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Discovery of Novel Protease Activated Receptors 1 Antagonists with Potent Antithrombotic Activity in Vivo

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Protease activated receptors (PARs) or thrombin receptors constitute a class of G-protein-coupled receptors (GPCRs) implicated in the activation of many physiological mechanisms. Thus, thrombin activates many cell types such as vascular smooth muscle cells, leukocytes, endothelial cells, and platelets via activation of these receptors. In humans, thrombin-induced platelet aggregation is mediated by one subtype of these receptors, termed PAR1. This article describes the discovery of new antagonists of these receptors and more specifically two compounds: 2-[5-oxo-5-(4-pyridin-2-ylpiperazin-1-yl)penta-1,3-dienyl]benzonitrile **36** (F 16618) and 3-(2-chlorophenyl)-1-[4-(4-fluorobenzyl)piperazin-1-yl]propenone **39** (F 16357), obtained after optimization. Both compounds are able to inhibit SFLLR-induced human platelet aggregation and display antithrombotic activity in an arteriovenous shunt model in the rat after iv or oral administration. Furthermore, these compounds are devoid of bleeding side effects often observed with other types of antiplatelet drugs, which constitutes a promising advantage for this new class of antithrombotic agents.

Introduction

Antiaggregant drugs such as ADP antagonists, thromboxane A2 inhibitors, or GPIIb/IIIa[a] antagonists constitute very important classes of antithrombotic therapies.[1,2] These drugs are essential for the treatment of life-threatening pathologies such as stroke, acute coronary syndrome, or myocardial infarction. Platelets are activated by a variety of agonists such as thrombin, ADP, thromboxane A2, collagen, serotonin, or epinephrine. Among these, thrombin is probably the most potent activator of platelet aggregation. Thrombin is also a well-known serine protease involved in the coagulation cascade that converts soluble fibrinogen to fibrin. The aggregation of platelets by thrombin is mediated via proteolytic activation of specific cell surface receptors known as protease activated receptors (PARs) or thrombin receptors.[3−5] Four PARs have been identified so far: PAR1, PAR2, PAR3, and PAR4. PAR1, PAR3, and PAR4 are activated by thrombin, while PAR2 is activated by trypsin. PAR1 is the major thrombin-activated receptor on human, monkey, and guinea pig platelets.[6]

PARs are activated by a unique mechanism in which a proteolytic enzyme such as thrombin (or trypsin) binds to the receptor and then cleaves its extracellular domain between Arg41 and Ser42.[7] The newly unmasked amino terminus binds intramolecularly to the proximally located transmembrane loop of the GPCR, eliciting intracellular signaling. This particular intramolecular activation mechanism, termed "tethered ligand mechanism", makes this target particularly difficult to address. Indeed, the aim of this project was not to inhibit thrombin-mediated receptor cleavage but to compete with the intramolecular activation of the receptor by the tethered ligand, an entropically favored interaction compared to an external antagonist ligand. Inhibiting thrombin-mediated platelet activation without affecting thrombin's role in the coagulation cascade could lead to a new promising class of antiplatelet drugs with a limited impact on bleeding.[8] Since all known antithrombotic therapies suffer from drawbacks mainly associated with hemorrhagic side effects, the lack of bleeding with PAR1 antagonists could constitute an essential advantage.

The proof of concept for this new antiaggregant approach and its low impact on bleeding has been recently established in a clinical trial with SCH-530348 (Figure 1), which is currently in phase 3 for the treatment of acute coronary syndrome.[9] Today, this is the only antithrombotic treatment by a PAR1 antagonist to have been demonstrated in humans.

Other compounds have been published as potent PAR1 antagonists without reaching the clinic. Among them ER-121958-06 (Figure 1) has been described as a potent antiaggregant compound on human platelets (IC_{50} = 21 nM).[8e] In a search for novel antagonists of PAR1, we have performed a screening of our proprietary library and found an interesting hit that, after a first optimization step, gave lead compound **4** derived from cinnamoylpiperazine (Scheme 1).

Chemistry

Lead compound **4** was prepared according to route A described in Scheme 1. Cinnamic acid **1a** was first transformed into an acid chloride, which was reacted with *tert*-butylpiperazine

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[a] Abbreviations: PARs, protease activated receptors; GPCRs, G-protein-coupled receptors; iv, intravenous; po, per os; ADP, adenosine diphosphate; GPIIb/IIIa, glycoprotein IIb/IIIa; FLIPR, fluorometric imaging plate reader.

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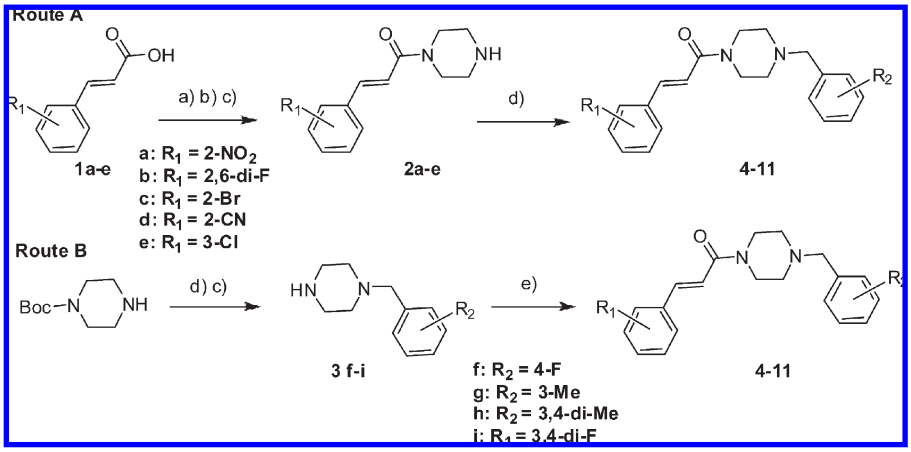
Scheme 1. Synthesis of Cinnamoyl Derivatives **4**−**11**[a]



[a] Conditions: (a) $SOCl_2$, reflux; (b) Boc-piperazine, CH_2Cl_2, Et_3N, room temp; (c) TFA, toluene, room temp; (d) benzyl bromide, CH_2Cl_2, Et_3N, room temp; (e) cinnamic acids **1a**−**e**, CH_2Cl_2, PS-carbodiimide, HOBT, room temp.

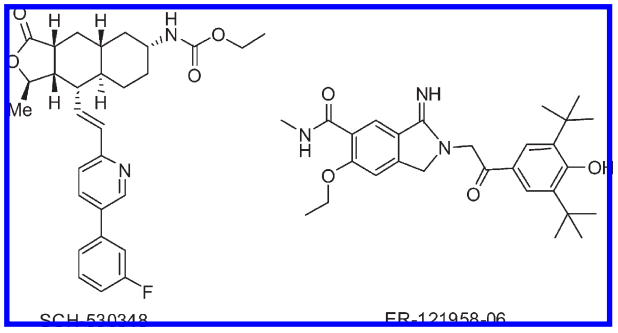


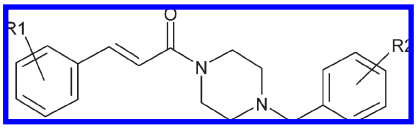
Figure 1. Structures of SCH-530348 and ER-121958-06.

1-carboxylate in the presence of Et_3N and then deprotected with TFA to give piperazine **2a**. This intermediate provided compound **4** after treatment with 4-fluorobenzyl bromide in the presence of Et_3N.

Optimization of compound **4** relied, in the first instance, on a screening assay based on the inhibition of fluorescent calcium release induced by the PAR1 selective agonist peptide ($SFLLR\text{-}NH_2$) in CHO cells (FLIPR).[10] This peptide, also termed thrombin receptor activating peptide (TRAP), has been shown to be sufficient to mimic human receptor activation by thrombin.[11−15] Table 1 summarizes the results obtained by modification of the substitution of the two aromatic rings of compound **4**. Compounds **5**−**11** were prepared according to the synthetic routes depicted in Scheme 1. Compound **12** was prepared by reduction of compound **4** with tin chloride. The ortho substitution on the left-hand phenyl gives the best results as exemplified by the comparison of compound **11** (3-Cl, 0.7%) with compound **6** (2-Br, 93.1%) or compound **39** (2-Cl, 92.8%) depicted in Table 4. The nature of the substituent in this position is particularly important, as exemplified by the comparison of compounds **4** ($2\text{-}NO_2$, 88.6%) and **12** ($2\text{-}NH_2$, 7.4%). Other analogues have been prepared with substitutions in meta or para positions that confirmed that the ortho position afforded the most active products (data not shown).

We then focused our attention on the modification of the unsaturated linker of compound **4**. Alkane and alkyne analogues **13** and **14**, respectively (Table 2), were prepared following a synthetic scheme similar to that used for compounds **4**−**11** but starting with commercially available

Table 1. SAR for the Substitution of the Aromatic Rings



compd	R1	R2	% antagonism at 10 μM ± SEM[a]
4	$2\text{-}NO_2$	4-F	88.6 ± 3.8
5	2,6-diF	3-Me	86.2 ± 4.3
6	2-Br	4-F	93.1 ± 0.9
7	2-CN	4-F	85.4 ± 1.9
8	2,6-diF	3,4-diMe	65.3 ± 4.6
9	2,6-diF	3,4-diF	73.1 ± 3.6
10	$2\text{-}NO_2$	3-Me	72.4 ± 5.3
11	3-Cl	4-F	0.7 ± 6.8
12	$2\text{-}NH_2$	4-F	7.4 ± 4.3

[a] Inhibition of calcium release induced by 1 μM $SFLLR\text{-}NH_2$.

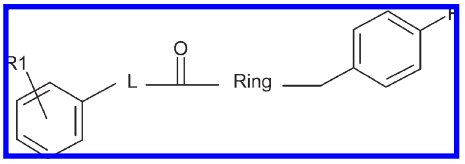
3-(2-nitrophenyl)propionic acid and (2-nitrophenyl)propynoic acid. Diene analogue **17** was synthesized according to Scheme 2 from commercially available (*E*)-3-(2-nitrophenyl)-propenal.

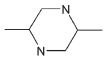
A number of analogues were obtained by substitution or replacement of the piperazine ring. 2,5-Dimethylpiperazine (**18**), piperazine-2-one (**19**), homopiperazine (**20**, **22**), and 4-aminopiperidine (**21**) analogues were prepared using synthetic approaches similar to the one depicted in Scheme 1. Introduction of a piperidine ring was performed according to Scheme 3.

Piperidine-4-carboxylic acid **23** was protected by a BOC group and then condensed with *N*,*O*-dimethylhydroxylamine after activation of the acid by EDCI and HOOBT. Boc deprotection of Weinreb amide **24** with TFA followed by reductive amination with 4-F-benzaldehyde afforded intermediate **25**. Diethyl methylphosphonate was first deprotonated with *n*-BuLi at −50 °C and then treated with amide **25** to give intermediate **26**. Horner−Wadsworth−Emmons reaction between this phoshonate and 2-Cl-benzaldehyde gave compound **27**. The evaluation in the PAR1 screening assay of these compounds is summarized in Table 2.

We also studied the replacement of the 4-F-benzyl part of compound **4** by a substituted phenyl (**28**, **29**) or a heterocycle (**30**).

Table 2. SAR of Analogues of Compound **4**[a]



Compound	R1	L	Ring	% antag. @ 10μM ± SEM*
13	2-NO_2	CH_2CH_2		45.3 ± 3.8
14	2-NO_2	C≡C		28.6 ± 5.1
17	2-NO_2	CH=CH-CH=CH		59.4 ± 1.9
18	2,5-diF	CH=CH		59.0 ± 1.9
19	2-NO_2	CH=CH		26.7 ± 1.9
20	2-NO_2	CH=CH		38.2 ± 3.1
21	2-NO_2	CH=CH		0 ± 3.1
22	2-Cl	CH=CH-CH=CH		37.8 ± 6.9
27	2-Cl	CH=CH		15.8 ± 4.5

[a] The asterisk (*) indicates inhibition of calcium release induced by 1 μM SFLLR-NH_2.

The synthesis of these compounds was accomplished following the same experimental routes as described in Scheme 1 except that we used an aryl- or heteroarylpiperazine instead of the Boc piperazine. In addition, we also synthesized amides, sulfonamides, or ureas in this position via the reaction of an acyl chloride, sulfonyl chloride, or isocyanate with intermediates **2**, as exemplified by compounds **31**−**33**. The evaluation in the PAR1 screening assay of these compounds is summarized in Table 3.

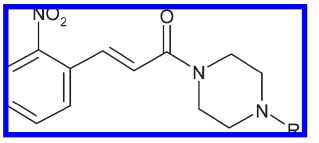
The results displayed in Table 3 show that replacement of the 4-F-benzyl group of compound **4** by either a 3-Cl-phenyl or a cycloheptylcarbonyl leads to potent compounds (**28** and **31**). On the other hand, close analogues such as **29**, **30**, or **32** are completely inactive. Urea analogue **33** gives very moderate activity.

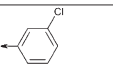
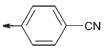
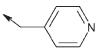
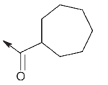
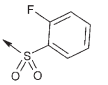
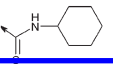
Finally, we combined the different modifications to further optimize the activity. The most interesting compounds, obtained by synthetic routes similar to the one described in Schemes 1 and 2, are summarized in Table 4.

Results and Discussion

In order to more precisely determine the activity of these compounds, additional pharmacological evaluations were conducted. First, a concentration−response was performed

Table 3. SAR on the Piperazine Substitution[a]



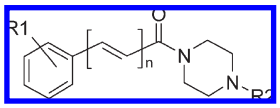
Compound	R1	% antag. @ 10μM ± SEM*
28		97.1 ± 2.1
29		18.8 ± 12.1
30		0 ± 2.2
31		99.9 ± 0.3
32		0 ± 4.6
33		39.2 ± 1.5

[a] The asterisk (*) indicates inhibition of calcium release induced by 1 μM SFLLR-NH_2.

on the inhibition of calcium release model (FLIPR assay) in order to classify the potency of these compounds as PAR1 antagonists. Since these compounds behave as competitive antagonists, pA_2 values were calculated for each of them. Second, antiaggregant activity was evaluated using an SFLLR-induced human platelet aggregation model.[6] Following this, an arteriovenous extracorporal shunt model in anesthetized rats was performed to address the antithrombotic potential of these compounds in vivo.[16] In this model, a shunt is realized between the left carotid artery and the right jugular vein. A silk thread, placed in the central portion of the shunt, was used as thrombogenic substrate. A thermal microprobe was placed onto the central part of the shunt to determine the time for occlusion to occur (indicated by a dramatic decrease in temperature). This assay was performed by either intravenous or oral administration in order to obtain the first evaluation of the bioavailability of the molecules. An antithrombotic agent is able to increase the occlusion time in such model.

Among the compounds listed in Table 4, several are potent antagonists of PAR1 with pA_2 or pK_b of > 7 (i.e., **34**, **35**, **37**). However, there is no clear relationship between this activity and the inhibition of human platelet aggregation. Thus, compound **39** has a weaker antagonistic activity compared to compound **34** (pA_2 of 6.42 vs 7.16, respectively) but a very comparable inhibitory activity on platelet aggregation (pK_b of 5.52 vs 5.50, respectively). The reference compound ER-121958-06 displays a moderate pK_b of 6.85 (noncompetitive displacement curve) but a very potent antiaggregant activity. Surprisingly, compound **38** was completely inactive in the aggregation model and very weakly active in the shunt model despite a good antagonistic activity (pA_2 of 6.67). Since there is no

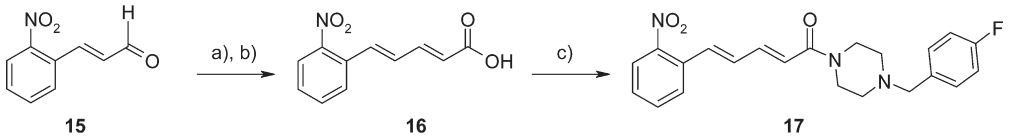
Table 4. SAR of Optimized Analogues of Compound **4**



compd	R1	*n*	R2	% antagonism at 10 μM ± SEM[a]	pA_2[a] [limit values]	hum plat (pK_b)[b] [limit values]	AV shunt (iv, %) ± SEM[c]	AV shunt (po, %) ± SEM[d]
ER-121958-06					6.85[e] [ND]	8.18 [8.09−8.49]	27 ± 13	43 ± 11
34	2-NO_2	2	c-pentyl	91.7 ± 2.8	7.16 [ND]	5.50 [5.34−6.11]	44 ± 8	7 ± 4
35	2-NO_2	2	phenyl	100.0 ± 0.2	7.23 [ND]	5.65 [5.27−5.71]	3 ± 5	ND
36	2-CN	2	2-pyridine	98.7 ± 0.5	6.49 [6.40−6.55]	5.30 [5.22−5.49]	58 ± 17	54 ± 14
37	2-CN	1	CO-cHeptyl	92.6 ± 5.3	7.06[e] [ND]	5.17 [5.07−5.27]	19 ± 6	ND
38	2,6-diF	1	CO-c-heptyl	98.9 ± 0.6	6.67 [6.28−6.88]	0 [ND]	14 ± 3	0 ± 4
39	2-Cl	1	4-F-benzyl	92.8 ± 0.9	6.67 [6.34−7.14]	5.52 [5.32−5.97]	28 ± 6	59 ± 11
40	2-NO_2	1	CH_2-c-hexyl	91.5 ± 3.0	6.31 [6.28−6.36]	5.31 [4.80−5.82]	29 ± 4	ND

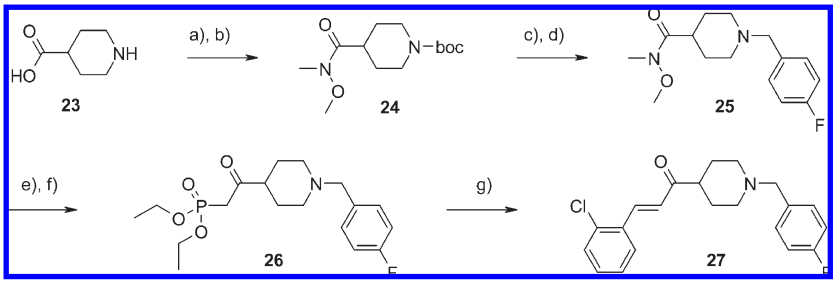
[a] Inhibition of calcium release induced by 1 μM SFLLR-NH_2: % at 10 μM or pA_2. [b] Inhibition of SFLLR-induced human platelet aggregation. [c] % increase of the occlusion time in an arteriovenous shunt in rat at 1.25 mg/kg iv. [d] % increase of the occlusion time in an arteriovenous shunt in rat at 40 mg/kg po. [e] pK_b value (noncompetitive profile).

Scheme 2. Synthesis of Diene Compound **17**[a]



[a] Conditions: (a) ethyl 2-(diethoxyphosphoryl)acetate, NaH, THF, room temp, 92%; (b) KOH, EtOH, 70 °C, 98%; (c) 4-F-benzylpiperazine, HOOBT, EDCI, DIEA, CH_2Cl_2, room temp, 77%.

Scheme 3. Synthesis of Piperidine Analogue **27**[a]



[a] Conditions: (a) $(BOC)_2O$, K_2CO_3, H_2O−THF, 99%; (b) EDCI, HOOBT, DMF, DIEA, MeONHMe·HCl, 88%; (c) TFA, toluene, room temp, 74%; (d) 4-F-benzaldehyde, $NaBH(OAc)_3$, AcOH, 1,2-DCE, room temp, 77% ; (e) $(EtO)_2P(O)Me$, *n*-BuLi, THF, −50 °C; (f) +amide **25**, THF; (g) 2-Cl-benzaldehyde, K_2CO_3, CH_3CN, room temp, 68% (three steps).

basic nitrogen in this compound (as well as in compound **37**), formation of a salt was not possible. Solubility issues are probably the main reason for the poor activity observed in most assays. Similarly and despite the fact that compound **35** was tested as a hydrochloride salt, its poor water solubility (< 1 mg/mL) could also explain the weak activity observed in the shunt model. In this model, after intravenous administration, uneven results were obtained. Compounds **34** and **36** displayed the most potent activities in these conditions with 44% and 58% increases in the occlusion time, respectively. The activity of compound **34** after iv administration was not confirmed after oral dosing (44% iv vs 7% po), probably because of a limited bioavailability. Inversely, compound **39** gave a much better activity after oral administration at 40 mg/kg (59%) compared to 1.25 mg/kg iv (28%), while compound **36** gave very homogeneous results (58% iv vs 54% po). Interestingly, the reference compound ER-121958-06 is slightly less potent in the in vivo shunt model compared to compound **36** (both routes of administration) and to a lesser extent to compound **39**, despite very potent antiaggregant activity (pK_b = 8.18). Overall, these results demonstrate a potent antithrombotic effect of molecules **36** and **39** in this model.

The selectivity of these compounds versus PAR2 and PAR4 was addressed. For that purpose, we used a serum response element (SRE) dependent luciferase activity model[18] in Cos-7 cells expressing PAR2, PAR4, or PAR1. We evaluated the ability of the compounds **36** and **39** to antagonize the increase of SRE-luciferase activity mediated by each selective agonist peptides. Thus, for PAR2, the activation by SLIGRL was weakly antagonized with pK_b values (±SEM, n = 3) of 4.39 ± 0.12 and 4.45 ± 0.07 for compounds **36** and **39**, respectively. For PAR4, the activation by AYPGKF was antagonized with pK_b values (±SEM, n = 3) of 4.93 ± 0.01 and 4.91 ± 0.11, respectively. For a clear comparison purpose, we also determined PAR1 antagonistic activity of these two compounds in the same SRE-luciferase model using SFLLR as selective agonist.

Table 5. ADME Profile of the Two Lead Compounds **36** and **39**

	36	39
water solubility[a] (mg/mL)	2.5	1.7
plasma $T_{1/2}$[b] (min)	167 ± 16	130 ± 30
F[a] (%)	42	43
CL[a] ((L/h)/kg)	0.36	2.4

[a] For the hydrochloride salt. [b] Dosed iv (1 mg/kg) and po (5 mg/kg) in male rats (mean value over three rats).

Compounds **36** and **39** displayed antagonistic activity with pK_b values (±SEM, $n = 3$) of 5.60 ± 0.03 and 5.55 ± 0.04, respectively. Thus, in this model, these compounds displayed a selectivity of about 15-fold vs PAR2 and 5-fold vs PAR4. It is noteworthy that none of these molecules displayed any agonistic activity in this model.

To address the impact on bleeding time, which is often a limitation of antithrombotic agents, these compounds were evaluated in a well-established tail cut rat model[17] in which they were devoid of significant effect (data not shown).

Compounds **36** (F 16618) and **39** (F 16357), being the most potent compounds after oral administration in the rat, were further evaluated in an ADME protocol (Table 5). Both compounds have reasonable water solubility and a good bioavailability and clearance, while the plasma half-lives in the rat are moderate.

Conclusions

We have identified new PAR1 antagonists that inhibit SFLLR-induced human platelet aggregation and are active by both iv and oral routes in a rat thrombosis model without any significant impact on bleeding time. Among them, compounds **36** and **39** are the most promising in terms of antithrombotic activity and ADME profile. Furthermore, these compounds are particularly easy to synthesize, which could constitute an interesting advantage compared with other known PAR1 antagonists such as SCH-530348 or ER-121958-06. Further pharmacological characterization of these compounds is currently ongoing.

Experimental Section

Chemistry. Melting points were determined on a Buchi 530 melting point apparatus and were not corrected. ^{1}H NMR spectra were recorded on a Bruker Avance 400 spectrometer operating at 400 MHz for ^{1}H and 100 MHz for ^{13}C. Chemical shifts are reported in δ value (ppm) relative to an internal standard of tetramethylsilane. Microanalyses were obtained on a Fison EA 1108/CHN analyzer. Mass spectra (TSQ 7000 Finnigan, Thermoelectron Corporation) were determined by electron spray ionization (ESI). Only 100% relative intensity peaks are given. HPLC analysis were performed on a Waters instrument with a photodiode array (PDA) detector (UV detection with chromatogram extracted at 220 nm). The columns used were either a C18 Symmetry, 50 mm × 4.6 mm, 5 μm, or a C18 XTerra MS, 50 mm × 4.6 mm, 5 μm (both from Waters), eluting at 3 mL/min with a 6 min gradient from 0 to 100% CH_3CN (+0.05% TFA) followed by 1 min at 100% CH_3CN. The purity of final compounds was determined by either elemental analysis or analytical HPLC. All compounds described hereafter have a minimum purity of 95%.

Representative Procedure A for the Synthesis of Cinnamoyl Derivatives. (*E*)-3-(2-Nitrophenyl)-1-(piperazin-1-yl)prop-2-en-1-one (2a). (*E*)-2-Nitrocinnamic acid (5.2 g, 26.92 mmol) was treated with thionyl chloride (50 mL) under reflux for 6 h. The mixture was then concentrated under reduced pressure to give the crude acid chloride. To a solution of this intermediate and triethylamine (4.7 mL, 33.6 mmol) in CH_2Cl_2 (60 mL) was added *tert*-butyl piperazine-1-carboxylate (4.18 g, 22.43 mmol). After being stirred overnight at room temperature, the reaction mixture was diluted with 1 N aqueous NaOH, extracted with CH_2Cl_2 (×2), dried with $MgSO_4$, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2/MeOH/NH_4OH, 98/1.5/0.5) gave (*E*)-*tert*-butyl 4-(3-(2-nitrophenyl)acryloyl)-piperazine-1-carboxylate (6.94 g, 86%): ^{1}H NMR (DMSO-d_6) δ 1.42 (s, 9H), 3.37 (broad s, 4H), 3.56 (broad s, 2H), 3.73 (broad s, 2H), 7.50 (d, J = 15 Hz, 1H), 7.62 (d, J = 15 Hz, 1H), 7.70 (t, J = 8 Hz, 1H), 8.19 (m, 2H), 8.63 (s, 1H); HPLC (Symmetry) t_R = 4.86 min, 97.8%; MS (ESI) m/z = 362 [MH^+].

To a solution of this intermediate (6.17 g, 17.07 mmol) in toluene (50 mL) was added TFA (47 mL, 614 mmol). After being stirred 2 h at room temperature, the reaction mixture was concentrated under reduced pressure. The reaction mixture was diluted with 1 N aqueous NaOH, extracted with CH_2Cl_2 (×2), dried with $MgSO_4$, and concentrated under reduced pressure to give the desired compound **2a** (3.9 g, 93%). This intermediate was used without further purification for the next step: ^{1}H NMR (DMSO-d_6) δ 3.69 (broad s, 4H), 3.50 (broad s, 2H), 3.65 (broad s, 2H), 7.48 (d, J = 15 Hz, 1H), 7.59 (d, J = 15 Hz, 1H), 7.69 (t, J = 8 Hz, 1H), 8.19 (m, 2H), 8.61 (s, 1H); MS (ESI+) m/z = 262 [MH^+].

(*E*)-1-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-(2-nitrophenyl)propenone (4). To a solution of intermediate **2a** (441 mg, 1.79 mmol) and triethylamine (375 μL, 2.68 mmol) in CH_2Cl_2 (10 mL) was added 4-F-benzyl bromide (268 μL, 2.15 mmol). After being stirred 5 h at room temperature, the reaction mixture was diluted with 1 N aqueous NaOH, extracted with CH_2Cl_2 (×2), dried with $MgSO_4$, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2/MeOH, 100/0 to 97/3) gave the desired product (428 mg, 65%). Treatment of this product by aqueous HCl afforded the hydrochloride salt as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 3.01 (broad m, 2H), 3.20 (broad t, J = 12 Hz, 1H), 3.36 (m, 2H), 3.62 (broad t, J = 12 Hz, 1H), 4.35 (broad s, 2H), 4.52 (broad t, J = 16 Hz, 2H), 7.27−7.34 (m, 3H), 7.66 (t, J = 7 Hz, 3H), 7.76−7.82 (m, 2H), 8.03 (dd, J = 7, 15 Hz, 1H), 11.32 (broad s, 1H); HPLC (Xterra MS) t_R = 3.52 min, 98.5%; MS (ESI+) m/z = 370 [MH^+]. Anal. Calcd for $C_{20}H_{20}F_1N_3O_3 \cdot HCl$: C, 59.19; H, 5.22; N, 10.35. Found: C, 58.80; H, 5.22; N, 10.15.

Representative Procedure B for the Synthesis of Cinnamoyl Derivatives. 1-(4-Fluorobenzyl)piperazine (3f). To a solution of *tert*-butyl piperazine-1-carboxylate (2.0 g, 10.74 mmol) and triethylamine (2.25 mL, 11.81 mmol) in CH_2Cl_2 (40 mL) was added 4-F-benzyl bromide (1.5 mL, 16.11 mmol). After being stirred overnight at room temperature, the reaction mixture was diluted with water, extracted with CH_2Cl_2 (×2), dried with $MgSO_4$, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, PE/AcOEt, 80/20) gave the desired product (2.1 g, 60%). To a solution of this intermediate (2.1 g, 7.13 mmol) in toluene (40 mL) was added TFA (16 mL, 214 mmol). After being stirred for 1.2 h at room temperature, the reaction mixture was concentrated under reduced pressure. The reaction mixture was diluted with 1 N aqueous NaOH, extracted with CH_2Cl_2 (×2), dried with $MgSO_4$, and concentrated under reduced pressure to give the desired compound **3f** as a yellow syrup (1.39 g, 99%): ^{1}H NMR (DMSO-d_6) δ 2.26 (broad s, 4H), 2.67 (broad s, 4H), 3.39 (s, 2H), 7.12 (t, J = 8 Hz, 2H), 7.31 (t, J = 8 Hz, 2H)

(*E*)-1-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-(2-bromophenyl)-propenone (6). To a solution of 2-bromocinnamic acid (140 mg, 0.62 mmol) and intermediate **3f** (101 mg, 0.52 mmol) in CH_2Cl_2 (4 mL) was added PS-carbodiimide (780 mg, 1.6 mmol/g, 1.24 mmol) and HOBT (104 mg, 0.78 mmol).[19] After being stirred overnight at room temperature, the reaction mixture was diluted with 1 N aqueous NaOH, extracted with CH_2Cl_2 (×2), dried with $MgSO_4$, and concentrated under reduced pressure.

Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2/MeOH, 100/0 to 95/5) gave the desired product (129 mg, 57%). Treatment of this product by aqueous HCl afforded the hydrochloride salt as a white powder: ^{1}H NMR (DMSO-d_6) δ 2.90−3.10 (m, 2H), 3.20 (broad t, J = 11 Hz, 1H), 3.34 (broad d, J = 11 Hz, 2H), 3.62 (broad t, J = 11 Hz, 1H), 4.34 (broad s, 2H), 4.52 (broad t, J = 11 Hz, 2H), 7.25−7.40 (m, 4H), 7.45 (t, J = 8 Hz, 1H), 7.64−7.72 (m, 3H), 7.80 (d, J = 15 Hz, 1H), 7.99 (dd, J = 1 Hz, 8 Hz, 1H), 11.40 (s, 1H); HPLC (Xterra MS) t_R = 3.86 min, 99%; MS (ESI+) m/z = 403 [MH^+].

(*E*)-3-(2,6-Difluorophenyl)-1-[4-(3-methylbenzyl)piperazin-1-yl]propenone (5). According to representative procedure A described above, compound **5** was isolated as an orange powder: ^{1}H NMR (DMSO-d_6) δ 2.30 (s, 3H), 2.36 (broad s, 4H), 3.46 (s, 2H), 3.59 (broad s, 4H), 7.00−7.30 (m, 7H), 7.40−7.55 (m, 2H); HPLC (Symmetry) t_R = 3.75 min, 98.7%; MS (ESI+) m/z = 357 [MH^+].

2-{(*E*)-3-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-oxopropenyl}-benzonitrile (7). According to representative procedure A described above, compound **7** was isolated as an HCl salt as an orange powder: ^{1}H NMR (DMSO-d_6) δ 2.90−3.15 (m, 2H), 3.22 (broad t, J = 11 Hz, 1H), 3.35 (broad d, J = 11 Hz, 2H), 3.64 (broad t, J = 11 Hz, 1H), 4.34 (broad s, 2H), 4.53 (broad t, J = 11 Hz, 2H), 7.32 (t, J = 8 Hz, 2H), 7.53 (d, J = 15 Hz, 1H), 7.60 (t, J = 8 Hz, 1H), 7.66 (broad s, 2H), 7.74 (d, J = 15 Hz, 1H), 7.79 (t, J = 8 Hz, 1H), 7.92 (d, J = 7 Hz, 1H), 8.17 (d, J = 7 Hz, 1H), 11.40 (s, 1H); HPLC (Xterra MS) t_R = 3.45 min, 98%; MS (ESI+) m/z = 350 [MH^+].

(*E*)-3-(2,6-Difluorophenyl)-1-[4-(3,4-dimethylbenzyl)piperazin-1-yl]propenone (8). According to representative procedure A described above, compound **8** was isolated as an HCl salt as an orange powder: ^{1}H NMR (DMSO-d_6) of the base δ 2.19 (s, 3H), 2.20 (s, 3H), 2.36 (broad s, 4H), 3.41 (s, 2H), 3.57 (broad s, 4H), 7.00 (d, J = 8 Hz, 1H), 7.05−7.1 (m, 2H), 7.15−7.25 (m, 3H), 7.40−7.55 (m, 2H); HPLC (Symmetry) t_R = 3.85 min, 98%; MS (ESI+) m/z = 371 [MH^+].

(*E*)-1-[4-(3,4-Difluorobenzyl)piperazin-1-yl]-3-(2,6-difluorophenyl)propenone (9). According to representative procedure A described above, compound **9** was isolated as an HCl salt as a yellow powder: ^{1}H NMR (DMSO-d_6) of the base δ 2.39 (broad s, 4H), 3.50 (s, 2H), 3.59 (broad s, 4H), 7.15−7.25 (m, 4H), 7.35−7.55 (m, 4H); HPLC (Symmetry) t_R = 3.64 min, 99%; MS (ESI+) m/z = 379 [MH^+].

(*E*)-1-[4-(3-Methylbenzyl)piperazin-1-yl]-3-(2-nitrophenyl)-propenone (10). According to representative procedure A described above, compound **10** was isolated as an HCl salt as a brown powder: ^{1}H NMR (DMSO-d_6) of the base δ 2.30 (s, 3H), 2.39 (broad s, 4H), 3.47 (s, 2H), 3.57 (broad s, 2H), 3.70 (broad s, 2H), 7.05−7.15 (m, 3H), 7.21 (t, J = 7 Hz, 1H), 7.26 (d, J = 15 Hz, 1H), 7.63 (t, J = 8 Hz, 1H), 7.71 (d, J = 15 Hz, 1H), 7.77 (t, J = 7 Hz, 1H), 8.00−8.05 (m, 2H); HPLC (Symmetry) t_R = 3.65 min, 99%; MS (ESI+) m/z = 366 [MH^+].

(*E*)-1-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-(3-chlorophenyl)-propenone (11). According to representative procedure B described above, compound **11** was isolated as an HCl salt as a white powder: ^{1}H NMR (DMSO-d_6) δ 2.90−3.15 (m, 2H), 3.20 (broad t, J = 11 Hz, 1H), 3.33 (broad d, J = 11 Hz, 2H), 3.62 (broad t, J = 11 Hz, 1H), 4.34 (broad s, 2H), 4.53 (broad d, J = 11 Hz, 2H), 7.32 (t, J = 9 Hz, 2H), 7.36 (d, J = 15 Hz, 1H), 7.45 (broad s, 2H), 7.51 (d, J = 15 Hz, 1H), 7.66 (broad s, 3H), 7.92 (s, 1H), 11.53 (s, 1H); HPLC (Xterra MS) t_R = 3.88 min, 99%; MS (ESI+) m/z = 359 [MH^+].

(*E*)-1-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-(2-aminophenyl)-propenone (12). To a solution of compound **4** (834 mg, 2.26 mmol) in ethanol (25 mL) was added tin chloride dihydrate (2.55 g, 11.3 mmol). After being stirred for 7 h under reflux, the reaction mixture was concentrated under reduced pressure, diluted with aqueous $NaHCO_3$, extracted with ethyl acetate (×2), dried with Na_2SO_4, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2/MeOH/Et_3N, 95/5/0.1) gave the desired product (370 mg, 48%) isolated as an HCl salt as a yellow powder: ^{1}H NMR (DMSO-d_6) of the base: δ 2.36 (broad s, 4H), 3.49 (s, 2H), 3.56 (broad s, 2H), 3.65 (broad s, 2H), 5.75 (s, 2H), 6.52 (t, J = 7 Hz, 1H), 6.66 (d, J = 8 Hz, 1H), 6.94 (d, J = 15 Hz, 1H), 7.02 (t, J = 7 Hz, 1H), 7.15 (t, J = 8 Hz, 2H), 7.35 (m, 2H), 7.47 (d, J = 7 Hz, 1H), 7.67 (d, J = 15 Hz, 1H); HPLC (Xterra MS) t_R = 3.22 min, 96%; MS (ESI+) m/z = 340 [MH^+].

1-[4-(4-Fluorobenzyl)piperazin-1-yl]-3-(2-nitrophenyl)propan-1-one (13). To a solution of 3-(2-nitrophenyl)propionic acid (11 mg, 0.056 mmol) and intermediate **3f** (12 mg, 0.061 mmol) in CH_2Cl_2 (2 mL) was added EDCI (12 mg, 0.061 mmol), HOOBT (10 mg, 0.061 mmol), and DIEA (19 μL, 0.11 mmol). After being stirred overnight at room temperature, the reaction mixture was diluted with 1 N aqueous NaOH, extracted with CH_2Cl_2 (×2), dried with $MgSO_4$, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2/MeOH, 100/0 to 85/15) gave the desired product (20 mg, 95%). Treatment of this product by aqueous HCl afforded the hydrochloride salt as a white powder: ^{1}H NMR (DMSO-d_6) δ 2.69−277 (m, 2H), 2.87−2.96 (m, 2H), 3.03 (t, J = 8 Hz, 3H), 3.27 (broad t, J = 11 Hz, 2H), 3.45 (t, J = 13 Hz, 1H), 4.03 (broad d, J = 14 Hz, 1H), 4.15 (broad s, 2H), 4.43 (broad d, J = 14 Hz, 1H), 7.31 (t, J = 8 Hz, 2H), 7.47 (dt, J = 1, 8 Hz, 1H), 7.55 (d, J = 8 Hz, 1H), 7.62−7.67 (m, 3H), 7.92 (dd, J = 1, 8 Hz, 1H); HPLC (Xterra MS) t_R = 3.56 min, 99%; MS (ESI+) m/z = 372 [MH^+].

1-(4-(4-Fluorobenzyl)piperazin-1-yl)-3-(2-nitrophenyl)prop-2-yn-1-one (14). 1-Iodo-2-nitrobenzene (10 g, 40 mmol) was dissolved in 120 mL of THF under argon atmosphere. Methyl propiolate (14.3 mL, 160 mmol) was added along with copper iodide (300 mg, 1.6 mmol), K_2CO_3 (11 g, 80 mmol), and $Pd(PPh_3)_4$ (920 mg, 0.8 mmol). The mixture was heated at 65 °C for 1.5 h and was concentrated under reduced pressure. The residue was taken up into EtOAc, washed with water, dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2/PE, 40/60 to 50/50) gave methyl 3-(2-nitrophenyl)propiolate (1.53 g, 19%): ^{1}H NMR (DMSO-d_6) δ 3.82 (s, 3H), 7.80−7.90 (m, 2H), 7.97 (dd, J = 2, 8 Hz, 1H), 8.26 (dd, J = 2 Hz, 8 Hz, 1H). This compound (1.53 g, 7.4 mmol) was dissolved in 22 mL of THF and was treated at room temperature with 11.2 mL of an aqueous solution of LiOH (1 M) until completion of the reaction (about 3 h). The solution was then concentrated, acidified with HCl (1 N), and extracted twice with EtOAc. The organic phases were combined, dried over $MgSO_4$, filtered, and concentrated under reduced pressure to give 3-(2-nitrophenyl)propiolic acid (1.12 g, 78%): ^{1}H NMR (DMSO-d_6) δ 7.80−7.90 (m, 2H), 7.93 (dd, J = 2, 8 Hz, 1H), 8.23 (dd, J = 2, 8 Hz, 1H). This compound (1.12 g, 5.8 mmol) was dissolved in 45 mL of CH_2Cl_2 under nitrogen atmosphere. EDCI (1.12 g, 5.8 mmol), HOOBT (950 mg, 5.8 mmol), DIEA (1.8 mL, 10.6 mmol), and 1-Boc-piperazine (990 mg, 5.3 mmol) were added, and the mixture was stirred at room temperature for 20 h. The solution was concentrated under reduced pressure, then diluted with EtOAc and washed with NaOH (1 M). The organic phase was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, EtOAc/CH_2Cl_2, 5/95 to 10/90) gave 1.37 g (71%) of *tert*-butyl 4-(3-(2-nitrophenyl)propioloyl)piperazine-1-carboxylate: ^{1}H NMR (DMSO-d_6) δ 1.42 (s, 9H), 3.37 (broad s, 2H), 3.44 (broad s, 2H), 3.54 (broad s, 2H), 3.79 (broad s, 2H), 7.79 (t, J = 8 Hz, 1H), 7.86 (t, J = 8 Hz, 1H), 7.96 (d, J = 8 Hz, 1H), 8.24 (d, J = 8 Hz, 1H). This compound (1.25 g, 3.4 mmol) was dissolved in 13 mL of a saturated solution of HCl in EtOAc. The mixture was stirred at room temperature for 14 h. The white precipitate formed was recovered, and the residue was concentrated and treated again with 7 mL of a

saturated solution of HCl in EtOAc. The white precipitate formed was recovered, and the residue was concentrated and treated again with 14 mL of a saturated solution of HCl in EtOAc. The three white solids were combined to give 682 mg (66%) of 3-(2-nitrophenyl)-1-(piperazin-1-yl)prop-2-yn-1-one hydrochloride: ^{1}H NMR (DMSO-d_6) δ 3.14 (t, J = 5 Hz, 2H), 3.23 (t, J = 5 Hz, 2H), 3.79 (t, J = 5 Hz, 2H), 4.03 (t, J = 5 Hz, 2H), 7.80 (t, J = 8 Hz, 1H), 7.87 (t, J = 8 Hz, 1H), 7.97 (d, J = 8 Hz, 1H), 8.26 (d, J = 8 Hz, 1H), 9.40 (broad s, 2H). A fraction of this compound (60 mg, 0.20 mmol) was dissolved in 3 mL of CH_2Cl_2 under nitrogen atmosphere. Triethylamine (51 μL, 0.36 mmol) and 4-fluorobenzyl bromide (30 μL, 0.24 mmol) were added, and the mixture was stirred at room temperature for 24 h. More CH_2Cl_2 was added, and the mixture was washed once with NaOH (2 N), dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 0.5/4.5/95), followed by treatment with an excess of HCl in EtOAc gave 32 mg (36%) of the hydrogen chloride salt of the title compound as an off-white powder: ^{1}H NMR (DMSO-d_6) δ 2.9−3.6 (m, 5H), 3.74 (broad t, J = 11 Hz, 1H), 4.30−4.55 (m, 4H), 7.32 (t, J = 9 Hz, 2H), 7.66 (broad s, 2H), 7.81 (td, J = 1 Hz, 8 Hz, 1H), 7.87 (td, J = 1 Hz, 8 Hz, 1H), 7.97 (dd, J = 1 Hz, 8 Hz, 1H), 8.26 (dd, J = 1 Hz, 8 Hz, 1H), 11.40 (broad s, 1H); HPLC (XTerra MS) t_R = 3.60 min, 95%; MS (ESI+) m/z = 368 [MH^+].

(2*E*,4*E*)-5-(2-Nitrophenyl)penta-2,4-dienoic Acid (16).[20] NaH (270 mg, 6.77 mmol) was put in suspension in 20 mL of anhydrous THF under nitrogen atmosphere. Ethyl 2-(diethoxyphosphoryl)acetate (1.23 mL, 6.21 mmol) dissolved in 5 mL of THF was added dropwise at room temperature over 10 min. (*E*)-3-(2-Nitrophenyl)acrylaldehyde (5.2 g, 26.92 mmol) dissolved in 20 mL of THF was then added, and the mixture was stirred until completion (about 3 h). Then the reaction mixture was sequentially quenched with a few drops of water, diluted with more water, extracted twice with EtOAc, dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2/PE, 70/30) gave (2*E*,4*E*)-ethyl 5-(2-nitrophenyl)penta-2,4-dienoate (1.29 g, 92%) as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 1.24 (t, J = 7 Hz, 3H), 4.16 (q, J = 7 Hz, 2H), 6.20 (d, J = 15 Hz, 1H), 7.17 (dd, J = 11, 15 Hz, 1H), 7.35 (d, J = 15 Hz, 1H), 7.46 (dd, J = 11, 15 Hz, 1H), 7.59 (t, J = 8 Hz, 1H), 7.75 (t, J = 8 Hz, 1H), 7.89 (d, J = 8 Hz, 1H), 7.99 (d, J = 8 Hz, 1H). This compound (1.29 g, 5.2 mmol) was dissolved in 20 mL of ethanol, and a solution of aqueous KOH (1 N, 6 mL) was added. The mixture was heated at 70 °C until completion (about 1 h). The mixture was cooled to room temperature, then quenched with HCl (1 N) and extracted 3 times with CH_2Cl_2/MeOH (90/10). The organic phases were combined, dried over Na_2SO_4, filtered, and concentrated under reduced pressure to give 1.12 g of the title compound (98%) as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 6.12 (d, J = 15 Hz, 1H), 7.16 (dd, J = 11, 15 Hz, 1H), 7.30 (d, J = 15 Hz, 1H), 7.38 (dd, J = 11, 15 Hz, 1H), 7.58 (t, J = 8 Hz, 1H), 7.75 (t, J = 8 Hz, 1H), 7.90 (d, J = 8 Hz, 1H), 7.99 (d, J = 8 Hz, 1H), 12.42 (s, 1H); HPLC (XTerra MS) t_R = 4.05 min, 99.8%.

(2*E*,4*E*)-1-(4-(4-Fluorobenzyl)piperazin-1-yl)-5-(2-nitrophenyl)-penta-2,4-dien-1-one (17). Compound **16** (70 mg, 0.31 mmol) was dissolved in 3 mL of CH_2Cl_2 under nitrogen atmosphere. EDCI (67 mg, 0.35 mmol), HOOBT (57 mg, 0.35 mmol), DIEA (84 μL, 0.48 mmol), and 1-(4-fluorobenzyl)piperazine (68 mg, 0.35 mmol) were added, and the mixture was stirred for 20 h. The solution was diluted with CH_2Cl_2 and washed with NaOH (1 M). The organic phase was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 0/0/100 to 1/10/90), followed by treatment with an excess of HCl in EtOAc gave 106 mg (77%) of the hydrogen chloride salt of the title compound as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 3.08 (broad s, 2H), 3.16 (broad s, 1H), 3.38 (m, 2H), 3.59 (broad s, 1H), 4.32−4.51 (m, 4H), 6.86 (d, J = 15 Hz, 1H), 7.11 (dd, J = 11, 15 Hz, 1H), 7.25−7.34 (m, 3H), 7.58 (t, J = 7 Hz, 1H), 7.64−7.67 (m, 2H), 7.76 (t, J = 7 Hz, 1H), 7.85 (d, J = 8 Hz, 1H), 7.99 (d, J = 8 Hz, 1H), 11.29 (broads, 1H); HPLC (XTerra MS) t_R = 3.75 min, 98%; MS (ESI+) m/z = 396 [MH^+].

(*E*)-3-(2,6-Difluorophenyl)-1-(4-(4-fluorobenzyl)-2,5-dimethylpiperazin-1-yl)prop-2-en-1-one (18). 2,5-Dimethylpiperazine (1.5 g, 2.62 mmol) was dissolved in 17 mL of CH_2Cl_2 under nitrogen atmosphere. Triethylamine (551 μL, 2.62 mmol) was added, and the mixture was cooled to 0 °C. 4-Fluorobenzyl bromide (360 μL, 2.62 mmol), diluted into 3 mL of CH_2Cl_2, was added dropwise over a 30 min period. Then the cold bath was removed, and the mixture was stirred at room temperature for 1 h. More CH_2Cl_2 was then added, and the mixture was washed once successively with NaOH (1 M), water, and brine. The aqueous phases were combined and extracted twice with CH_2Cl_2. The organic phases were combined, dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 0.5/6.5/93) gave 437 mg (75%) of 1-(4-fluorobenzyl)-2,5-dimethylpiperazine as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 0.81 (d, J = 6 Hz, 3H), 1.02 (d, J = 6 Hz, 3H), 1.52 (t, J = 11 Hz, 1H), 1.91 (broad s, 1H), 2.05−2.15 (m, 1H), 2.36 (t, J = 11 Hz, 1H), 2.45−2.60 (m, 2H), 2.75 (dd, J = 3, 12 Hz, 1H), 3.00 (d, J = 14 Hz, 1H), 3.98 (d, J = 14 Hz, 1H), 7.12 (t, J=9 Hz, 2H), (dd, J=6, 8 Hz), 7.30; MS (ESI+) m/z = 223 [MH^+]. This compound (50 mg, 0.22 mmol) was dissolved in 2 mL of CH_2Cl_2 under nitrogen atmosphere. EDCI (47 mg, 0.24 mmol), HOOBT (40 mg, 0.24 mmol), DIEA (79 μL, 0.45 mmol), and (*E*)-3-(2,6-difluorophenyl)acrylic acid (50 mg, 0.27 mmol) were added, and the mixture was stirred for 20 h. The solution was diluted with CH_2Cl_2 and washed with NaOH (1 M). The organic phase was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, MeOH/CH_2Cl_2, 0/100 to 5/95) followed by treatment with an excess of HCl in EtOAc gave 82 mg (85%) of the hydrogen chloride salt of the title compound as a white powder: HPLC (XTerra MS) t_R = 3.85 min, 95%; MS (ESI+) m/z = 389 [MH^+].

(*E*)-1-(4-Fluorobenzyl)-4-(3-(2-nitrophenyl)acryloyl)piperazin-2-one (19). (*E*)-3-(2-Nitrophenyl)acrylic acid (1.27 g, 6.59 mmol) was dissolved in 45 mL of CH_2Cl_2 under nitrogen atmosphere. EDCI (1.26 g, 6.59 mmol), HOOBT (1.07 g, 6.59 mmol), DIEA (2 mL, 11.9 mmol), and piperazin-2-one (600 mg, 5.99 mmol) were added, and the mixture was stirred for 20 h. The solution was diluted with CH_2Cl_2 and washed with NaOH (1 M). The organic phase was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 0.5/2.5/97 to 0.5/4.5/95) gave 1.35 g (75%) of (*E*)-4-(3-(2-nitrophenyl)acryloyl)piperazin-2-one: ^{1}H NMR (DMSO-d_6) δ 3.22 (broad s, 1H), 3.30 (broad s, 1H), 3.72 (broad s, 1H), 3.90 (broad s, 1H), 4.07 (s, 1H), 4.33 (s, 1H), 7.24−7.32 (m, 1H), 7.65 (t, J=8 Hz, 1H), 7.75−7.80 (m, 2H), 8.0−8.2 (m, 3H). This compound (140 mg, 0.50 mmol) was dissolved in 15 mL of THF, and grounded KOH (29 mg, 0.51 mmol) was added along with 3 drops of water. 4-Fluorobenzyl bromide (53 μL, 0.42 mmol) was then added. The mixture was stirred at room temperature for 24 h, heated under reflux conditions for 4 h, and then concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 0.5/2.5/97 to 0.5/4.5/95) gave 31 mg (15%) of the title compound as an off-white powder: ^{1}H NMR (DMSO-d_6) δ 3.35 (broad s, 2H), 3.79 (broad s, 1H), 3.96 (broad s, 1H), 4.23 (s, 1H), 4.50 (s, 1H), 4.56 (s, 2H), 7.17 (t, J = 8 Hz, 2H), 7.26 (d, J = 15 Hz, 1H), 7.32 (dd, J = 6, 8 Hz, 2H), 7.64 (t, J = 8 Hz, 1H), 7.75−7.84 (m, 2H), 8.00−8.15 (m, 2H); HPLC (XTerra MS) t_R = 4.36 min, 95%; MS (ESI+) m/z = 384 [MH^+].

(*E*)-1-(4-(4-Fluorobenzyl)-1,4-diazepan-1-yl)-3-(2-nitrophenyl)prop-2-en-1-one (20). *tert*-Butyl 1,4-diazepane-1-carboxylate (275 mg, 1.37 mmol) was dissolved in 7 mL of CH_2Cl_2 under nitrogen atmosphere. Triethylamine (288 μL, 1.94 mmol) was added followed by 4-fluorobenzyl bromide (286 mL, 1.51 mmol). The mixture was stirred at room temperature for 18 h. More CH_2Cl_2 was added, and the mixture was washed once successively with NaOH (1 M), water, and brine. The organic phase was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, CH_2Cl_2) gave 324 mg (77%) of *tert*-butyl 4-(4-fluorobenzyl)-1,4-diazepane-1-carboxylate as a colorless oil: ^{1}H NMR (DMSO-d_6) δ 1.40 (s, 9H), 1.69 (broad s, 2H), 2.45−2.60 (m, 4H), 3.35 (broad s, 4H), 3.58 (s, 2H), 7.13 (t, J = 8 Hz, 2H), 7.33 (dd, J = 6, 8 Hz, 2H); MS (ESI+) m/z = 309 [MH^+]. This compound (324 mg, 1.05 mmol) was dissolved in 8.5 mL of toluene under nitrogen atmosphere. TFA (2.4 mL, 32 mml) was added, and the reaction was stirred until completion (about 1 h). It was then concentrated under reduced pressure, diluted into CH_2Cl_2, washed once successively with NaOH (1 M), water, and brine, then dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 1/9/90) gave 192 mg (88%) of 1-(4-fluorobenzyl)-1,4-diazepane as a colorless oil: ^{1}H NMR (DMSO-d_6) δ 1.64 (pentet, J = 6 Hz, 2H), 2.53 (broad s, 2H), 2.58 (t, J = 6 Hz, 2H), 2.71 (t, J = 5 Hz, 2H), 2.79 (t, J = 8 Hz, 2H), 3.1−3.5 (m, 1H, NH), 3.58 (s, 2H), 7.12 (t, J = 8 Hz, 2H), 7.34 (dd, J = 6, 8 Hz, 2H); MS (ESI+) m/z = 209 [MH^+]. This compound (50 mg, 0.24 mmol) was dissolved in 2 mL of CH_2Cl_2 under nitrogen atmosphere. EDCI (51 mg, 0.26 mmol), HOOBT (43 mg, 0.26 mmol), DIEA (84 μL, 0.48 mmol), and (*E*)-3-(2-nitrophenyl)acrylic acid (56 mg, 0.29 mmol) were added, and the mixture was stirred for 20 h. The solution was diluted with CH_2Cl_2 and washed with NaOH (1 M). The organic phase was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, MeOH/CH_2Cl_2, 0/100 to 10/90) followed by treatment with an excess of HCl in EtOAc gave 81 mg (80%) of the hydrogen chloride salt of the title compound as a white powder: ^{1}H NMR (DMSO-d_6) δ 2.12 (broad s, 1H), 2.37 (broad s, 1H), 3.0−3.2 (m, 2H), 3.3−3.6 (m, 3H), 3.7−3.9 (m, 2H), 4.15−4.4 (m, 3H), 7.22 (m, 1H), 7.31 (t, J = 9 Hz, 2H), 7.60−7.75 (m, 3H), 7.75−7.81 (m, 2H), 8.04 (td, J = 1 Hz, 8 Hz, 2H), 10.90 (s, 1H); HPLC (XTerra MS) t_R = 3.64 min, 99%; MS (ESI+) m/z = 384 [MH^+].

(*E*)-1-(4-(4-Fluorobenzylamino)piperidin-1-yl)-3-(2-nitrophenyl)prop-2-en-1-one (21). (*E*)-3-(2-Nitrophenyl)acrylic acid (2 g, 10.3 mmol) was dissolved in 75 mL of CH_2Cl_2 under nitrogen atmosphere. EDCI (1.98 g, 10.3 mmol), HOOBT (1.68 g, 10.3 mmol), DIEA (4.9 mL, 28 mmol), and the TFA salt of piperidin-4-one (2.0 g, 9.38 mmol) were added, and the mixture was stirred for 20 h. The solution was diluted with CH_2Cl_2 and washed with NaOH (1 M) and brine. The organic phase was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, acetone/CH_2Cl_2, 5/95 to 10/90) gave 2.22 g (86%) of (*E*)-1-(3-(2-nitrophenyl)acryloyl)piperidin-4-one: ^{1}H NMR (DMSO-d_6) δ 2.4−2.5 (m, 4H), 3.85 (t, J = 6 Hz, 2H), 4.00 (t, J = 6 Hz, 2H), 7.17 (d, J = 15 Hz, 1H), 7.64 (t, J = 8 Hz, 1H), 7.75−7.80 (m, 2H), 8.06 (t, J = 8 Hz, 2H); HPLC (XTerra MS) t_R = 3.51 min, 98%; MS (ESI+) m/z = 275 [MH^+]. A fraction of this compound (804 mg, 2.93 mmol) was dissolved in 13 mL of 1,2-dichloroethane under nitrogen atmosphere. 4-Fluorobenzylamine (337 μL, 2.93 mmol) was added, followed by 918 μL of acetic acid. The mixture was stirred for half an hour at room temperature prior to the addition of $NaBH(OAc)_3$ (684 mg, 3.2 mmol). After 18 h the reaction was quenched with 30 mL of water, basified with saturated $NaHCO_3$, and extracted with CH_2Cl_2. The organic phase was dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 1/9/90) gave 676 mg (60%) of the title compound as an off-white powder: ^{1}H NMR (DMSO-d_6) δ 1.57 (broad q, J = 10 Hz, 2H), 2.19 (broad d, J = 10 Hz, 2H), 2.72 (broad t, J = 12 Hz, 1H), 3.13 (broad t, J = 12 Hz, 1H), 3.33 (broad s, 1H), 4.19 (broad s, 2H), 4.40 (broad d, J = 14 Hz, 1H), 4.55 (broad d, J = 14 Hz, 1H), 7.29 (t, J = 9 Hz, 2H), 7.34 (d, J = 15 Hz, 1H), 7.60−7.69 (m, 3H), 7.73 (d, J = 15 Hz, 1H), 7.78 (t, J = 8 Hz, 1H), 8.05 (td, J = 1 Hz, 7 Hz, 2H), 9.40 (broad s, 2H); HPLC (XTerra MS) t_R = 3.60 min, 98%; MS (ESI+) m/z = 384 [MH^+].

***tert*-Butyl 4-(Methoxy(methyl)carbamoyl)piperidine-1-carboxylate (24).** Synthesis of the title compound has already been reported.[21] Boc protection of piperidine-4-carboxylic acid was performed using the described protocol, but a different methodology was used for the amidation step. Therefore, 1-(*tert*-butoxycarbonyl)piperidine-4-carboxylic acid (8.63 g, 37.6 mmol) was dissolved in 250 mL of DMF under nitrogen atmosphere. EDCI (7.93 g, 41.3 mmol), HOOBT (6.75 g, 41.3 mmol), DIEA (19.7 mL, 113 mmol), and MeONHMe·HCl (4.1 g, 41.3 mmol) were added, and the mixture was stirred for 24 h at 50 °C. The solution was cooled to room temperature, then hydrolyzed with NaOH (1 M) and extracted twice with CH_2Cl_2. The organic phase was washed once with HCl (1 N), dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, EtOAc/PE, 30/70 to 60/40) gave 9.05 g (88%) of the title compound as a yellow oil: ^{1}H NMR (DMSO-d_6) δ 1.3−1.45 (m, 11H), 1.63 (broad d, J = 12 Hz, 2H), 2.6−2.95 (m, 3H), 3.09 (s, 3H), 3.68 (s, 3H), 3.94 (broad d, J = 12 Hz, 2H).

1-(4-Fluorobenzyl)-*N*-methoxy-*N*-methylpiperidine-4-carboxamide (25). Compound **24** (2.95 g, 10.8 mmol) was dissolved in 36 mL of toluene under nitrogen atmosphere and cooled at 0 °C prior to the addition of 18 mL of TFA. The cooling bath was removed, and the mixture was stirred for 1 h at room temperature. The solvent was removed under reduced pressure, and the residue was dissolved in CH_2Cl_2, then quenched by the addition of NaOH (1 M). The aqueous phase was extracted 6 times with CH_2Cl_2. The organic phase was dried over $MgSO_4$, filtered, and concentrated under reduced pressure to give 1.38 g (74%) of *N*-methoxy-*N*-methylpiperidine-4-carboxamide as a pale-yellowish oil: ^{1}H NMR (DMSO-d_6) δ 1.42 (ddd, J = 4, 12, 24 Hz, 2H), 1.55 (broad d, J = 11 Hz, 2H), 2.4−2.6 (m, 2H), 2.94 (broad d, J = 12 Hz, 2H), 2.9−3.4 (m, 1H, NH), 3.08 (s, 3H), 3.66 (s, 3H); MS (ESI+) m/z = 173 [MH^+]. A fraction of this compound (750 mg, 4.3 mmol) was dissolved in 20 mL of 1,2-dichloroethane under nitrogen atmosphere. 4-Fluorobenzaldehyde (595 mg, 4.8 mmol) was added, followed by 1.4 mL of acetic acid. The mixture was stirred for half an hour at room temperature prior to the addition of $NaBH(OAc)_3$ (1.02 g, 4.8 mmol). After 24 h of reaction, the starting material was not fully consumed; therefore, an aliquot of 4-fluorobenzaldehyde (216 mg) and $NaBH(OAc)_3$ (370 mg) was added. Twenty-four hours later, the reaction was quenched with 30 mL of water, basified with NaOH (1 M), and extracted 3 times with CH_2Cl_2. The organic phase was dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, MeOH/CH_2Cl_2, 0/100 to 5/95) gave 940 mg (77%) of the title compound as a yellow oil: ^{1}H NMR (DMSO-d_6) δ 1.5−1.75 (m, 4H), 1.95 (td, J = 2, 11 Hz, 2H), 2.61 (broad t, 1H), 2.80 (d, J = 11 Hz, 2H), 3.08 (s, 3H), 3.43 (s, 2H), 3.66 (s, 3H), 7.13 (t, J = 8 Hz, 2H), 7.31 (dd, J = 8, 6 Hz, 2H); HPLC (XTerra MS) t_R = 2.97 min, 100%; MS (ESI+) m/z = 281 [MH^+].

(*E*)-3-(2-Chlorophenyl)-1-(1-(4-fluorobenzyl)piperidin-4-yl)-prop-2-en-1-one (27). Diethyl methylphosphonate (1.1 mL, 7.8 mmol) was dissolved in 20 mL of anhydrous THF under nitrogen atmosphere and cooled at −50 °C prior to the dropwise addition *n*-buthyllithium (1.6 M in hexane, 5.0 mL, 8 mmol).

The resulting white suspension was stirred for 1 h between −40 and −50 °C. In a separated flask, compound **25** (880 mg, 3.1 mmol) was dissolved in 11 mL of anhydrous THF under nitrogen atmosphere. This clear solution was slowly added into the above cold suspension while maintaining the temperature between −45 and −43 °C. The resulting reaction mixture was stirred for 1 h at −45 °C, then quenched with a few drops of water prior to the removal of the cooling bath. It was then diluted with 20 mL of HCl (1 N) and washed 3 times with EtOAc. The aqueous phase was brought to pH 10 with concentrated NaOH and was extracted 3 times with CH_2Cl_2. The organic phases were combined, dried over $MgSO_4$, filtered, and concentrated under reduced pressure. The residue (**26**) was taken up into 25 mL of CH_3CN under nitrogen atmosphere. 2-Chlorobenzaldehyde (700 μL, 6.2 mmol) and potassium carbonate (850 mg, 6.2 mmol) were added, and the mixture was stirred at room temperature until completion (about 24 h). The reaction mixture was filtered and concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, NH_4OH/MeOH/CH_2Cl_2, 0/0/100 to 0.5/4.5/95) gave 611 mg (68%) of the title compound as a colorless oil: ^{1}H NMR (DMSO-d_6) δ 1.52 (ddd, J = 3, 12, 24 Hz, 2H), 1.83 (broad d, J = 12 Hz, 2H), 2.02 (td, J = 2, 11 Hz, 2H), 2.68 (tt, J = 4, 11 Hz, 1H), 2.82 (broad d, J = 12 Hz, 2H), 3.46 (s, 2H), 7.05−7.25 (m, 3H), 7.33 (dd, J = 6, 8 Hz, 2H), 7.1−7.46 (m, 2H), 7.55 (dd, J = 2, 8 Hz, 1H), 7.82 (d, J = 16 Hz, 1H), 7.98 (dd, J = 2, 8 Hz, 1H); HPLC (XTerra MS) t_R = 4.10 min, 100%; MS (ESI+) m/z = 358 [MH^+].

(***E***)**-1-[4-(3-Chlorophenyl)piperazin-1-yl]-3-(2-nitrophenyl)propenone (28).** According to compound **13** synthesis described above, compound **28** was isolated in 75% yield as an HCl salt as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 3.24 (broad s, 4H), 3.72 (broad s, 2H), 3.87 (broad s, 2H), 6.82 (d, J = 8 Hz, 1H), 6.95 (d, J = 8 Hz, 1H), 7.00 (s, 1H), 7.24 (t, J = 8 Hz, 1H), 7.35 (d, J = 15 Hz, 1H), 7.64 (t, J = 8 Hz, 1H), 7.74−7.87 (m, 2H), 8.05 (t, J = 8 Hz, 1H); HPLC (XTerra MS) t_R = 5.18 min, 98%; MS (ESI) m/z = 372 [MH^+]. Anal. Calcd for $C_{19}H_{18}N_3O_3Cl_1$·HCl: C, 55.89; H, 4.69; N, 10.29. Found: C, 55.77; H, 4.77; N, 10.03.

(***E***)**-3-(2-Nitrophenyl)-1-[4-(4-cyanophenyl)piperazin-1-yl]propenone (29).** According to compound **13** synthesis described above, compound **29** was isolated in 95% yield as a red-brown powder: ^{1}H NMR (DMSO-d_6) δ 3.43 (broad s, 4H), 3.72 (broad s, 2H), 3.88 (broad s, 2H), 7.05 (d, J = 8 Hz, 1H), 7.34 (d, J = 15 Hz, 1H), 7.60−7.66 (m, 3H), 8.05 (t, J = 8 Hz, 2H); HPLC (Symmetry) t_R = 4.87 min, 98%; MS (ESI+) m/z = 363 [MH^+].

(***E***)**-3-(2-Nitrophenyl)-1-(4-pyridin-4-ylmethylpiperazin-1-yl)-propenone (30).** According to representative procedure A described above, compound **30** was isolated in 77% yield as an orange powder: ^{1}H NMR (DMSO-d_6) δ 2.30−2.45 (m, 4H), 3.56 (s, 2H), 3.58 (broad s, 2H), 3.71 (broad s, 2H), 7.28 (d, J = 15 Hz, 1H), 7.38 (dd, J = 4 Hz, 8 Hz, 1H), 7.62 (td, J = 1 Hz, 8 Hz, 1H), 7.70−7.79 (m, 3H), 8.00−8.05 (m, 2H), 8.45−8.55 (m, 2H); HPLC (Symmetry) t_R = 2.94 min, 95%; MS (ESI+) m/z = 353 [MH^+].

(***E***)**-1-(4-Cycloheptanecarbonylpiperazin-1-yl)-3-(2-nitrophenyl)propenone (31).** According to representative procedure A described above, compound **31** was isolated in 92% yield as a white powder: ^{1}H NMR (DMSO-d_6) δ 1.44−1.56 (m, 8H), 1.67−1.72 (m, 4H), 2.74−2.77 (m, 1H), 3.53−3.69 (broad m, 6H), 3.69−3.74 (broad m, 2H), 7.29 (d, J = 15 Hz, 1H), 7.64 (t, J = 8 Hz, 1H), 7.75 (d, J = 15 Hz, 1H), 7.79 (d, J = 8 Hz, 1H), 8.04 (d, J = 8 Hz, 1H); HPLC (XTerra MS) t_R = 4.77 min, 98%; MS (ESI+) m/z = 386 [MH^+]. Anal. Calcd for $C_{21}H_{27}N_3O_4$: C, 65.44; H, 7.06; N, 10.90. Found: C, 65.49; H, 7.21; N, 10.84.

(***E***)**-1-[4-(2-Fluorobenzenesulfonyl)piperazin-1-yl]-3-(2-nitrophenyl)propenone (32).** According to representative procedure A described above, compound **32** was isolated in 50% yield as an off-white powder: ^{1}H NMR (DMSO-d_6) δ 3.10 (broad s, 4H), 3.67 (broad s, 2H), 3.81 (broad s, 2H), 7.24 (d, J = 15 Hz, 1H), 7.45 (t, J = 8 Hz, 1H), 7.51 (td, J = 2 Hz, 8 Hz, 1H), 7.64 (td, J = 1 Hz, 8 Hz, 1H), 7.70 (d, J = 15 Hz, 1H), 7.74−7.82 (m, 3H), 7.99 (d, J = 7 Hz, 1H), 8.02 (dd, J = 1 Hz, 8 Hz, 1H); HPLC (XTerra MS) t_R = 4.78 min, 95%; MS (ESI+) m/z = 420 [MH^+].

(***E***)**-4-(3-(2-Nitrophenyl)acryloyl)piperazine-1-carboxylic Acid Cyclohexylamide (33).** According to representative procedure A described above, compound **33** was isolated in 66% yield as a white powder: ^{1}H NMR (DMSO-d_6) δ 1.00−1.30 (m, 5H), 1.50−1.80 (m, 5H), 3.33 (broad s, 4H), 3.41 (broad s, 1H), 3.54 (broad s, 2H), 3.67 (broad s, 2H), 6.28 (d, J = 8 Hz, 1H), 7.29 (d, J = 15 Hz, 1H), 7.64 (td, J = 1 Hz, 9 Hz, 1H), 7.72−7.80 (m, 2H), 8.02 (d, J = 8 Hz, 2H); HPLC (XTerra MS) t_R = 4.38 min, 99%; MS (ESI+) m/z = 381 [MH^+].

(**2*E*,4*E*)-1-(4-Cyclopentylpiperazin-1-yl)-5-(2-nitrophenyl)penta-2,4-dien-1-one (34).** According to compound **17** synthesis described above, compound **34** was isolated in 87% yield as an HCl salt and a yellow powder: ^{1}H NMR (DMSO-d_6) δ 1.56 (broad s, 2H), 1.73 (broad s, 4H), 2.05 (broad s, 2H), 2.85−3.25 (m, 3H), 3.4−3.75 (m, 4H), 4.28 (broad s, 1H), 4.53 (broad s, 1H), 6.90 (d, J = 15 Hz, 1H), 7.13 (dd, J = 11 Hz, 15 Hz, 1H), 7.27 (d, J = 15 Hz, 1H), 7.34 (dd, J = 11 Hz, 15 Hz, 1H), 7.58 (t, J = 8 Hz, 1H), 7.75 (t, J = 8 Hz, 1H), 7.86 (d, J = 8 Hz, 1H), 7.99 (d, J = 8 Hz, 1H), 10.87 (broad s, 1H, NH^+); HPLC (XTerra MS) t_R = 3.62 min, 99%; MS (ESI+) m/z = 356 [MH^+]. Anal. Calcd for $C_{20}H_{25}N_3O_3$·HCl: C, 61.30; H, 6.69; N, 10.72. Found: C, 60.91; H, 6.72; N, 10.94.

(**2*E*,4*E*)-5-(2-Nitrophenyl)-1-(4-phenylpiperazin-1-yl)penta-2,4-dien-1-one (35).** According to compound **17** synthesis described above, compound **35** was isolated in 70% yield as an HCl salt and a yellow powder: ^{1}H NMR (DMSO-d_6) δ 3.26 (broad s, 4H), 3.82 (broad s, 4H), 5.4−6.4 (broad m, 1H, NH^+), 6.9−7.0 (m, 2H), 7.05−7.4 (m, 7H), 7.58 (t, J = 8 Hz, 1H), 7.75 (t, J = 8 Hz, 1H), 7.87 (d, J = 8 Hz, 1H), 7.99 (d, J = 8 Hz, 1H); HPLC (XTerra MS) t_R = 4.60 min, 97.8%; MS (ESI+) m/z = 364 [MH^+]. Anal. Calcd for $C_{21}H_{21}N_3O_3$·HCl: C, 63.08; H, 5.55; N, 10.51. Found: C, 63.17; H, 5.61; N, 10.81.

2-((1*E*,3*E*)-5-Oxo-5-(4-(pyridin-2-yl)piperazin-1-yl)penta-1,3-dienyl)benzonitrile (36). Bromobenzonitrile (910 mg, 5 mmol) was dissolved in 10 mL of dimethylacetamide under nitrogen atmosphere. DIEA (1 mL), 3,3-diethoxyprop-1-ene (2.3 mL, 15 mmol), and Herrmann's catalyst (90 mg, 0.1 mmol) were added, and the mixture was heated at 90 °C for 18 h. The mixture was then cooled to room temperature, diluted with ether, washed with HCl (1 N), dried with Na_2SO_4, filtered, and concentrated under reduced pressure. Flash column chromatography of the residue (SiO_2, CH_2Cl_2) gave 314 mg of (*E*)-2-(3-oxoprop-1-enyl)benzonitrile partially purified. ^{1}H NMR (DMSO-d_6) δ 7.07 (dd, J = 7.5, 15 Hz, 1H), 7.67 (t, J = 8 Hz, 1H), 7.81 (t, J = 8 Hz, 1H), 7.87 (d, J = 15 Hz, 1H), 7.98 (d, J = 8 Hz, 1H), 8.13 (d, J = 8 Hz, 1H), 9.82 (d, J = 8 Hz, 1H). This aldehyde was carried out in the following step without further purification. This compound (314 mg) was dissolved in 10 mL of toluene under nitrogen atmosphere, and (carbethoxymethylene)triphenylphosphorane (740 mg, 2.1 mmol) was added. The mixture was stirred under reflux conditions for 4 h, then concentrated under reduced pressure. Purification of the crude material by flash column chromatography (SiO_2, EtOAc/PE, 10/90) gave 383 mg (33%, two steps) of a mixture of (2*E*,4*E*)- and (2*E*,4*Z*)-ethyl 5-(2-cyanophenyl)penta-2,4-dienoates (80/20). This mixture was dissolved in 4 mL of CH_3CN and treated with a catalytic amount of iodine (4 mg) for 3 h at room temperature. The mixture was then concentrated under reduced pressure, diluted with CH_2Cl_2, washed with an aqueous solution of Na_2SO_3 (1%), dried over $MgSO_4$, filtered, and concentrated under reduced pressure to give 333 mg (87%) of (2*E*,4*E*)-ethyl 5-(2-cyanophenyl)penta-2,4-dienoate: ^{1}H NMR (DMSO-d_6) δ 1.25 (t, J = 7 Hz, 3H), 4.16 (q, J = 7 Hz, 2H), 6.21 (d, J = 15 Hz, 1H), 7.25−7.40 (m, 2H), 7.45−7.56 (m, 2H), 7.74 (t, J = 8 Hz, 1H), 7.87 (d, J = 8 Hz, 1H), 7.96 (d, J = 8 Hz, 1H). This compound (2.0 g, 8.8 mmol) was dissolved in 50 mL of ethanol, and a

solution of aqueous KOH (1 N, 13.2 mL) was added. The mixture was heated under refluxing conditions until completion (about 1.5 h). The mixture was cooled to room temperature, then quenched with HCl (1 N), filtered, and concentrated under reduced pressure to give 1.63 g of (2*E*,4*E*) 5-(2-cyanophenyl)penta-2,4-dienoic acid (93%): ^{1}H NMR (DMSO-d_6) δ 6.14 (d, J = 15 Hz, 1H), 7.22 (d, J = 15 Hz, 1H), 7.30−7.48 (m, 2H), 7.52 (t, J = 8 Hz, 1H), 7.73 (t, J = 8 Hz, 1H), 7.86 (d, J = 8 Hz, 1H), 7.96 (d, J = 8 Hz, 1H); HPLC (XTerra MS) t_R = 4.00 min, 95%; MS (ESI−) m/z = 198 [M − H]. This compound was used according to compound **17** synthesis described above. The HCl salt of compound **36** was obtained in 78% yield as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 3.81 (broad s, 8H), 3.60−4.30 (broad m, 1H), 6.9−7.0 (m, 2H), 7.10−7.45 (m, 4H), 7.51 (t, J=8 Hz, 1H), 7.75 (t, J = 8 Hz, 1H), 7.87 (d, J = 8 Hz, 1H), 7.91 (d, J = 8 Hz, 1H), 7.97 (t, J = 5 Hz, 1H), 8.07 (d, J = 5 Hz, 1H); HPLC (XTerra MS) t_R = 3.45 min, 99%; MS (ESI) m/z = 345 [MH$^+$]. Anal. Calcd for $C_{21}H_{20}N_4O_1 \cdot HCl \cdot 1.5H_2O$: C, 61.84; H, 5.93; N, 13.74. Found: C, 61.65; H, 5.60; N, 13.45.

2-[(*E*)-3-(4-Cycloheptanecarbonylpiperazin-1-yl)-3-oxopropenyl]benzonitrile (37). According to representative procedure A described above, compound **37** was isolated in 61% yield as a white powder: ^{1}H NMR (DMSO-d_6) δ 1.44−1.53 (m, 8H), 1.67−1.73 (m, 4H), 2.75 (broad s, 1H), 3.53−3.70 (broad m, 6H), 3.70−3.75 (broad m, 2H), 7.53 (d, J = 15 Hz, 1H), 7.58 (t, J = 8 Hz, 1H), 7.73 (d, J = 15 Hz, 1H), 7.78 (d, J = 8 Hz, 1H), 7.90 (d, J = 8 Hz, 1H), 8.18 (broad s, 1H); HPLC (XTerra MS) t_R = 4.60 min, 98%; MS (ESI+) m/z = 366 [MH$^+$]. Anal. Calcd for $C_{22}H_{27}N_3O_2 \cdot 0.6H_2O$: C, 70.22; H, 7.55; N, 11.17. Found: C, 70.28; H, 7.57; N, 11.07.

(*E*)-1-(4-Cycloheptanecarbonylpiperazin-1-yl)-3-(2,6-difluorophenyl)propenone (38). According to representative procedure A described above, compound **38** was isolated in 27% yield as a white powder: ^{1}H NMR (DMSO-d_6) δ 1.44−1.55 (m, 8H), 1.67−1.72 (m, 4H), 2.67 (broad s, 1H), 3.51−3.61 (broad m, 8H), 7.20−7.27 (m, 3H), 7.48−7.53 (m, 2H); HPLC (XTerra MS) t_R = 4.86 min, 98%; MS (ESI+) m/z = 377 [MH$^+$]. Anal. Calcd for $C_{21}H_{26}N_2F_2O_2$: C, 67.00; H, 6.96; N, 7.44. Found: C, 66.82; H, 7.13; N, 7.44.

(*E*)-3-(2-Chlorophenyl)-1-[4-(4-fluorobenzyl)piperazin-1-yl]-propenone (39). According to representative procedure A described above, compound **39** was isolated in 69% yield as an HCl salt as a white powder: ^{1}H NMR (DMSO-d_6) δ 3.01−3.17 (m, 3H), 3.34−3.38 (m, 2H), 3.59 (broad t, 1H), 4.34 (broad s, 1H), 4.53 (broad t, 1H), 7.31−7.35 (m, 2H), 7.41−7.44 (m, 1H), 7.53−7.55 (m, 1H), 7.64 (broad s, 2H), 7.84 (d, J = 15 Hz, 1H), 7.99−8.01 (m, 1H), 11.1 (s, 1H); HPLC (XTerra MS) t_R = 3.73 min, 99%; MS (ESI+) m/z = 359 [MH$^+$]. Anal. Calcd for $C_{20}H_{20}N_2Cl_1F_1O_1 \cdot 0.9HCl \cdot 0.2H_2O$: C, 60.78; H, 5.43; N, 7.09. Found: C, 60.36; H, 5.44; N, 7.02.

(*E*)-1-(4-Cyclohexylmethylpiperazin-1-yl)-3-(2-nitrophenyl)-propenone (40). According to representative procedure A described above, compound **40** was isolated in 73% yield as an HCl salt as a yellow powder: ^{1}H NMR (DMSO-d_6) δ 0.92−1.00 (m, 2H), 1.07−1.30 (m, 3H), 1.61−1.70 (m, 3H), 1.81 (broad s, 3H), 2.09−3.03 (m, 3H), 3.27−3.30 (m, 1H), 3.53 (broad s, 2H), 3.70 (broad t, 1H), 4.48 (t, J = 14 Hz, 2H), 7.31 (d, J = 15 Hz, 1H), 7.66 (t, J = 8 Hz, 1H), 7.78 (m, 2H), 8.03 (d, J = 8 Hz, 1H), 8.05 (d, J = 8 Hz, 1H), 10.18 (s, 1H). HPLC (XTerra MS) t_R = 3.65 min, 99%; MS (ESI+) m/z = 358 [MH$^+$]. Anal. Calcd for $C_{20}H_{27}N_3O_3 \cdot HCl \cdot 0.6H_2O$: C, 60.68; H, 7.17; N, 10.43. Found: C, 60.30; H, 7.19; N, 10.36.

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Supporting Information Available: Experimental protocols for the biological assays. This material is available free of charge via the Internet at http://pubs.acs.org.

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