was added and the chunky solid was converted to a fine crystal with vigorous stirring. The solid was filtered and washed with water to remove any remaining acid. Upon drying, the chloride was obtained in 62% (700 mg) yield. MS (ESI$^+$) m/e 260.0, 262.0 (M+H). Phenethylamine (762 mg, 6.3 mmol) and the chloride (220 mg, 0.84 mmol) were dissolved in dioxane (7 mL) with K_2CO_3 (232 mg, 1.68 mmol) and the reaction was heated at reflux for 36 h. The reaction mixture was concentrated to small volume and H_2O added to incipient cloudiness. The resultant precipitate was filtered, washed with water, and air dried to give the desired product in 62% (180 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 10.37 (s, 1H), 7.45 (s, 1H), 7.38–7.23 (m, 6H), 7.19 (m, 1H), 7.02–6.96 (m, 2H), 6.05–5.90 (m, 1H), 3.57–3.35 (m, 2H), 2.88–2.80 (m, 2H), 2.22 (s, 3H); MS (ESI$^+$) m/e 344.86 (M+H); Anal. Calcd for $C_{20}H_{20}N_6$ (0.2H_2O): C, 69.02; H, 5.91; N, 24.15. Found: C, 69.02; H, 5.82; N, 24.24.

5.3.3. General procedure C, for synthesis of compounds of type 7 (67, 71–85, Scheme 2)

5.3.3.1. 6-Methyl-N^4-(2-phenylethyl)-2,4-pyrimidinediamine (6). To a solution of 2-amino-4-chloro-6-methylpyrimidine (10.0g, 70 mmol) in EtOH (270 mL) was added sodium acetate (11.4 g, 139 mmol) and phenethylamine (11.0 g, 90 mmol). The reaction mixture was heated at reflux for 8 h, filtered, and concentrated in vacuo. The residue was partitioned between EtOAc/H_2O, the organic phase separated, washed with H_2O, dried over $MgSO_4$, filtered, and concentrated to a white solid. This material was triturated with Et_2O, filtered, washed 2×Et_2O, and air dried to give pure product, **6**, in 54% (8.58 g) yield.

5.3.3.2. *N*-Alkyl-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N*″-[4-(trifluoromethyl)phenyl]guanidine (7). Compound **6** (5.6 g, 24.6 mmol) was dissolved in dioxane (100 mL) and treated with 4-(trifluoromethyl)phenyl isothiocyanate (5 g, 24.6 mmol) at reflux for 20 h. The reaction mixture was concentrated to a yellow solid that was recrystallized from EtOH (70 mL). The reaction flask was cooled in ice, filtered, and the cake washed with cold EtOH (3×25 mL) to give the resultant thiourea in 51% (5.4 g) yield. MS (ESI$^+$) m/e 432.83 (M+H). The thiourea (4.05 g, 9.38 mmol) was dissolved in acetone (16 mL) and treated with iodomethane (4.0 g, 28.2 mmol) at ambient temperature for 16 h. The resultant precipitate was filtered off and washed with Et_2O. A second crop was obtained by concentrating the filtrate to small volume, diluting with EtOAc, and filtering. The combined materials gave the isothiourea intermediate in 91% (4.9 g) yield as the HI salt. MS (ESI$^+$) m/e 446.08 (M+H). Final products of type **7** were typically prepared by dissolving the above material in EtOH (0.5 M) and treating with the appropriate amine (1.5 equiv) at 60 °C for 16 h. Reaction mixtures were diluted with EtOH (3×original volume) and H_2O was added to incipient cloudiness. The resultant precipitates were cooled in ice, filtered, washed with H_2O, and air dried to give the title compounds below. In cases where unsatisfactory results were obtained by this

method, products were isolated by flash chromatography as detailed individually. Variations from this procedure are also detailed individually.

5.3.3.3. *N*-Methyl-*N′*-{4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)phenyl]-guanidine (67). A 2 M solution of methylamine in THF (8.7 mL, 17.4 mmol) was used in place of EtOH as reaction solvent with the isothiourea (1.0 g, 1.74 mmol) at reflux for 1 h. The reaction mixture was concentrated, partitioned between EtOAc/H_2O, the organic phase separated, washed with satd $NaHCO_3$ and brine, dried over $MgSO_4$, filtered, and concentrated. The residue was crystalized from EtOH/H_2O as described above to give the desired product in 74% (550 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.92 (br s, 1H), 7.58 (d, 2H, $J = 8.3$ Hz), 7.60–7.45 (br s, 1H), 7.30–7.13 (m, 5H), 7.02–6.80 (m, 2H), 5.92 (s, 1H), 3.47–3.30 (m, 2H), 2.85 (d, 3H, $J = 4.4$ Hz), 2.79 (t, 2H, $J = 7.3$ Hz), 2.11 (s, 3H); MS (ESI$^+$) m/e 429.0 (M+H); MS (ESI$^-$) m/e 426.49 (M−1); Anal. Calcd for $C_{22}H_{23}F_3N_6$: C, 61.67; H, 5.41; N, 19.61. Found: C, 61.69; H, 5.47; N, 19.40.

5.3.3.4. *N*-Ethyl-*N′*-{4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)phenyl]guanidine (71). A 2 M solution of ethylamine in THF (8.7 mL, 17.4 mmol) was used in place of EtOH as reaction solvent with the isothiourea (1.0 g, 1.74 mmol) at ambient temperature for 4 days. The reaction mixture was concentrated, partitioned between EtOAc/H_2O, the organic phase separated, washed with satd $NaHCO_3$ and brine, dried over $MgSO_4$, filtered, and concentrated. The residue was crystalized from EtOH/H_2O as described above to give the desired product in 87% (666 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.98 (s, 1H), 7.58 (d, 2H, $J = 8.5$ Hz), 7.60–7.45 (br s, 1H), 7.30–7.17 (m, 5H), 7.03–6.90 (m, 2H), 5.93 (s, 1H), 3.50–3.37 (m, 2H), 3.37–3.25 (m, 2H), 2.80 (t, 2H, $J = 7.3$ Hz), 2.10 (s, 3H), 1.21–1.08 (m, 3H); MS (ESI$^+$) m/e 443.05 (M+H); MS (ESI$^-$) m/e 440.85 (M−1); Anal. Calcd for $C_{23}H_{25}F_3N_6$ 0.27HI: C, 57.91; H, 5.34; N, 17.62. Found: C, 57.99; H, 5.43; N, 17.25.

5.3.3.5. *N*-Isopropyl-*N′*-{4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)phenyl]-guanidine (72). Desired product was obtained in 100% (310 mg) yield as per general procedure C. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.91 (d, 1H, $J = 6.3$ Hz), 7.54 (d, 2H, $J = 8.4$ Hz), 7.60–7.50 (br s, 1H), 7.22 (d, 2H, $J = 7.2$ Hz), 7.17 (d, 2H, $J = 7.5$ Hz), 7.24–7.10 (m, 1H), 6.94–6.89 (m, 2H), 5.88 (s, 1H), 3.99–3.90 (m, 1H), 3.45–3.30 (m, 2H), 2.80–2.76 (m, 2H), 2.04 (s, 3H), 1.12 (d, 6H, $J = 4.9$ Hz); MS (ESI$^+$) m/e 456.87 (M+H); Anal. Calcd for $C_{24}H_{27}F_3N_6$: C, 63.14; H, 5.96; N, 18.41. Found: C, 63.32; H, 5.95; N, 18.45.

5.3.3.6. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-propyl-*N″*-[4-(trifluoromethyl)phenyl]guanidine (73). Desired product was obtained in 70% (282 mg) yield as per general procedure C. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 9.05 (s, 1H), 7.58 (d, 2H, $J = 8.5$ Hz), 7.32–7.16 (m, 5H), 7.03–6.91 (m, 2H), 5.93 (s, 1H), 3.50–3.30 (m, 2H), 3.30–3.21 (m, 2H), 2.79 (t, 2H, $J = 7.4$ Hz), 2.10 (s, 3H), 1.62–1.50 (m, 2H), 0.90 (t, 3H, $J = 7.2$ Hz); MS (ESI$^+$) m/e 456.91 (M+H); MS (ESI$^-$) m/e 454.95 (M−1); Anal. Calcd for $C_{24}H_{27}F_3N_6$: C, 63.14; H, 5.96; N, 18.41. Found: C, 63.23; H, 5.98; N, 18.46.

5.3.3.7. *N*-(2-Hydroxyethyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)-phenyl]guanidine (74) . The *tert*-butyl ether of the title compound was prepared in 100% (450 mg) yield as per general procedure C. The ether (358 mg, 0.69 mmol) was subsequently treated with trifluoroacetic acid (3 mL) at ambient temperature for 3 h. The reaction mixture was concentrated and partitioned between EtOAc/satd $NaHCO_3$, the organic phase separated, dried over $MgSO_4$, filtered, and the filtrate concentrated. The residue was purified by flash chromatography on silica gel eluted with a gradient from 0% to 3% CH_3OH in EtOAc to give the desired product in 35% (110 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 9.23 (s, 1H), 7.59 (d, 2H, $J = 7.9$ Hz), 7.51 (br s, 1H), 7.31–7.13 (m, 5H), 7.01–6.92 (m, 2H), 5.92 (s, 1H), 4.85 (br s, 1H), 3.62–3.57 (m, 2H), 3.51–3.30 (m, 4H), 2.79 (t, 2H, $J = 6.9$ Hz), 2.10 (s, 3H); MS (ESI$^+$) m/e 458.95 (M+H); MS (ESI$^-$) m/e 456.97 (M−1); Anal. Calcd for $C_{23}H_{25}F_3N_6O$: C, 60.25; H, 5.50; N, 18.33. Found: C, 60.27; H, 5.44; N, 18.44.

5.3.3.8. *N*-Butyl-*N′*-{4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)phenyl]guanidine (75). Desired product was obtained in 73% (300 mg) yield as per general procedure C. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 9.05 (br s, 1H), 7.57 (d, 2H, $J = 8.2$ Hz), 7.60–7.55 (br s, 1H), 7.30–7.18 (m, 5H), 7.02–6.90 (m, 3H), 5.92 (s, 1H), 3.45–3.35 (m, 2H), 3.34–2.25 (m, 2H), 2.79 (t, 2H, $J = 7.4$ Hz), 2.10 (s, 3H), 1.60–1.43 (m, 2H), 1.42–1.29 (m, 2H), 0.88 (t, 3H, $J = 6.8$ Hz); MS (ESI$^+$) m/e 470.93 (M+H); MS (ESI$^-$) m/e 468.93 (M−1); Anal. Calcd for $C_{25}H_{29}F_3N_6$: C, 63.81; H, 6.21; N, 17.86. Found: C, 63.82; H, 6.16; N, 17.92.

5.3.3.9. *N*-(2-Methoxyethyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)-phenyl]guanidine (76). Desired product was obtained in 96% (200 mg) yield as per general procedure C. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 9.25 (br s, 1H), 7.65–7.50 (m, 3H), 7.22 (d, 2H, $J = 6.8$ Hz), 7.16 (d, 2H, $J = 7.2$ Hz), 7.26–7.14 (m, 1H), 7.00–6.90 (m, 2H), 5.88 (s, 1H), 3.57–3.40 (m, 6H), 3.18 (s, 3H), 2.82–2.73 (m, 2H), 2.08 (s, 3H); MS (ESI$^+$) m/e 476.00 (M+H); Anal. Calcd for $C_{24}H_{27}F_3N_6O$ 0.33HI: C, 56.00; H, 5.35; N, 16.33. Found: C, 55.98; H, 5.39; N, 16.05.

5.3.3.10. *N,N*-Dimethyl-*N′*-{4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)phenyl] guanidine (77). A 2 M solution of dimethylamine in CH_3OH (4.3 mL, 8.7 mmol) was used in place of EtOH as reaction solvent with the isothiourea (500 mg, 0.88 mmol) at 60 °C for 16 h. The reaction mixture was concentrated, partitioned between EtOAc/satd $NaHCO_3$, organic phase separated, dried over $MgSO_4$,

filtered, and concentrated. The residue was purified by flash chromatography on silica gel eluted with a gradient from 0% to 3% CH_3OH in EtOAc to give the desired product in 75% (295 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.30 (br s, 1H), 7.42–7.18 (m, 8H), 6.93–6.70 (m, 2H), 5.70 (s, 1H), 3.47–3.30 (m, 2H), 2.95 (s, 6H), 2.76 (t, 2H, $J = 7.1$ Hz), 2.02 (s, 3H); MS (ESI^+) m/e 442.97 (M+H); Anal. Calcd for $C_{23}H_{25}F_3N_6$ 0.30HI: C, 57.45; H, 5.30; N, 17.48. Found: C, 57.60; H, 5.41; N, 17.48.

5.3.3.11. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-[4-(trifluoromethyl)phenyl]-4-morpholine carboximidamide (78). Desired product was obtained in 61% (260 mg) yield as per general procedure C. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.40 (br s, 1H), 7.39 (d, 2H, J = 8.0Hz), 7.36–7.03 (m, 5H), 6.91–6.81 (m, 2H), 5.70 (s, 1H), 3.63–3.58 (m, 4H), 3.44–3.36 (m, 4H), 3.40–3.30 (m, 2H), 2.78 (t, 2H, $J = 7.4$ Hz), 2.01 (s, 3H); MS (ESI^+) m/e 484.90 (M+H); MS (ESI^-) m/e 482.90 (M−1); Anal. Calcd for $C_{25}H_{27}F_3N_6O$ 0.07HI: C, 60.85; H, 5.53; N, 17.03. Found: C, 60.82; H, 5.65; N, 17.01.

5.3.3.12. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-phenyl-*N″*-[4-(trifluoromethyl)phenyl]guanidine (79). Desired product was obtained as the HI salt in 99% (270 mg) yield as per general procedure C. ^1H NMR (DMSO-d_6, 300 MHz) δ 8.62 (br s, 1H), 7.63 (d, 2H, $J = 8.4$ Hz), 7.48 (d, 2H, $J = 8.6$ Hz), 7.40–7.23 (m, 5H), 7.21–7.08 (m, 4H), 6.98 (d, 2H, $J = 7.3$ Hz), 5.87 (s, 1H), 3.43–3.37 (m, 2H), 2.66 (t, 2H, $J = 7.4$ Hz), 2.14 (s, 3H); MS (ESI^+) m/e 491.24 (M+H); Anal. Calcd for $C_{27}H_{25}F_3N_6$ 1.0HI: C, 52.44; H, 4.24; N, 13.59. Found: C, 52.61; H, 4.29; N, 13.54.

5.3.3.13. *N*-(4-Methoxybenzyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)-phenyl]guanidine (80). Desired product was obtained as the HI salt in 81% (235 mg) yield as per general procedure C. ^1H NMR (DMSO-d_6, 300 MHz, 100C) δ 9.10 (br s, 1H), 7.59 (d, 2H, $J = 8.3$ Hz), 7.31 (d, 2H, $J = 8.6$ Hz), 7.25–7.11 (m, 6H), 6.88 (d, 2H, $J = 8.7$ Hz), 5.93 (s, 1H), 4.52 (s, 2H), 3.73 (s, 3H), 3.35 (m, 2H), 2.72 (t, 2H, $J = 7.1$ Hz), 2.12 (s, 3H); MS (ESI^+) m/e 535.05 (M+H); MS (ESI^-) m/e 532.99 (M−1); Anal. Calcd for $C_{29}H_{29}F_3N_6O$ 0.43HI: C, 59.08; H, 5.03; N, 14.25. Found: C, 59.12; H, 5.10; N, 14.39; HRMS calcd for $C_{29}H_{29}F_3N_6O$ (M+H) 535.2433, found 535.2424.

5.3.3.14. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-(2-phenylethyl)-*N″*-[4-(trifluoromethyl)phenyl]-guanidine (81). The reaction mixture was concentrated, partitioned between EtOAc/satd $NaHCO_3$, organic phase separated, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by flash chromatography on silica gel eluted with EtOAc to give the desired product in 67% (378 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 9.08 (s, 1H), 7.58 (d, 2H, $J = 8.4$ Hz), 7.60–7.45 (m, 1H), 7.37–7.10 (m, 9H), 7.03–6.90 (m, 2H), 5.90 (s, 1H), 3.63–3.52 (m, 2H), 3.35–3.25 (m, 2H), 2.97–2.83 (m, 2H), 2.83–2.70 (m, 2H), 2.02 (s, 3H); MS (ESI^+) m/e 518.99 (M+H); MS (ESI^-) m/e 516.95 (M−1); Anal. Calcd for $C_{29}H_{29}F_3N_6$ 0.79HI: C, 56.21; H, 4.85; N, 13.56. Found: C, 56.19; H, 4.98; N, 13.48.

5.3.3.15. *N*-(2-Furylmethyl)-*N′*-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N″*-[4-(trifluoromethyl)phenyl]-guanidine (82). Desired product was obtained in 69% (301 mg) yield as per general procedure C. ^1H NMR (DMSO-d_6, 300 MHz) δ 9.43 (br s, 1H), 7.63–7.50 (m, 4H), 7.30–6.93 (m, 8H), 6.39 (s, 1H), 6.33 (s, 1H), 5.91 (s, 1H), 4.51 (d, 2H, $J = 4.4$ Hz), 3.35–3.25 (m, 2H), 2.80–2.70 (m, 2H), 2.07 (s, 3H); MS (ESI^+) m/e 494.89 (M+H); MS (ESI^-) m/e 492.88 (M−1); Anal. Calcd for $C_{26}H_{25}F_3N_6O$: C, 63.15; H, 5.10; N, 16.99. Found: C, 63.21; H, 5.08; N, 16.91.

5.3.3.16. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-(4-pyridinylmethyl)-*N″*-[4-(trifluoromethyl)-phenyl]guanidine (83). Desired product was obtained in 27% (60 mg) yield as per general procedure C. ^1H NMR (DMSO-d_6, 300 MHz) δ 9.62–9.43 (m, 1H), 8.46 (d, 2H, $J = 5.8$ Hz), 7.53 (d, 2H, $J = 7.9$ Hz), 7.55–7.40 (m, 1H), 7.33 (d, 2H, $J = 5.5$ Hz), 7.15–7.00 (m, 6H), 6.92–6.85 (m, 2H), 6.00–5.90 (m, 1H), 4.55 (d, 2H, $J = 5.3$ Hz), 3.35–3.25 (m, 2H), 2.80–2.65 (m, 2H), 2.06 (s, 3H); MS (ESI^+) m/e 506.02 (M+H); MS (ESI^-) m/e 503.96 (M−1); Anal. Calcd for $C_{27}H_{26}F_3N_7$: C, 64.15; H, 5.18; N, 19.39. Found: C, 64.10; H, 5.37; N, 19.08.

5.3.3.17. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-[2-(4-morpholinyl)ethyl]-*N″*-[4-(trifluoromethyl)phenyl]guanidine (84). Product oiled out upon addition of water. Water was decanted off and residue washed with water to give the desired product in 73% (170 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 9.10 (br s, 1H), 7.71–7.62 (m, 3H), 7.33–7.17 (m, 5H), 7.10–6.92 (m, 2H), 5.93 (s, 1H), 3.60–3.35 (m, 8H), 2.87–2.75 (m, 2H), 2.40–2.30 (m, 2H), 2.17 (s, 3H); MS (ESI^-) m/e 526.00 (M−1); Anal. Calcd for $C_{27}H_{32}F_3N_7O_1$ 0.5HI: C, 54.82; H, 5.54; N, 16.57. Found: C, 54.92; H, 5.64; N, 16.48.

5.3.3.18. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N′*-(2,2,2-trifluoroethyl)-*N″*-[4-(trifluoromethyl)-phenyl]guanidine (85). Reaction was performed in a sealed tube with 2,2,2-trifluoroethylamine (9 equiv). Desired product was obtained in 71% (313 mg) yield. ^1H NMR (DMSO-d_6, 300 MHz) δ 9.66 (t, 1H, $J = 5.8$ Hz), 7.61 (d, 2H, $J = 8.3$ Hz), 7.30–7.15 (m, 5H), 6.97 (d, 2H, $J = 8.0$ Hz), 5.96 (s, 1H), 4.21 (m, 2H), 3.42 (m, 2H), 2.78 (m, 2H), 2.11 (s, 3H); MS (ESI^+) m/e 496.85 (M+H); MS (ESI^-) m/e 494.87 (M−1); Anal. Calcd for $C_{23}H_{22}F_6N_6$: C, 55.64; H, 4.47; N, 16.93. Found: C, 55.57; H, 4.44; N, 16.83.

5.3.4. General procedure D, for synthesis of compounds of type 7 (86–88, Scheme 3). A stirring solution of **31** (1.0 equiv) in anhydrous DCM was treated with an appropriate electrophile (1.0 equiv) and DIEA (1.1 equiv) sequentially. The resulting solution was stirred at room temperature until the starting material was consumed as judged by LCMS (2–8 h). The reaction

mixture was then concentrated under reduced pressure and purified by silica gel flash column chromatography using the solvent system indicated below.

5.3.4.1. *N*″-(Ethanoyl)-*N*-{4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}-*N*′-[4-(trifluoromethyl) phenyl]guanidine (86). Solvent system used: 70% EtOAc in hexanes. Yield 63%; Electrophile used, Acetyl chloride; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 12.80 (br s, 1H), 12.48 (s, 1H), 7.90–7.88 (m, 2H), 7.70–7.62 (m, 2H), 7.19–7.27 (m, 5H), 6.10 (s, 1H), 3.53–3.57 (m, 2H), 2.84 (t, $J = 7.3$ Hz, 2H), 2.22 (s, 3H), 2.06 (s, 3H); HRMS calcd for $C_{23}H_{23}F_3N_6O$ (M+H) 457.1964, found 457.1960.

5.3.4.2. Benzyl(*E*)-({4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}amino)[4-(trifluoromethyl)anilino]-methylidenecarbamate (87). Solvent system used: 80% EtOAc in hexanes. Yield 78%; Electrophile used, Benzyl 2-nitrophenylcarbonate; ^{1}H NMR (CD_3OD, 400 MHz) δ 7.74 (br s, 3H), 7.39–7.14 (m, 12H), 6.03 (s, 1H), 5.15 (s, 2H), 3.58 (br s, 2H), 2.87 (t, 2H, $J = 8$ Hz), 2.24 (s, 3H); HRMS calcd for $C_{29}H_{27}F_3N_6O_2$ (M+H) 549.2226, found 549.2233.

5.3.4.3. *N*-Ethyl-*N*′-{(*E*)-({4-methyl-6-[(2-phenylethyl)-amino]-2-pyrimidinyl}amino)[4-(trifluoromethyl)anilino]-methylidene}urea (88). Solvent system used: 10% Methanol in DCM. Yield 47%; Electrophile used, Ethyl isocyanate; ^{1}H NMR (DMSO-d_6, 400 MHz) δ 12.60 (br s, 1H), 12.41 (s, 1H); 7.90–7.92 (m, 2H), 7.70–7.75 (m, 1H), 7.54–7.59 (m, 2H), 7.17–7.29 (m, 6H), 6.03 (s, 1H), 3.50–3.52 (m, 2H), 3.05–3.11 (m, 2H), 2.48 (t, $J = 9.5$ Hz, 2H), 2.24 (s, 3H), 1.04 (t, $J = 7.2$ Hz, 3H); HRMS calcd for $C_{24}H_{26}F_3N_7O$ (M+H) 486.2229, found 486.2234.

5.3.4.4. *N*-(4-Chlorophenyl)-*N*′-{4-methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}urea (89). 1-(4-Chloro-phenyl)-3-amidineurea prepared by the method of Skowronska-Serafin and Urbanski[28] and methyl acetoacetate (1.2 equiv) were combined in MeOH followed by dropwise addition of NaOMe (1 equiv). The reaction mixture was heated at reflux for 2 h during which time a precipitate formed. The reaction mixture was cooled, filtered, and washed with cold MeOH to give the sodium salt. This material was dissolved in water and glacial acetic acid added until solution reached pH 5. A white precipitate formed and was filtered, washed with water, and dried in vacuum oven to obtain the desired product. MS (ESI$^+$) m/e 301.0 (M+Na). This intermediate was dissolved in $POCl_3$ and stirred at 100 °C in a sealed tube for 2 h. The $POCl_3$ was removed in vacuo and crushed ice added to the residue. Vigorous stirring afforded the chloride as a white solid that was isolated by filtration. Phenethylamine (0.23 mL, 1.81 mmol) and the chloride (258 mg, 0.91 mmol) were dissolved in EtOH (10 mL) with K_2CO_3 (250 mg, 1.81 mmol) and the reaction was heated at reflux for 16 h. H_2O was added to incipient cloudiness. The resultant precipitate was filtered, washed with water, and air dried to give the desired product in 17% (59 mg) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 10.40 (br s, 1H), 8.75 (br s, 1H), 7.60–7.15 (m, 9H), 6.20 (s, 1H), 3.60 (m, 2H), 2.87 (t, 2H, $J = 7.3$ Hz), 2.33 (s, 3H); HRMS calcd for $C_{20}H_{20}ClN_5O$ (M+H) 382.1435, found 382.1453.

5.3.4.5. *N*-{4-Methyl-6-[(2-phenylethyl)amino]-2-pyrimidinyl}-*N*′-[4-(trifluoromethyl)phenyl]thiourea (90). Produced as an intermediate according to general procedure C in 51% (5.4 g) yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 10.42 (s, 1H), 8.04 (d, 2H, $J = 8.2$ Hz), 7.88 (s, 1H), 7.75 (d, 2H, $J = 8.0$ Hz), 7.40–7.23 (m, 5H), 6.13 (s, 1H), 3.65–3.35 (m, 2H), 2.90 (t, 2H, $J = 7.5$ Hz), 2.30 (s, 3H); MS (ESI$^+$) m/e 432.06 (M+H); MS (ESI$^-$) m/e 429.88 (M−1); Anal. Calcd for $C_{21}H_{20}F_3N_5S$: C, 58.46; H, 4.67; N, 16.23. Found: C, 58.35; H, 4.66; N, 16.36.

Acknowledgements

We gratefully acknowledge Doug Minick, Andrea Sefler, Randy Rutkowske, Wendy White, and Peter Kitrinos for analytical support; Ben Travis and Frank VanMiddleworth for additional synthetic support; the In Vitro Metabolism Group headed by Ivin Silver for conducting the rat liver S9 assay, and Thimysta Burnette for consultation and assistance in preparing the manuscript.

References and notes

1. Kanatani, A.; Ishihara, A.; Asahi, S.; Tanaka, T.; Ozaki, S.; Ihara, M. *Endocrinology* **1996**, *137*, 3177–3182.
2. Daniels et al. *Proc. Natl. Acad. Sci.* **1995**, *92*, 9067–9071.
3. Doods, H. N.; Wieland, H. A.; Engel, W.; Eberlein, W.; Willim, K.-D.; Entzeroth, M.; Wienen, W.; Rudolf, K. *Reg. Pept.* **1996**, *65*, 71–77.
4. Gerald, C.; Walker, M. W.; Criscione, L.; Gustafson, E. L.; Batzl-Hartmann, C.; Smith, K. E.; Vaysse, P.; Durkin, M. M.; Laz, T. M., et al. *Nature (London)* **1996**, *382*, 168–171.
5. Wyss, P.; Stricker-Krongrad, A.; Brunner, L.; Miller, J.; Crossthwaite, A. *Reg. Pept.* **1998**, *75–76*, 363–371.
6. Schaffhauser, A. O.; Stricker-Krongrad, A.; Brunner, L.; Cumin, F.; Gerald, C.; Whitebread, S.; Criscione, L.; Hofbauer, K. G. *Diabetes* **1997**, *46*, 1792–1798.
7. Kirby, D. A.; Koerber, S. C.; May, J. M.; Hagaman, C.; Cullen, M. J.; Pelleymounter, M. A.; Rivier, J. E. *J. Med. Chem.* **1995**, *38*, 4579–4586.
8. Criscione, L.; Rigollier, P.; Batzl-Hartmann, C.; Rueger, H.; Stricker-Krongrad, A.; Wyss, P.; Brunner, L.; Whitebread, S.; Yamaguchi, Y.; Gerald, C.; Heurich, R. O.; Walker, M. W.; Chiesi, M.; Schilling, W.; Hofbauer, K. G.; Levens, N. *J. Clin. Invest.* **1998**, *102*, 2136–2145.
9. Daniels, A. J.; Grizzle, M. K.; Wiard, R. P.; Matthews, J. E.; Heyer, D. *Reg. Pept.* **2002**, *106*, 47–56.
10. Rueger, H.; Schmidlin, T.; Rigollier, P.; Yamaguchi, Y.; Tintelnot-Blomley, M.; Schilling, W.; Criscione, L.; Stutz, S. PCT Int. Appl. (Novartis AG), WO9720821, 1997.
11. Rueger, H.; Schmidlin, T.; Rigollier, P.; Yamaguchi, Y.; Tintelnot-Blomley, M.; Schilling, W.; Criscione, L. PCT Int. Appl. (Novartis AG), WO9720820, 1997.
12. Fukami, T.; Fukuroda, T.; Kanatani, A.; Ihara, M. PCT Int. Appl. (Banyu Pharmaceutical), WO9825908, 1998.

13. Fukami, T.; Fukuroda, T.; Kanatani, A.; Ihara, M. PCT Int. Appl. (Banyu Pharmaceutical, Japan), WO9827063, 1998.
14. Sato, N.; Takahashi, T.; Shibata, T.; Haga, Y.; Sakuraba, A.; Hirose, M.; Sato, M.; Nonoshita, K.; Koike, Y.; Kitazawa, H.; Fujino, N.; Ishii, Y.; Ishihara, A.; Kanatani, A.; Fukami, T. *J. Med. Chem.* **2003**, *46*, 666–669.
15. Thurkauf, A.; Yuan, J.; Hutchison, A.; Blum, C. A.; Elliot, R. L.; Hammond, M. PCT Int. Appl. (Neurogen Corporation, USA; Pfizer), WO9901128, 1999.
16. Buehlmayer, P. PCT Int. Appl. (Novartis AG, Switz.; Novartis-Erfindungen Verwaltungsgesellschaft m.b.H.), WO9932466, 1999.
17. Satoh, Y.; Hatori, C.; Ito, H. *Biorg. Med. Chem. Lett.* **2002**, *12*, 1009–1011.
18. Dugar, S.; Neustadt, B. R.; Stamford, A. W.; Wu, Y. PCT Int. Appl. (Schering Corp.), WO9964394, 1999.
19. Youngman, M. A.; McNally, J. J.; Lovenberg, T. W.; Reitz, A. B.; Willard, N. M.; Nepomuceno, D. H.; Wilson, S. J.; Crooke, J. J.; Rosenthal, D.; Vaidya, A. H.; Dax, S. L. *J. Med. Chem.* **2000**, *43*, 346–350.
20. Fotsch, C.; Sonnenberg, J. D.; Chen, N.; Hale, C.; Karbon, W.; Norman, M. H. *J. Med. Chem.* **2001**, *44*, 2344–2356.
21. Kreutzberger, A.; Sellheim, M. *J. Heterocycl. Chem.* **1985**, *22*, 721–723, 2-Cyanoamino-4-hydroxy-6-methylpyrimidine is now commercially available.
22. Taylor, E. C., Jr.; Barton, J. W. *J. Org. Chem.* **1959**, *24*, 127–128.
23. Naim, S. S.; Sharma, S. *Synthesis* **1992**, 664–666.
24. For a description of the method used to analyze drug levels in whole brain, see: Polli, J. W.; Humphreys, J. E.; Wring, S. A.; Burnette, T. C.; Read, K. D.; Hersey, A.; Butina, D.; Bertolotti, L.; Pugnaghi, P.; Serabjiy-Singh, C. S. Comparison of MDCK and Bovine Brain Endothelial Cells (BBEC's) as a Blood-Brain Barrier Screen. In *Early Drug Development in Progress in the Reduction, Refinement, and Replacement of Animal Experimentation*; Balls, M., van Zeller, A. M., Halder, M., Eds.; Elsevier Science: New York, 2000; pp 271–289.
25. Matthews, J. E.; Jansen, M.; Lyerly, D.; Cox, R.; Chen, W. J.; Koller, K. J.; Daniels, A. J. *Reg. Pept.* **1997**, *72*, 113–119.
26. Cheng, Y. C.; Prusoff, W. H. *Biochem. Pharmacol.* **1973**, *22*, 3099–3108.
27. Wring, S. A.; Silver, I. S.; Serabjit-Sighn, C. J. *Methods Enzymol.* **2000**, *357*, 285–295.
28. Skowronska-Serafin, B.; Urbanski, T. *Tetrahedron* **1960**, *10*, 12–25.