Design and Synthesis of Novel Sulfonamide-Containing Bradykinin hB_2 Receptor Antagonists. 2. Synthesis and Structure−Activity Relationships of α,α-Cycloalkylglycine Sulfonamides

Daniela Fattori,*,† Cristina Rossi,† Christopher I. Fincham,† Valerio Caciagli,† Fernando Catrambone,† Piero D'Andrea,† Patrizia Felicetti,† Martina Gensini,‡ Elena Marastoni,† Rossano Nannicini,‡ Marielle Paris,† Rosa Terracciano,† Alessandro Bressan,† Sandro Giuliani,‡ Carlo A. Maggi,‡ Stefania Meini,‡ Claudio Valenti,‡ and Laura Quartara‡

Menarini Ricerche, Via Tito Speri 10, 00040 Pomezia (Rome), Italy, and Menarini Ricerche, Via Rismondo 12A, 50131 Florence, Italy

Received October 3, 2006

Recently we reported on the design and synthesis of a novel class of selective nonpeptide bradykinin (BK) B_2 receptor antagonists (*J. Med. Chem.* 2006, 3602−3613). This work led to the discovery of MEN 15442, an antagonist with subnanomolar affinity for the human B2 receptor (hB2R), which also displayed significant and prolonged activity in vivo (for up to 210 min) against BK-induced bronchoconstriction in the guinea-pig at a dose of 300 nmol/kg (it), while demonstrating only a slight effect on BK-induced hypotension. Here we describe the further optimization of this series of compounds aimed at maximizing the effect on bronchoconstriction and minimizing the effect on hypotension, with a view to developing topically delivered drugs for airway diseases. The work led to the discovery of MEN 16132, a compound which, after intratracheal or aerosol administration, inhibited, in a dose-dependent manner, BK-induced bronchoconstricton in the airways, while showing minimal systemic activity. This compound was selected as a preclinical candidate for the topical treatment of airway diseases involving kinin B2 receptor stimulation.

Introduction

Bradykinin is a nonapeptide (BK: Arg^1-Pro^2-Pro^3-Gly^4-Phe^5-Ser^6-Pro^7-Phe^8-Arg^9) generated in plasma and tissue by activation of kininogens and exerts its effects through the constitutively expressed B_2 receptor or through the induced B_1 receptor.

In humans and guinea pigs, the kinin B_2 receptors are expressed in the upper and lower airways.[1,2] In humans, BK induces potent bronchoconstriction and cough when inhaled by asthmatic patients[3] and rhinitis-like symptoms when instilled into the nose.[4] Furthermore, BK is generated in human nasal secretions during rhinovirus infections[5] and allergic rhinitis.[6] On the basis of these findings, a potential therapeutic role for kinin B_2 receptor antagonists has been hypothesized in the treatment of airway inflammatory pathologies, such as chronic bronchial asthma[7] or perennial and seasonal allergic rhinitis.[8] In addition, a B_2 receptor is expressed also in the cardiovascular system, where kinins exert protective effects. Several studies have provided evidence suggesting that BK contributes to the antihypertensive and cardioprotective effects of angiotensin-converting enzyme inhibitors[9] and angiotensin II antagonists.[10] On the basis of these considerations, it would be ideal to block B_2 receptors in the airways without affecting them at the cardiovascular level.

We argued that the development of new, selective, human B_2 receptor (hB_2R) antagonists for topical administration would be the best way of achieving the goal of airway selectivity. With this aim in mind, we undertook work on the sulfonamide moiety of Anatibant, the most advanced hB_2R antagonist, which led to the discovery of MEN 15422 (Chart 1).[11] In an in vivo assay in the guinea pig, the latter, at a dose of 300 nmol/kg, was able to lower the residual bronchoconstriction activity (Σ%)[12] to 39 following a challenge with BK (10 nmol/kg iv); at the same dose Anatibant had a Σ% of 73 (Figure 1). However, at this dose, MEN 15422 had still a noticeable effect on BK-induced hypotension (Σ% 79), indicating that the further improvement is needed. This current paper describes our efforts to optimize the pharmacodynamic properties of our lead compound (lower dose, greater airway selectivity).

As a general working strategy, all compounds showing a $pK_i \geq 9.0$ in a hB_2R binding assay were assessed for their ability to antagonize a BK-induced functional response, that is, the accumulation of inositolphosphate (IP) as an index of the activation of the hB_2R expressed in CHO cells (pA_2). Promising candidates were subsequently screened in a bioassay for their ability to antagonize the BK-induced contraction of the guinea pig ileum longitudinal smooth muscle (GPI) to avoid any possible problems arising from species related selectivity when performing in vivo tests in the guinea pig. Compounds having a pA_2 (IP) ≥ 8.7 and a pA_2 (GPI) ≥ 9 were then tested in vivo, via intratracheal administration, for their effect on BK-induced bronchoconstriction and hypotension.

Chemistry

The compounds described in this study are shown in Tables 1−3, and the synthetic methods for their preparation are outlined in Schemes 1−7. Chlorosulfonic acid **1**[11] was subjected to radical bromination under standard conditions (NBS, AIBN) to obtain benzylbromide **2**. Formation of the sulfonamides with the corresponding α,α-aminoacids was performed in two ways, either via a classical base-catalyzed reaction with the amino acid methyl ester or by the addition of the amino acid pretreated with BSA (*N,O-bis*-trimethylsilylacetamide). The resulting

* To whom correspondence should be addressed. Phone: +39 06 91184522. Fax: +39 06 9106137. E-mail: dfattori@menarini-ricerche.it.
† Menarini Ricerche, Pomezia (Rome), Italy.
‡ Menarini Ricerche, Florence, Italy.

[a] Abbreviations: AcOH, acetic acid; AcCN, acetonitrile; BSA, *N,O*-bis-trimethylsilylacetamide; DIAD, diisopropyl azodicarboxylate; DIPEA, di-isopropylethyl amine; DCM, dichloromethane; DMF, dimethylformamide; DMSO, dimethylsulfoxide; EDAC, *N*-(3-dimethylaminopropyl)-*N*′-ethyl-carbodiimide; EtOAc, ethyl acetate; Et_2O, diethyl ether; EtOH, ethanol; HOAt, 7-aza-1-hydroxybenzotriazole; HOBt, 1-hydroxybenzotriazole; MeOH, methanol; NBS, *N*-bromosuccinimide; PyBOP, (benzotriazol-1-yloxy)-tripyrrolidinophosphonium hexafluorophosphate; TFA, trifluoroacetic acid; THF, tetrahydrofuran; TMSCl, trimethylsilyl chloride.

10.1021/jm061143k CCC: $37.00
Published on Web 01/06/2007

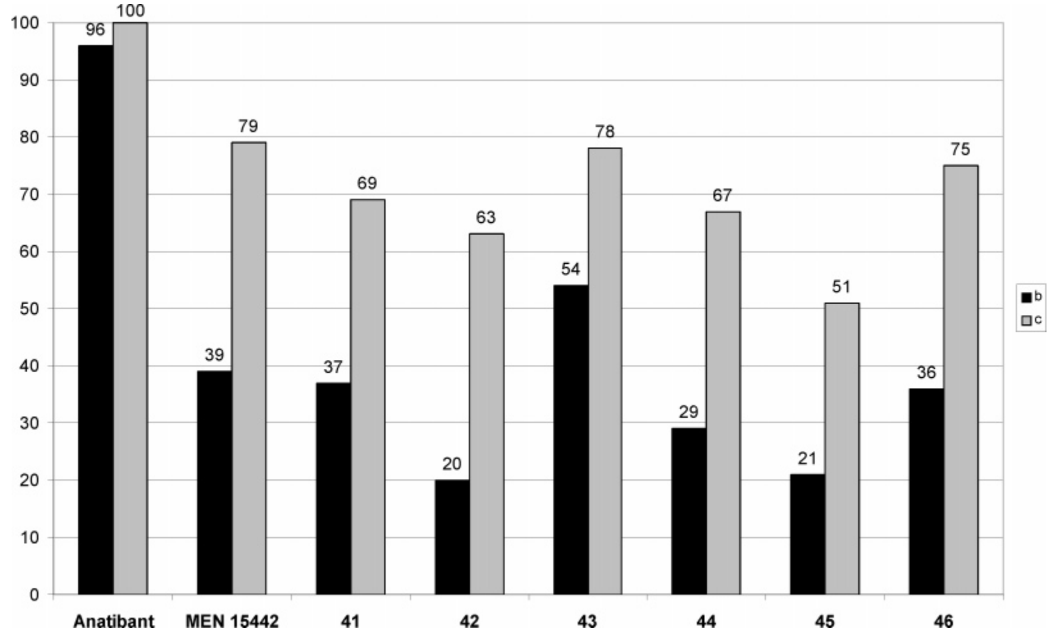
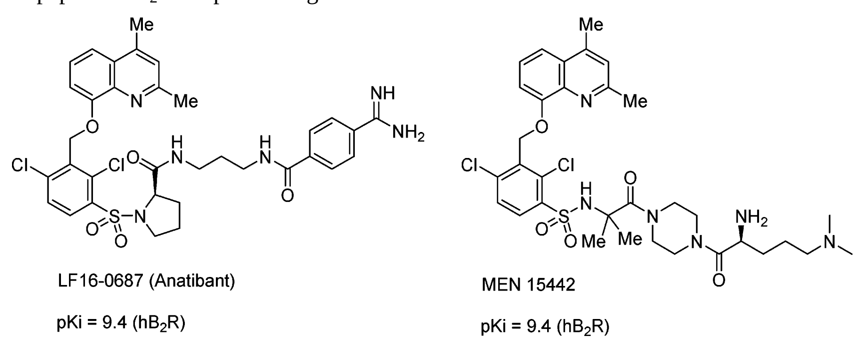


Figure 1. In vivo activity of compounds **41**−**46** and reference compounds after intratracheal administration at 300 nmol/kg. For details, see the Experimental Section. b = Σ% (BC), bronchoconstriction, see ref 12. c = Σ% (BP), blood pressure, see ref 12.

Chart 1. Competitive, Nonpeptidic hB_2 Receptor Antagonists Anatibant and MEN 15442



methyl esters (**3a**, **3b**, and **3c**) or free acids (**3d**, **3e**, and **3f**) were reacted with 2,4-dimethyl-quinolin-8-ol in the presence of a base and, in the case of methyl esters, then subjected to basic hydrolysis to obtain acids **4a**−**f** (Scheme 1). These were coupled with either functionalized piperazines (**19**, **21**, **26**, **38**, and **40**; Scheme 2, route a) or with a monoprotected piperazine residue (Scheme 2, route b). In the latter case, removal of the protecting group (Boc or Fmoc) gave intermediates **6**, which were coupled with the chosen acids. The carboxylic acids with suitably protected basic and/or charged groups were derived from L-ornithine (Scheme 3), L-*β*-lysine (Scheme 4), L-lysine (Scheme 5), or simple *ω*-amino acids of differing chain lengths (Scheme 6). The functionalized piperazines were mainly two kinds: dimethylamines (**9**, **18**, **19**, **24**, and **31**) generated via reductive amination with aq HCHO in the presence of H_2/Pd or $NaCNBH_3$ or trimethylammonium salts (**10**, **12**, **14**, **21**, **26**, **29**, **32**, **34**, and **40**) obtained via treatment of the amines with *O*-methyl-*N*,*N′*-diisopropylurea or dimethylsulfate. Finally, a guanidino moiety was introduced into compound **36** using 1,3-di-Boc-2-(trifluoromethylsulfonyl)guanidine (Goodman's reagent) and alkylated via a Mitsunobu reaction (Scheme 7). In all cases, removal of the protecting groups and HPLC purification gave the final products.

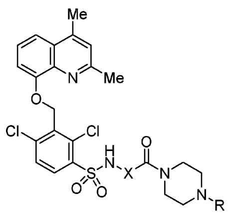
Results and Discussion

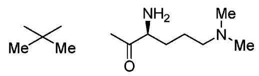
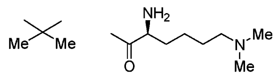
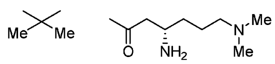
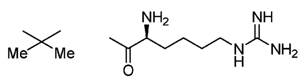
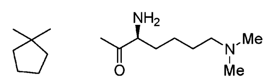
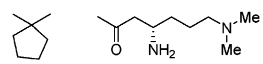
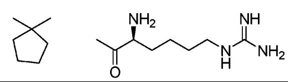
Initially we decided to explore the effect, in a small set of analogues, of replacing the α,α-dimethylglycine group of MEN 15422 with the slightly more bulky α,α-cyclopentaneglycine residue. Comparison of the in vitro activity of these compounds (**41**−**46**) revealed that the cyclopentaneglycine derivatives, generally, had similar, or slightly lower, binding affinities but slightly higher potencies in the functional assay on the hB_2 receptor (Table 1). These compounds, as well as all the others mentioned in this paper, had a pA_2 (GPI) ≥ 9.

The in vivo activity of the compounds in the guinea pig was measured after intratracheal administration (300 nmol/kg), followed 5 min later by a BK challenge (10 nmol/kg iv, repeated every 30 min for 210 min). The pulmonary insufflation pressure and the blood pressure were monitored as a measure of BK-induced bronchoconstriction and BK-induced hypotension, respectively. The results are shown in Figure 1. Under these experimental conditions, all the compounds tested were more active than Anatibant and all showed greater activity at the pulmonary B_2 receptors compared to that at the cardiovascular receptors, though for some compounds, for example, **45**, this too was appreciable (Σ% 51). In two out of the three cases examined, the cyclopentaneglycine derivatives showed better inhibition of bronchoconstriction than their dimethylglycine counterparts (see Figure 1, **41** vs **44** and **43** vs **46**).

This small improvement led us to investigate further the new series, and several modifications were made to the substituents on the piperazine ring (Table 2). Efforts focused on modifying the structure of these substituents so that, following intratracheal administration, they would help the molecule to remain "stuck"

Table 1. Binding and In Vitro Functional Activity on the hB_2 Receptor and Functional Activity on the Guinea-Pig Ileum (GPI) B_2 Receptor for Compounds **41**−**46**



Compd	X	R	pK_i[a]	pA_2(h)[b]	pA_2(GPI)[c]
Anatibant			9.4	9.4	9.5
MEN 15442			9.4	9.1	10
41			9.2	8.5	9.7
42			9.2	8.6	10
43			9.2	9.1	10
44			8.8	9.1	9.8
45			9.2	8.8	10.4
46			9.0	9.6	10

[a] pK_i for inhibition of specific binding of [^{3}H]-BK to hB_2 receptor in stably transfected CHO cells membrane preparations. [b] pA_2 for the hB_2-mediated accumulation of inositol monophosphate in stably transfected CHOdhfr-/hB_2R cells. [c] pA_2 for the antagonism of BK-induced contractile responses in guinea-pig ileum longitudinal smooth muscle. For details see the Experimental Section.

to the surface of the respiratory tract,[14] with a minimal amount being released into circulation to interact with the cardiovascular receptors. A similar strategy has been used for the muscarinic receptor antagonists ipratropium, flutropium, and tiotropium.[15] Thus, it was decided to increase the polarity of the molecule via the introduction of positive charges through the use of very basic functional groups (protonated at physiological pH) and quaternary ammonium salts. These groups have the added advantage of rendering the molecules poorly bioavailable thus avoiding absorption of the compounds following accidental ingestion during intratracheal or aerosol administration.

Carboxylic acids containing amines, ammonium salts, and guanidines at varying distances and geometries were linked to the piperazine moiety (Table 2). All the new compounds, with the single exception of compound **50**, showed subnanomolar binding affinities, and all had good activity in the functional test ($pA_2 \geq 8.2$). Best of all was ammonium salt **63**, with a pK_i of 10.3 and a pA_2 of 9.3. The combination of a primary amine and a quaternary ammonium salt at the correct distance from one another appeared to be the key for both high binding affinity and high antagonist potency (compound **63** vs compounds **49** and **61**). The majority of the compounds were then screened in vivo, at a dose of 100 nmol/kg, to assess their potency and selectivity of action in inhibiting bronchoconstriction over hypotension (Figure 2a). At this dose, only compound **63** showed very good inhibition of BK-induced bronchoconstriction (Σ% 24), with a minimal effect on BK-induced hypotension.

Therefore, we decided to use this molecule as the starting point for a final set of compounds in which a heteroatom was inserted into the central cyclic α,α-disubstituted amino acid to increase the polar surface area in this region of the molecule (Table 3). Again, all the compounds showed outstanding binding affinities, and the introduction of a tetrahydropyran ring (**72**) significantly enhanced the pA_2 in comparison to compound **63**. Those new compounds with a $pA_2 \geq 9$ were tested in vivo, and this confirmed the excellent activity of compound **72** (MEN 16132; Figure 3). The activity of this compound remained high even at lower doses; at 30 nmol/kg it showed a Σ% of 39 against BK-induced bronchoconstriction, and at an even lower dose (10 nmol/kg, i.t.) it maintained a Σ% of 63, while any significant effect on BK-induced hypotension was undetectable (Figure 4). At all three doses the inhibitory effect lasted for at least 210 min.

All the compounds reported in this paper were also tested for their affinity at the hB1 receptor and all showed a pK_i value of <5. Finally, given the presence of a trimethylammonium moiety in MEN 16132 (**72**), the compound was tested, at 1 μM, for any activity on the muscarinic or nicotinic receptors; none was found.

Conclusions

In this paper we have described the extensive work carried out on MEN 15422, the lead compound from our first paper.[11] Changes to the amino acid used to form the central sulfonamide unit and to the nature and distribution of the polar groups in the terminal chain produced a compound, MEN 16132 (**72**), which was 100× more active in vivo. This new molecule, when administered locally, was a potent and long lasting antagonist of BK-induced bronchoconstriction in the airways, devoid of any significant effects on BK-induced hypotension.[16] It could be potentially useful in the treatment of chronic airway diseases involving the proinflammatory activity of BK and is under study. Additional pharmacological results will be reported in due course.

Experimental Section

(A) Chemistry. Commercial chemicals and solvents were of reagent grade and used without further purification.

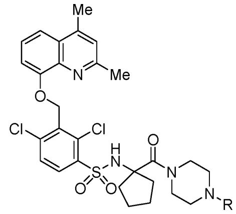
Merck silica gel (Kieselgel 60) was used for analytical thin-layer chromatography (TLC, F254 plates) and flash chromatography (230−400 mesh).

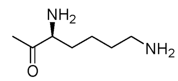
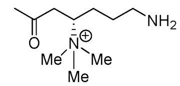
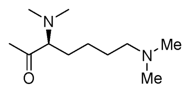
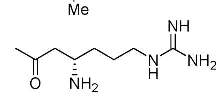
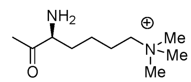
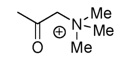
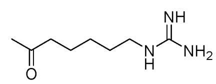
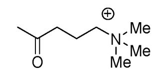
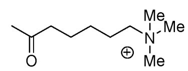
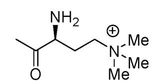
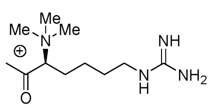
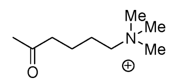
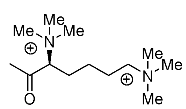
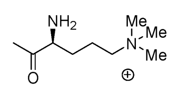
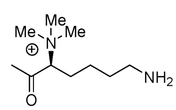
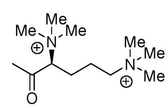
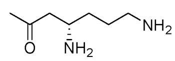
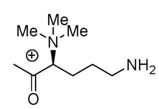
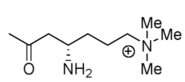
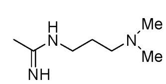
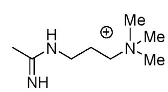
Purity was evaluated by analytical HPLC using a 600 E Waters pump coupled to a Jasco 875 UV detector and a Merck-Hitachi D-2500 integrator, or a system comprising a Jasco PU-980 pump, LG-980−02 gradient unit, a UV-975 UV/vis detector, and a Merck-Hitachi D-2500 integrator, a Beckman System Gold apparatus, or an Agilent 1100 analytic HPLC system. The solvents were (A) water + 0.1% TFA and (B) AcCN + 0.1% TFA, flow 1 mL/min.

System A: Column Vydac RP-18, 5 μm, 250 × 4.6 mm, λ = 220 nm; from 95% to 20% solvent (A) in 25 min. System B: Column Symmetry 300 RP-18, 250 × 4.6 mm, λ = 220 or 254 nm; from 80% to 20% solvent (A) in 20 min. System C: Column Agilent Zorbax Eclipse XDB C8, 5 μm, 150 × 4.6 mm; λ = 210, 240 nm; from 80% to 20% solvent (A) in 20 min. System D: column Jupiter RP-18, 5 μm, 150 × 4.6 mm, λ = 210, 240 nm; from 80% to 20% solvent (A) in 20 min.

Preparative reverse phase HPLC was performed on a Waters 600E apparatus with a Jasco 874 UV detector or on a Waters Delta-Prep 3000 apparatus. The mobile phases were the same as for the analytical systems. Gradient elution was employed. The columns used were a SymmetryPrep C18, 7 μm, 19 × 300 mm, a Hibar

Table 2. Binding and In Vitro Functional Activity on the hB_2 Receptor of Compounds **47**−**67**



Compd	R	pK_i[a]	pA_2[b]	Compd	R	pK_i[a]	pA_2[b]
47		9.5	8.7	**57**		9.9	8.9
48		9.7	8.5	**58**		9.1	9.1
49		9.2	8.9	**59**		9.4	8.3
50		8.9	8.7	**60**		9.4	8.7
51		9.3	8.3	**61**		9.5	8.7
52		10.0	9.2	**62**		9.6	8.8
53		9.7	8.2	**63**		10.3	9.5
54		9.9	8.8	**64**		9.7	8.6
55		9.2	9.3	**65**		9.6	9.3
56		9.7	8.8	**66**		9.2	9.3
				67		9.4	8.8

[a] See Table 1. [b] See Table 1. For details see the Experimental Section.

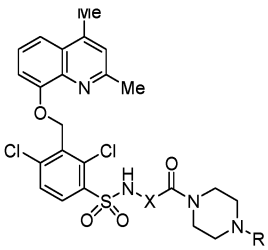
Lichrosorb RP-18, 7 μm, 25 × 250 mm, a Vydac C18, 10 μm, 22 × 250 mm or Jupiter, 15 μm, 250 × 21.2 mm. Peak detection was at 220 and 254 nm. Chemical yields are not optimized.

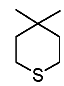
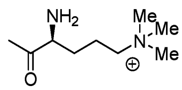
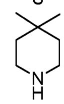
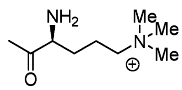
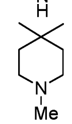
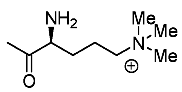
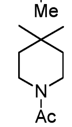
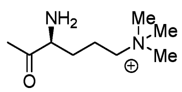
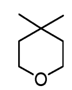
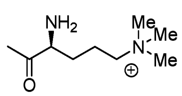
NMR experiments were recorded on a Varian Gemini 200 mod. J200 HC, a Varian 300 MHz spectrometer (equipped with a 5 mm inverse probe), a Bruker Avance 400 MHz, or a Bruker Avance 600 MHz machine and are referenced to residual solvent signals: $CDCl_3$ (δ 7.26) or DMSO-d_6 (δ 2.49). Chemical shifts are reported in δ units (parts per million) and are assigned as singlets (s), doublets (d), doublets of doublets (dd), triplets (t), quartet (q), quintet (quin), multiplets (m), broad signals (br), or very broad signals (v br). Coupling constants (*J*) are reported in hertz (Hz).

Mass spectra were recorded using a Waters Alliance 2795 HPLC system fitted with a UV-PDS 996 diode array detector, a ZMD mass spectrometer, and a GL Science Inertsil ODS-3 column (50 × 3 mm, 3 μm) or a ThermoFinnigan LCQ equipped with APCI or ESI source.

1-(3-Bromomethyl-2,4-dichloro-benzenesulfonylamino)-cyclopentanecarboxylic Acid Methyl Ester (3a). A mixture of **2** (150 mg, 0.445 mmol), 1-aminocyclopentancarboxylic acid methyl ester (87 mg, 0.490 mmol), and K_2CO_3 (150 mg, 0.980 mmol) in DMF (15 mL) was stirred at room temperature for 18 h. At the end of the reaction (TLC control) the solution was diluted with water (150 mL) and extracted with EtOAc (3 × 50 mL). The organic layer was washed with 1 N HCl (3 × 50 mL), water (50 mL), and brine (50 mL), dried over Na_2SO_4, and concentrated to afford a solid (100 mg) as a mixture of **3a** and the corresponding benzyl chloride in a ratio of 3:7. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.06−7.97 (1H, m), 7.54−7.48 (1H, m), 5.44 (1H, s), 4.94 (1.4H, s, CH_2Cl), 4.82 (0.6H, s, CH_2Br), 3.66−3.60 (3H, m), 2.52−2.03 (2H, m), 2.01−

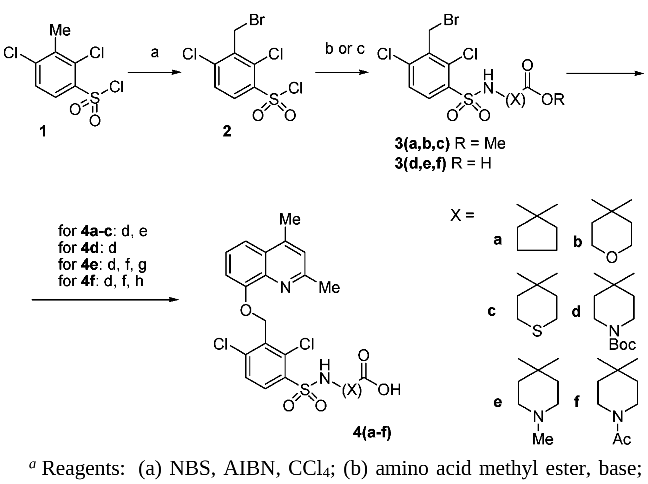
Table 3. Binding and In Vitro Functional Activity on the hB_2 Receptor of Compounds **68**−**76**



Compd	X	R	pK_i[a]	pA_2[b]
68			10.3	9.8
69			9.3	8.8
70			10.1	9.6
71			9.8	8.9
72			10.3	10.3

[a] See Table 1. [b] See Table 1. For details see the Experimental Section.

Scheme 1[a]

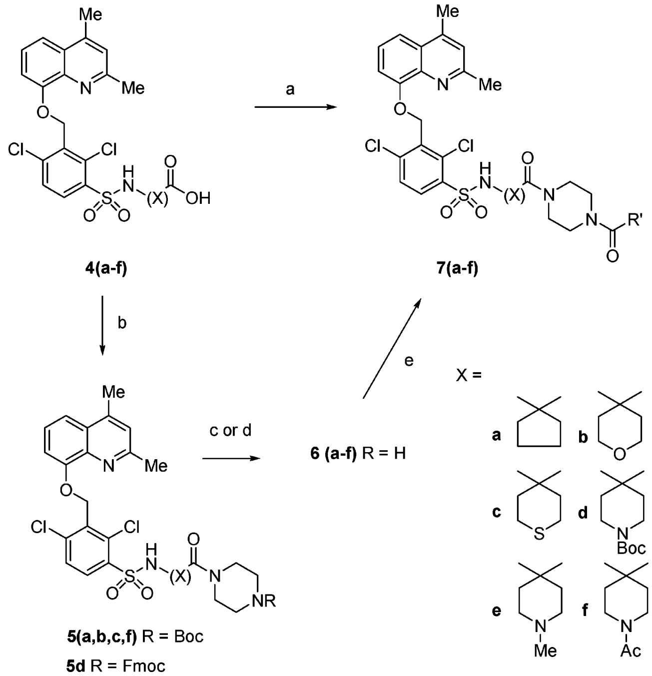


[a] Reagents: (a) NBS, AIBN, CCl_4; (b) amino acid methyl ester, base; (c) amino acid, BSA, TMSCl, THF; (d) 2,4-dimethyl-quinolin-8-ol, KI, base; (e) LiOH, THF/H_2O or NaOH, THF/H_2O; (f) HCl/dioxane; (g) HCHO, BH_3−CN resin, MeOH; (h) Ac_2O, DIPEA, DMF.

1.86 (2H, m), 1.80−1.51 (4H, m). MS *m/z* calcd for $C_{14}H_{16}BrCl_2NO_4S$, 442.9; found, 460.8, 462.9, 464.8 $[M + NH_4]^+$. MS *m/z* calcd for $C_{14}H_{16}Cl_3NO_4S$, 399.0; found, 416.9, 419.0, 421.0 $[M + NH_4]^+$. HPLC purity: system A, t_R 17.24 min (68.4%), 17.62 min (27.0%).

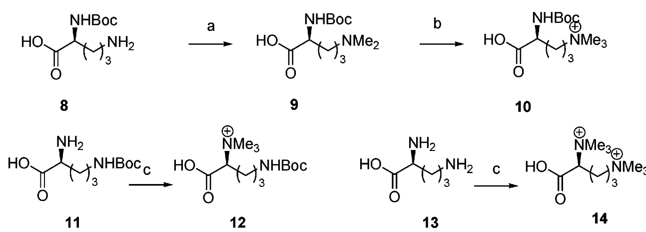
4-(3-Bromomethyl-2,4-dichloro-benzenesulfonylamino)-tetrahydro-pyran-4-carboxylic Acid Methyl Ester (3b). A solution of 5% $NaHCO_3$ (53 mL, 31.5 mmol) was added at a rate of 10 mL/h to a solution of the sulfonyl chloride **2** (2.91 g, 8.61 mmol) and the 4-amino-tetrahydropyran-4-carboxylic acid methyl ester hydrochloride (2.059 g, 10.52 mmol) in AcCN/water 10:1 (58.3 mL) at 5 °C. Stirring was continued at this temperature for 19.5 h, after which time HPLC analysis showed almost complete consumption of the sulfonyl chloride. The AcCN was removed in vacuo, the residue was diluted with water (80 mL), and the mixture was filtered under reduced pressure. The solid was washed with water (3×), air-dried for 30 min, then transferred to a tared flask and dried under high vacuum to constant weight to give **3b** as an off-white solid in a 3:1 mixture with the corresponding benzyl chloride (2.981 g, 75% yield). HPLC purity: system B, t_R 17.46 min (12.4%), 17.88 min (83.8%). ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.99−7.91 (1H, m), 7.53−7.46 (1H, m), 5.30 (1H, br s), 4.94 (0.5H, s, CH_2Cl), 4.81 (1.5H, s, CH_2Br), 3.71−3.49 (7H, m), 2.25−2.07 (2H, m), 1.99−1.81 (2H, m). MS *m/z* calcd for $C_{14}H_{16}BrCl_2NO_5S$, 458.93; found, 460.1, 462.1, 464.1 $[M + H]^+$. MS *m/z* calcd for $C_{14}H_{16}Cl_3NO_5S$, 414.98; found, 438.3, 440.1 $[M + Na]^+$. HPLC purity: system B, t_R 17.46 min (12.4%), 17.88 min (83.8%).

Scheme 2[a]



[a] Reagents: (a) EDAC, HOAt, R′CO-piperazine; (b) HOAt or HOBt, EDAC, Boc-piperazine (**5a**−**5c**, **5e**, **5f**) or Fmoc-piperazine (**5d**); (c) HCl/dioxane; (d) piperidine, DMF; (e) R′CO_2H, EDAC, HOBt, DIPEA.

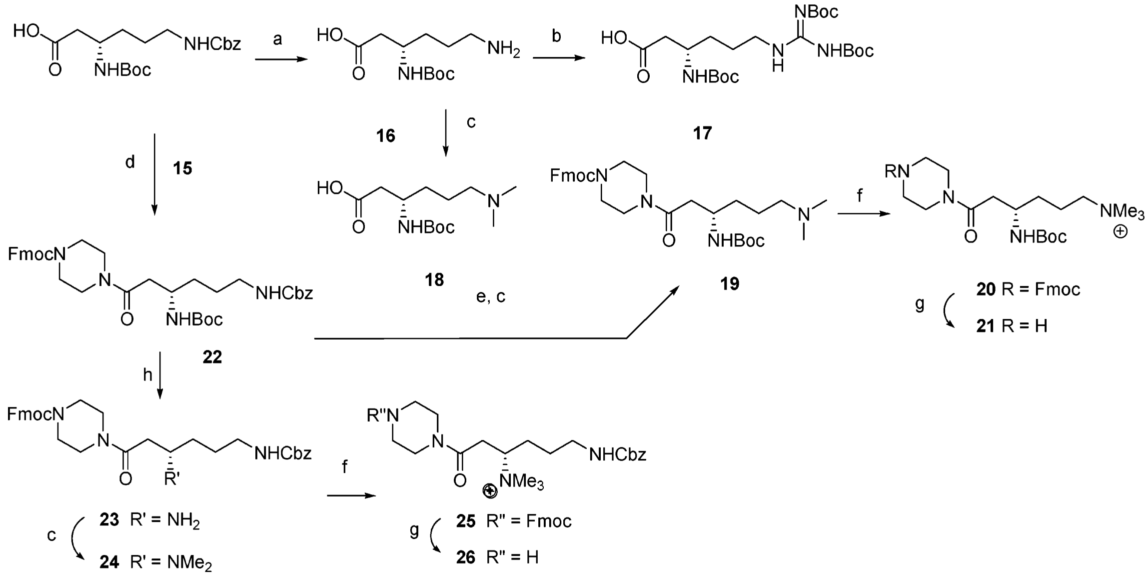
Scheme 3[a]



[a] Reagents: (a) HCHO, H_2, 10% Pd/C, MeOH; (b) *O*-methyl-*N*,*N*′-diisopropylisourea, MeOH; (c) Me_2SO_4, NaOH.

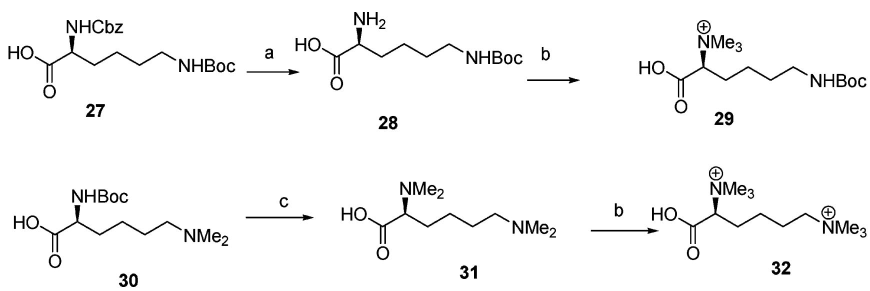
4-(3-Bromomethyl-2,4-dichloro-benzenesulfonylamino)-tetrahydro-thiopyran-4-carboxylic Acid Methyl Ester (3c). A solution of 1-amino-tetrahydrothiopyran carboxylic acid methyl ester hydrochloride (100 mg, 0.47 mmol) in 5% $NaHCO_3$ (0.7 mL) was added to a solution of sulfonyl chloride **2** (132 mg, 0.39 mmol) in AcCN (2 mL). The resulting mixture was stirred at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, and the residue was dissolved in EtOAc, washed with 1 M $KHSO_4$ (3×), dried over Na_2SO_4, and concentrated to obtain **3c** (88 mg) together with the corresponding benzyl chloride in the ratio 7:3 (50% yield). MS *m/z* calcd for $C_{14}H_{16}$-

Scheme 4[a]



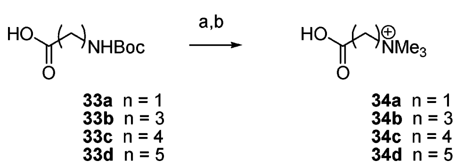
[a] Reagents: (a) H_2, 10% Pd/C, MeOH; (b) Goodman's reagent; (c) HCHO, $NaCNBH_3$; (d) Fmoc-piperazine, EDAC, HOBt, DIPEA; (e) H_2, 10% Pd/C, AcOH; (f) MeI; (g) piperidine/DMF; (h) HCl/dioxane.

Scheme 5[a]



[a] Reagents: (a) H_2, 10% Pd/C, MeOH; (b) Me_2SO_4, NaOH; (c) TFA, CH_2Cl_2, then HCHO, $NaCNBH_3$.

Scheme 6[a]



[a] Reagents: (a) TFA, CH_2Cl_2; (b) Me_2SO_4, NaOH.

$BrCl_2NO_4S_2$, 474.9; found, 492.9, 495.0, 496.9 $[M + NH_4]^+$. HPLC purity: system B t_R 16.21 min (16.3%), 16.54 min (77.4%).

4-(3-(Bromomethyl)-2,4-dichloro-phenylsulfonamido)-1-(*tert*-butoxycarbonyl)piperidine-4-carboxylic Acid (3d). A suspension of 4-Boc-1,1-aminopiperidinylcarboxylic acid (195 mg, 0.80 mmol) in dry THF (2.4 mL) was stirred at reflux temperature under nitrogen, and BSA (0.59 mL, 2.4 mmol) and TMSCl (0.31 mL, 2.4 mmol) were added. Refluxing was continued for an additional 2 h until dissolution of the amino acid was complete. Then a solution of sulfonyl chloride **2** (134 mg, 0.40 mmol) in dry THF (3 mL) was added dropwise with stirring. At the end of the reaction (HPLC control, RP-C18), the mixture was diluted with MeOH/H_2O (1:1) and concentrated in vacuo to afford an oily yellow residue. This was diluted with EtOAc, washed with water (2×) and 1 M $NaHSO_4$ (3×), dried over Na_2SO_4, and concentrated in vacuo to afford an oil that solidified upon drying. This solid consisted of a mixture of **3d** and the corresponding benzyl chloride in a 4:1 mixture (83 mg, 41% yield). It was dissolved in acetone/DMF (1:2; 1.6 mL), NaBr (130 mg, 1.27 mmol) was added, and it was stirred at room temperature to convert the benzyl chloride into the benzyl bromide. At the end of the reaction (HPLC control), the solvent was distilled off in vacuo, the residue was dissolved in EtOAc, washed with water (4×), and dried over Na_2SO_4 to obtain crude **3d** (86 mg), which was used as such in the following reaction. MS *m/z* calcd for $C_{18}H_{23}BrCl_2N_2O_6S$, 543.98; found, 542.8, 544.8, 546.8 $[M - H^+]^-$. HPLC purity: system B (100−0% A in 30 min), t_R 21.00 min (20.0%), 21.26 min (80.0%).

1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarboxylic Acid (4a). A solution of 2,4-dimethyl-quinolin-8-ol (42 mg, 0.24 mmol), **3a** (and the corresponding benzyl chloride in the ratio 3:7; 100 mg, 0.24 mmol), KI (43 mg, 0.26 mmol), and K_2CO_3 (74 mg, 0.48 mmol) in dry acetone (10 mL) was refluxed for 7 h under nitrogen. At the end of the reaction (HPLC control), the mixture was poured over acetate buffer at pH 4.2 (100 mL) and extracted with EtOAc (3 × 50 mL). The organic layers were washed with water (3 × 70 mL) and brine, dried over Na_2SO_4, filtered, and concentrated to afford 1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonyl-amino]-cyclopentanecarboxylic acid methyl ester (100 mg). A portion of this material (50 mg, 0.093 mmol) was dissolved in THF/MeOH/water (3:2:1; 6 mL) together with LiOH (24 mg, 0.1 mmol), and the resulting mixture was refluxed for 4 h. At the end of the reaction (HPLC control), the reaction was cooled to room temperature, the organic solvents were removed in vacuo, and the residue was diluted with water (10 mL). The pH was made acidic (ca. 5) by the cautious addition of 1 N HCl, and the solution was extracted with EtOAc (3 × 25 mL). The organic extracts were combined, washed with water (3 × 30 mL) and brine, dried over Na_2SO_4, filtered, and concentrated to afford crude **4a** (41 mg). ^1H NMR (300 MHz, $CDCl_3$): δ 8.13 (1H, d, $J = 8.6$ Hz), 7.68−7.50 (3H, m), 7.25 (1H, d, $J = 7.8$ Hz), 7.21 (1H, s), 5.80 (1H, s), 5.60 (2H,

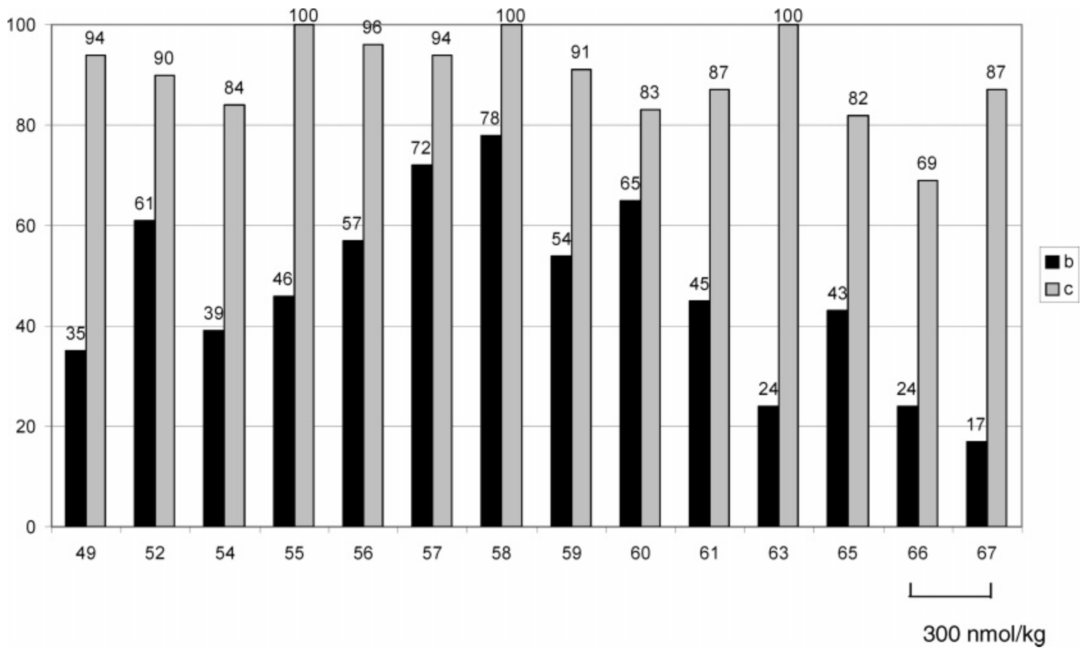
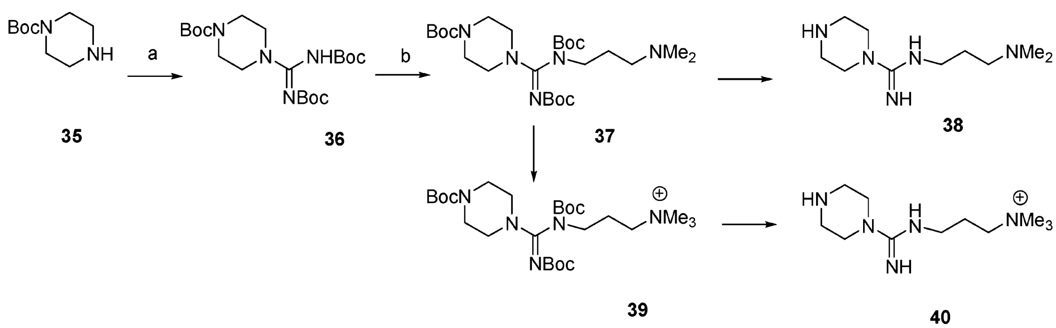


Figure 2. In vivo activity of compounds **49**−**67** after intratracheal administration at 100 nmol/kg. For details, see the Experimental Section; **66** and **67** were tested at 300 nmol/kg. b = Σ% (BC), bronchoconstriction, see ref 12. c = Σ% (BP), blood pressure, see ref 12.

Scheme 7[a]



[a] Reagents: (a) Goodman's reagent; (b) DIAD, PPh_3, 3-dimethylaminopropanol; (c) HCl/dioxane; (d) MeI.

s), 2.71 (3H, s), 2.58 (3H, s), 2.24−1.57 (8H, m). HPLC purity: system A, 80.0%, t_R 10.07 min.

4-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-tetrahydro-pyran-4-carboxylic Acid (4b). 2,4-Dimethyl-quinolin-8-ol (2.253 g, 13.02 mmol) was dissolved in a 1.5 M solution of Bu_4NOH (8.6 mL, 13 mmol) within 30 min. The resulting intense orange solution was added to Bu_4NBr (347 mg, 1.08 mmol) and compound **3b** (as a 4:1 mixture with the corresponding benzyl chloride; 4.88 g, 10.8 mmol) in AcCN (20 mL). The mixture immediately turned green, and after about 4 h, a white solid separated from the solution. The course of the reaction was monitored by HPLC. After 24 h, the mixture was diluted with water (50 mL) and filtered, and the resulting solid was washed with water (100 mL), petroleum ether (15 mL), and MeOH (15 mL) and then dried in vacuo to yield the desired compound in the form of methyl ester (4.68 g). A batch of the methyl ester so obtained (5.46 g, 9.86 mmol) was suspended in AcCN (50 mL), and the mixture was heated to reflux. A 15% aq NaOH solution (52 mL, 197 mmol) was added, and complete solution occurred to give a biphasic system, which was refluxed for 24 h. At the end of the reaction (HPLC control), the mixture was cooled to room temperature and concentrated in vacuo to remove the AcCN, and the resulting cloudy solution was carefully acidified with 4 N HCl to pH 4−5. After stirring for 1 h, a fine white precipitate formed, which was filtered off, washed with water (100 mL) and Et_2O (30 mL), and dried in vacuo to afford **4b** in 97% yield (5.45 g). ^1H NMR (400 MHz, DMSO-d_6): δ 8.64 (1H, br s), 8.03 (1H, d, J = 8.8 Hz), 7.75 (1H, d, J = 8.8 Hz), 7.67 (1H, d, J = 8.3 Hz), 7.46 (1H, dd, J = 8.3, 8.3 Hz), 7.37 (1H, d, J = 8.3 Hz), 7.30 (1H, s), 5.58 (2H, s), 3.50−3.38 (2H, m), 3.35−3.10 (2H, m), 2.64 (3H, s), 2.56 (3H, s), 1.90−1.75 (4H, m). MS m/z calcd for $C_{24}H_{24}Cl_2N_2O_6S$, 538.1; found, 539.2, 541.2 $[M + H]^+$. HPLC purity: system B, 95.0%, t_R 11.63 min.

4-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-tetrahydro-thiopyran-4-carboxylic Acid (4c). A mixture of 2,4-dimethyl-quinolin-8-ol (35 mg, 0.20 mmol) and NaH (6.0 mg, 0.24 mmol) in DMF (3 mL) was stirred at room temperature for 45 min. Then a solution of **3c** (and the corresponding benzyl chloride in ratio 7:3; 91 mg, 0.20 mmol) and KI (60 mg, 0.40 mmol) in DMF (3 mL) was added dropwise. At the end of the reaction (HPLC control), the solvent was distilled off in vacuo, and the residue was dissolved in EtOAc. The organic solution was washed with pH 4.2 buffer (3×), dried over Na_2SO_4, and concentrated to give an orange solid (88 mg, 78% yield). This was dissolved in dioxane (3.0 mL) and heated to 100 °C, and a 5% aq NaOH solution (2.9 mL) was added while stirring. At the end of the reaction (HPLC control), the solution was concentrated in vacuo, and the orange residue was partitioned between EtOAc and pH 4.2 buffer. The two phases were separated, and the organic one was dried over Na_2SO_4 and concentrated to obtain **4c** (61 mg, 72% yield) as a white solid. MS m/z calcd for $C_{24}H_{24}Cl_2N_2O_5S_2$, 554.05; found, 555.0, 557.0 $[M + H]^+$. HPLC purity: system B, 85%; t_R 10.43 min.

4-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-piperidine-1,4-dicarboxylic Acid Mono-*tert*-butyl Ester (4d). A mixture of 1,1-dimethyl-quinolin-8-ol (28 mg,

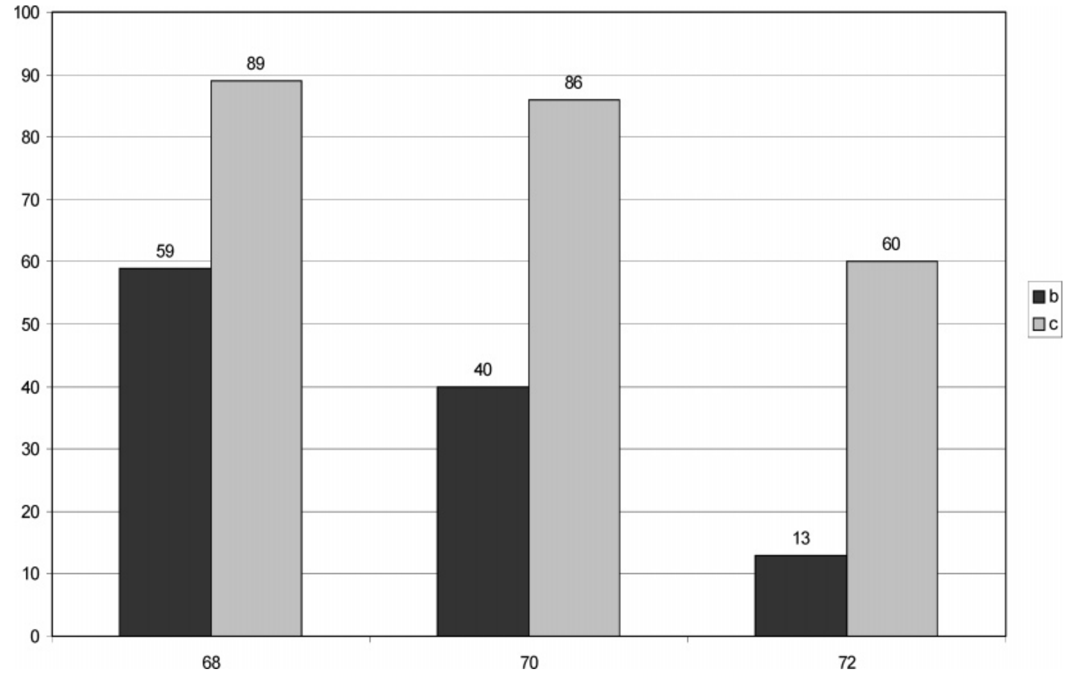


Figure 3. In vivo activity of compounds **68**, **70**, and **72** after intratracheal administration at 100 nmol/kg. For details, see the Experimental Section. b = Σ% (BC), bronchoconstriction, see ref 12. c = Σ% (BP), blood pressure, see ref 12.

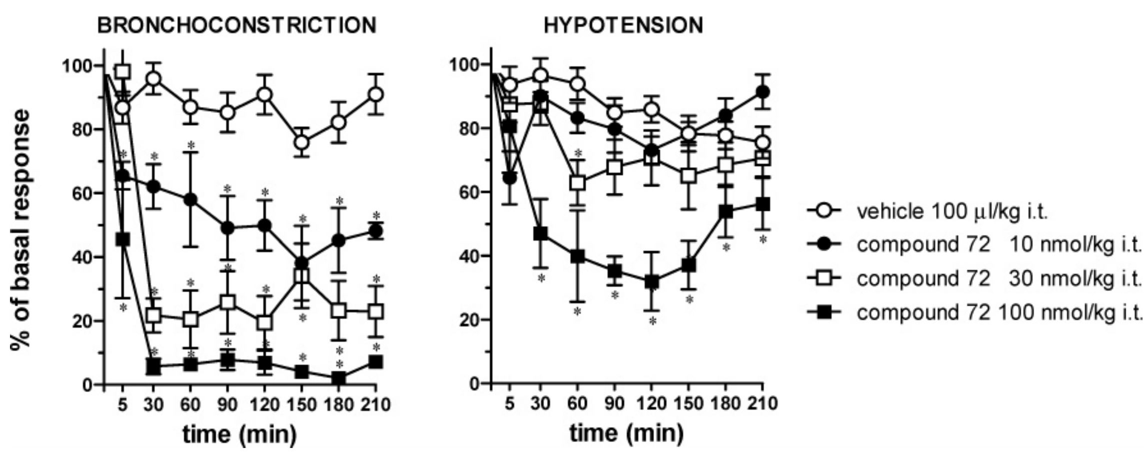


Figure 4. In vivo activity of compound **72** after intratracheal administration on bronchoconstriction and hypotension induced by repeated challenges with BK (10 nmol/kg iv) in anaesthetized guinea pigs.[16]

0.16 mmol) and LiOH (10 mg, 0.4 mmol) in dry AcCN (2.5 mL) was stirred at room temperature for 1.5 h under nitrogen. Then a solution of crude **3d** (86 mg, 0.16 mmol) in AcCN/DMF (2:1) (2 mL) was added dropwise, and the resulting mixture was stirred at room temperature. At the end of the reaction (HPLC control), MeOH/water (1:1; 3 mL) was added, stirring was continued for an additional 15 min, and then the solvents were distilled off in vacuo. The residue was partitioned between EtOAc and pH 4.2 buffer. The two phases were separated, and the organic one was washed again with buffer (5×). The organic layer was dried over Na_2SO_4, filtered, and concentrated in vacuo. The residue was triturated with Et_2O to afford crude **4d** (70 mg, 70% yield). MS *m*/*z* calcd for $C_{29}H_{33}Cl_2N_3O_7S$, 637.14; found, 638.1, 640.0 $[M + H]^+$. HPLC purity: system B, (100−0% A in 30 min), 70%, t_R 17.59 min.

2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-*N*-[1-(piperazine-1-carbonyl)-cyclopentyl]-benzenesulfonamide (6a). EDAC (981 mg, 5.12 mmol) and HOBt (629 mg, 5.12 mmol) were added to a solution of acid **4a** (2.43 g, 4.66 mmol) in CH_2Cl_2 (50 mL) stirred in an ice bath. The resulting mixture was stirred for an additional hour, then *N*-Boc-piperazine (697 mg, 5.12 mmol) and DIPEA (0.88 mL, 5.12 mmol) were added, and stirring was continued at room temperature overnight. Then the mixture was diluted with CH_2Cl_2 (20 mL) and washed with 0.1 N $KHSO_4$ (50 mL), 0.1 N NaOH (50 mL), and brine (50 mL), dried over Na_2SO_4, filtered, and concentrated in vacuo. The crude product was purified by flash chromatography to give the Boc-protected derivative in 99% purity. This was suspended in EtOAc (15 mL), and 4 N HCl in dioxane (20 mL) was added dropwise while stirring at room temperature. At the end of the reaction (HPLC control), the mixture was concentrated in vacuo, dissolved in water, and lyophilized to give **6a** (2.15 g, 73% yield) as a white solid in the form of the hydrochloride salt. ^1H NMR (300 MHz, $CDCl_3$): δ 8.94 (2H, s), 8.7 (s, 1H), 8.34 (1H, br s), 8.04−8.01 (1H, d), 7.83−7.81 (1H, d), 7.61−7.45 (4H, m), 5.56 (2H, s), 3.16 (4H, br s), 2.75 (3H, s), 2.69 (3H, s), 2.65 (3H, s), 1.93 (2H, m), 1.73 (2H, m), 1.42 (4 H, m). MS *m*/*z* calcd for $C_{28}H_{32}Cl_2N_4O_4S$, 591.56; found, 592.2 $[M + H]^+$. HPLC purity: system B (100−0% A in 30 min), 94%, t_R 10.37 min.

2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-*N*-[4-(piperazine-1-carbonyl)-tetrahydro-pyran-4-yl]-benzenesulfonamide (6b). EDAC (51 mg, 0.27 mmol) was added to a chilled solution of acid **4b** (105 mg, 0.195 mmol) and HOAt (37 mg, 0.27 mmol) in DCM (6.0 mL). The resulting mixture was stirred for 30 min at 0 °C and then *N*-Boc-piperazine (66 mg, 0.35 mmol) was added. Stirring was continued at 0 °C for 30 min then at room temperature. At the end of the reaction (HPLC control), the DCM was removed in vacuo, and the residue was partitioned between EtOAc (50 mL) and pH 4.2 buffer (50 mL). The layers were separated, and the organic phase was washed with buffer (50 mL). The aqueous washes were combined and re-extracted with EtOAc (50 mL), then the organic extracts were combined and washed with 5%$NaHCO_3$ (2 × 50 mL) and brine (2 × 50 mL), dried over Na_2SO_4, and filtered, and the filtrate was concentrated in vacuo to give crude **5b**. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.02 (1H, d, J = 8.7 Hz), 7.64 (1H, d, J = 8.3 Hz), 7.50 (1H, d, J = 8.6 Hz), 7.39 (1H, t, J = 8.0 Hz), 7.20 (1H, d, J = 7.6 Hz), 7.15 (1H, s), 5.73 (2H, s), 5.52 (1H, s), 3.87−3.69 (4H, m), 3.63−3.43 (8H, m), 2.69 (3H, s), 2.64 (3H, s), 2.21−2.10 (2H, m), 1.65−1.52 (2H, m), 1.47 (9H, s). MS *m*/*z* calcd for $C_{33}H_{40}Cl_2N_4O_7S_2$, 706.2; found, 707.2 $[M + H]^+$. HPLC purity: system B, 95.9%, t_R 15.22 min. HCl/dioxane (4 N, 4 mL) was added at room temperature to a solution of **5b** (138 mg, 0.195 mmol) in DCM (4 mL). At the end of the reaction (HPLC control), the solvents were removed in vacuo, and the residue was triturated with Et_2O, filtered, washed with Et_2O, and dried under a stream of N_2 to give **6b** (131 mg, 98%) as a pale yellow solid. ^{1}H NMR (300 MHz, DMSO-d_6): δ 9.73−9.50 (2H, m), 8.92 (1H, s), 8.46−8.12 (4H, m), 7.34−7.08 (2H, m), 5.68 (2H, br s), 4.15−3.77 (4H, m), 3.27−2.97 (6H, m), 2.83 (6H, br s), 1.98−1.77 (2H, m), 1.74−1.54 (2H, m). MS *m*/*z* calcd for $C_{29}H_{34}Cl_2N_4O_4S$, 606.15; found, 607.2, 609.2 $[M + H]^+$. HPLC purity: system B, 94.9%, t_R 9.46 min.

2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-*N*-[4-(piperazine-1-carbonyl)-tetrahydro-thiopyran-4-yl]-benzenesulfonamide (6c). A solution of **4c** (45 mg, 0.083 mmol) and DIPEA (28 μL, 0.083 mmol) in DMF (2 mL) was stirred at room temperature. PyBOP (43 mg, 0.083 mmol) was added, followed by *N*-Boc-piperazine (15 mg, 0.083 mmol). At the end of the reaction (HPLC control), the solvent was distilled off in vacuo, and the orange residue was dissolved in EtOAc and washed with acetate buffer, pH 4.2 (3×). The organic layer was dried over Na_2SO_4, filtered, and concentrated to obtain crude **5c** (45 mg). MS *m*/*z* calcd for $C_{33}H_{40}Cl_2N_4O_6S_2$, 722.18; found, 723.1, 725.1 $[M + H]^+$. This crude product was dissolved in TFA/CH_2Cl_2 (1:1; 2 mL) and stirred at room temperature. After 1 h, Et_2O was added with stirring and a white solid precipitated. This was filtered off and dried under nitrogen to give crude **6c** as trifluoroacetate salt (26 mg), which was used as such without further purification. MS *m*/*z* calcd for $C_{28}H_{32}Cl_2N_4O_4S_2$, 622.12; found, 623.1, 625.1 $[M + H]^+$.

4-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-4-(piperazine-1-carbonyl)-piperidine-1-carboxylic Acid *tert*-Butyl Ester (6d). A solution of **4d** (70 mg, 0.11 mmol) in DMF (3 mL) was stirred at room temperature. EDAC (25 mg, 0.13 mmol) and HOBt (18 mg, 0.13 mmol) were added. Fmoc-piperazine hydrochloride (38 mg, 0.110 mmol) was dissolved in DMF (1 mL), DIPEA was added (0.04 mL, 0.26 mmol), and the resulting solution was added dropwise to the solution of the active ester. The resulting mixture was stirred overnight. The solvents were distilled off in vacuo, and the residue was partitioned between EtOAc and pH 4.2 buffer. The organic layer was washed with buffer (2×) and then dried over Na_2SO_4, concentrated, and triturated with Et_2O to obtain crude **5d** as a white solid (76 mg, 75% yield). MS *m*/*z* calcd for $C_{48}H_{51}Cl_2N_5O_8S$, 927.28; found, 928.2, 930.2 $[M + H]^+$. A solution of **5d** (76 mg, 0.081 mmol) in piperidine/DMF 1:10 (3 mL) was stirred at room temperature for 1 h. The solution was concentrated in vacuo, and the resulting yellow oil was triturated with Et_2O to obtain **6d** as a white solid (47 mg, 81% yield). MS *m*/*z* calcd for $C_{33}H_{41}Cl_2N_5O_6S$, 705.22; found, 706.2, 708.2 $[M + H]^+$.

2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-*N*-[1-methyl-4-(piperazine-1-carbonyl)-piperidin-4-yl]-benzenesulfonamide (6e). A solution of **4d** (173 mg, 0.27 mmol) in dry dioxane (1 mL) was cooled to 0 °C, 4 N HCl in dioxane (10 mL) was added, and the mixture was warmed to room temperature. At the end of the reaction (2 h, HPLC control), the solvents were distilled off under reduced pressure and the residue was triturated with Et_2O (3×) to obtain crude 4-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-piperidine-4-carboxylic acid as the hydrochloride salt (176 mg). A portion of this product (50 mg, 0.08 mmol) was dissolved in MeOH (4.5 mL), and 37% aq HCHO (30.7 μL, 0.41 mmol) was added, together with $NaCNBH_3$ polystyrene resin (38 mg, 4.3 mmol/gr, 0.16 mmol), and stirring was continued at room temperature. At the end of the reaction (HPLC control), the resin was filtered off and the residual solution was concentrated in vacuo, with the help of toluene (4×), to obtain crude **4e**.

Crude **4e** (40 mg, 0.07 mmol) in dry DMF (1 mL) was stirred at room temperature. HOAt (98 mg, 0.07 mmol), DCC (14.8 mg, 0.07 mmol), and Boc-piperazine (13 mg, 0.07 mmol) were added in that order and stirring was continued overnight. Na_2CO_3 (5%, 5 mL) was added and stirring was continued for an additional 30 min. The mixture was then extracted with EtOAc (3×) and the organic layer was washed with 5% Na_2CO_3 (2×). The EtOAc layer was acidified with diluted HCl to pH 4, washed with water, dried over Na_2SO_4, and concentrated under reduced pressure. The resulting amorphous solid was dissolved in THF, cooled for 3 days at −20 °C, filtered to eliminate the residual DCU, and concentrated again to obtain crude **5e**. This material was dissolved in dry EtOAc (1 mL), 4 N HCl in dioxane (5 mL) was added, and the resulting mixture was stirred for 30 min. The solvents were distilled off and the residue was dried in vacuo to obtain crude **6e** (114 mg), which was used as such without further purification.

***N*-[1-Acetyl-4-(piperazine-1-carbonyl)-piperidin-4-yl]-2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide (6f).** A portion of 4-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-piperidine-4-carboxylic acid hydrochloride salt (see preparation for **6e**; 60 mg, 0.098 mmol) was dissolved in dry DMF (1 mL). Acetic anhydride (9.3 μL, 0.098 mmol) and DIPEA (50 μL, 0.29 mmol) were added while stirring at room temperature. After 4 h (HPLC control), water (2 mL) was added and the solvents were distilled off in vacuo. Toluene was added, and the mixture was concentrated again (4×). The solid residue was triturated with Et_2O to obtain crude **4f** (78 mg). This crude product (0.098 mmol) was dissolved in dry DMF (2 mL) and then EDAC (20.7 mg, 0.108 mmol), HOAt (14.7 mg, 0.108 mmol), DIPEA (32 μL, 0.098 mmol), and Boc-piperazine (20.1 mg, 0.108 mmol) were added in that order while stirring at room temperature. After 12 h (HPLC control), 5% $NaHCO_3$ (5 mL) was added and stirring was continued for an additional 30 min. Then EtOAc was added and the two phases were separated. The organic phase was washed with 5% $NaHCO_3$ (3×) and pH 4.2 buffer (3×), dried over Na_2SO_4, and concentrated. Flash chromatographic purification (silica, $CHCl_3$, then $CHCl_3$/MeOH 99:1, then $CHCl_3$/MeOH 98:2) afforded pure **5f** (19 mg, 0.025 mmol). This was dissolved in dry dioxane (1 mL), and 4 N HCl in dioxane (2 mL) was added while cooling in an ice bath. At the end of the reaction (HPLC control), the solvents were distilled off and the residue was triturated with Et_2O to obtain **6f** (18 mg, quantitative). MS *m*/*z* calcd for $C_{30}H_{35}Cl_2N_5O_5S$, 647,17; found, 648.1, 650.1 $[M + H]^+$.

(*S*)-2-*tert*-Butoxycarbonylamino-5-dimethylamino-pentanoic Acid (9). BocOrn-OH (8) was suspended in MeOH (100 mL), and 37% aq HCHO (1.6 mL, 0.02 mmol) was added, followed by 10% Pd/C (200 mg). The reaction mixture was stirred under H_2 for 3 h, then the catalyst was filtered off, and the filtrate was concentrated to dryness. Et_2O (100 mL) was added to the resulting oil, and stirring was continued to obtain a white solid, which was filtered off and washed with Et_2O. A second precipitation was performed by dissolving the solid in the minimum amount of MeOH (3 mL) and then adding Et_2O (100 mL). The desired product was finally collected by filtration, following a further wash with Et_2O, as a highly hygroscopic white solid that was stored under nitrogen. Total yield, 1.60 g (60%). ^{1}H NMR (400 MHz, DMSO-d_6): δ 6.49 (1H, d), 6.05 (1H, d), 3.75−3.65 (1H, m), 2.70−2.53 (2H, m), 2.39 (6H, s), 1.70−1.41 (4H, m), 1.38 (9H, s). ^{13}C NMR (100 MHz, DMSO-d_6): δ 175.27, 155.94, 78.54, 58.21, 54.74, 43.99, 30.23, 29.09, 22.5. MS *m*/*z* calcd for $C_{12}H_{24}N_2O_4$, 260.0; found, 261.0 $[M + H]^+$.

(*S*)-2-*tert*-Butoxycarbonylamino-5-trimethylammonium-pentanoic Acid (10). Amino acid **9** (1.16 g, 4.37 mmol) was dissolved in MeOH (5 mL). *O*-Methyl-*N,N′*-diisopropylisourea (0.87 mL, 4.81 mmol) was added, and the reaction mixture was stirred at room temprature for 18 h (MS-HPLC control). Solvent removal under reduced pressure, followed by addition of water (30 mL), caused the precipitation of the diisopropylurea, which was filtered off. The aqueous layer was washed with EtOAc (3 × 30 mL) to remove the unreacted *O*-methyl-*N,N′*-diisopropylisourea, and then the water was again distilled off under reduced pressure. Absolute EtOH (3 × 50 mL) was added and then removed under reduced pressure until a white foam formed. Et_2O (50 mL) was added, and the resulting suspension was stirred at room temperature for 1 h. Ammonium salt **10** was finally collected, after filtration and washing with Et_2O, as a highly hygroscopic white solid that was stored under nitrogen (1.10 g, 90% yield). ^{1}H NMR (400 MHz, DMSO-d_6): δ 6.49 (1H, d), 3.45 (1H, m), 3.29 (2H, m), 3.03 (9H, s), 1.75−1.58 (4H, m), 1.38 (9H, s). ^{13}C NMR (100 MHz, DMSO-d_6): δ 172.38, 155.73, 78.19, 66.3, 55.15, 52.6, 29.9, 28.06, 18.7. MS *m*/*z* calcd for $C_{13}H_{27}N_2O_4$, 275.0; found, 275.0 [M]$^+$.

(*S*)-(4-*tert*-Butoxycarbonylamino-1-carboxy-butyl)-trimethyl-ammonium (12). Amino acid **11** (0.100 g, 0.40 mmol) was dissolved in 10% NaOH (4 mL), Me_2SO_4 (0.4 mL) was added, and the reaction mixture was stirred for 30 min at 0 °C and then for 30 min at room temperature (MS-HPLC control). The solution was neutralized with 0.5 N HCl, the solvent was removed under reduced pressure, and the residue was triturated with Et_2O to obtain **12**, which was used without further purification. MS *m*/*z* calcd for $C_{13}H_{27}N_2O_4$, 275.37; found, 275.4 [M]$^+$.

(*S*)-(1-Carboxy-butyl)-1,4-ditrimethyl-ammonium (14). Amino acid **13** monochloride (0.200 g, 1.20 mmol) was dissolved in 10% NaOH (5 mL), Me_2SO_4 (0.5 mL) was added, and the reaction mixture was stirred for 30 min at 0 °C and for 30 min at room temperature (MS-HPLC control). The solution was neutralized with 0.5 N HCl, the solvent was removed under reduced pressure, and the residue was triturated with Et_2O to obtain **14**, which was used without further purification. MS *m*/*z* calcd for $C_{11}H_{26}N_2O_2$, 218.34; found, 218.5 [M]$^+$.

(*S*)-6-Amino-3-*tert*-butoxycarbonylamino-hexanoic Acid (16). A solution of Boc-β-Lys(*Z*)-OH (210 mg, 0.55 mmol) in MeOH (20 mL) was stirred under H_2 in the presence of 10% Pd/C (50 mg). At the end of the reaction (TLC control, silica, $CHCl_3$/MeOH 9:1), the solution was filtered through Celite and the filtrate was concentrated in vacuo to obtain **16** (130 mg, 96%) as a colorless oil. MS *m*/*z* calcd for $C_{11}H_{26}N_2O_2$, 218.34; found, 219.2 [M + H]$^+$.

(*S*)-6-[(*tert*-Butoxycarbonimidoylimino-*tert*-butoxycarbonylamino-methyl)-amino]-3-*tert*-butoxycarbonylamino Hexanoic Acid (17). A suspension of **16** (130 mg, 0.52 mmol) in CH_2Cl_2 (5 mL) was treated with BSA (0.40 mL, 1.56 mmol) until complete solution occurred. Goodman's reagent (600 mg, 1.56 mmol) was added, and stirring was continued at room temperature for 24 h. At the end of the reaction (TLC control, silica, $CHCl_3$/MeOH 9:1), the mixture was concentrated under reduced pressure, and the crude residue was purified by flash chromatography (silica, CH_2Cl_2/MeOH 9:1) to obtain **17** (112 mg, 44%). MS *m*/*z* calcd for $C_{22}H_{40}N_4O_8$, 488.594; found, 489.6 [M + H]$^+$.

(*S*)-3-*tert*-Butoxycarbonylamino-6-dimethylamino-hexanoic Acid (18). A solution of **16** (113 mg, 0.45 mmol) in dioxane (10 mL) was treated with 37% aq HCHO (250 μL, 3.0 mmol), and $NaCNBH_3$ (75 mg, 1.2 mmol) in water (3 mL) was added. At the end of the reaction (TLC control, silica, AcCN/water 5:15), the mixture was concentrated under reduced pressure, and the crude product was purified by flash chromatography (silica, CH_3CN/H_2O 1:4) to obtain **18** (65 mg, 55%). MS *m*/*z* calcd for $C_{131}H_{26}N_2O_4$, 274.36; found, 275.4 [M + H]$^+$.

4-[(*S*)-3-*tert*-Butoxycarbonylamino-6-dimethylamino-hexanoyl]-piperazine-1-carboxylic Acid 9*H*-Fluoren-9-ylmethyl Ester (19). Pd/C (10%, 45 mg) was wetted with three drops of water, suspended in AcOH (2 mL), and added, under nitrogen, to a solution of **22** (400 mg, 0.596 mmol) in AcOH (4 mL). The resulting mixture was cooled in a water bath to 10 °C and degassed with nitrogen for 10 min. Hydrogen was then bubbled through the mixture while the temperature was maintained below 15 °C by periodic addition of solid CO_2 to the water bath. At the end of the deprotection reaction (HPLC control), the solution was degassed with nitrogen for 20 min, while maintaining the temperature at 0 °C, and then filtered. The filtrate was concentrated in vacuo to give the free amine as the acetate salt. This crude product was purified by flash chromatography ($CHCl_3$/MeOH/AcOH 85:10:5) to obtain the pure amine as the acetate salt. MS *m*/*z* calcd for $C_{30}H_{40}N_4O_5$, 536.30; found, 537.3 [M + H]$^+$. HPLC purity: system B, >99%, t_R 16.42 min.

This salt was dissolved in CH_2Cl_2/AcOH 9:1 (10 mL), and 37% aq HCHO (220 μL, 2.71 mmol) was added. The resulting solution was agitated (150 oscillations/min) at room temperature for 20 min, (polystyrylmethyl)trimethylammonium cyanoborohydride (339 mg, 1.39 mmol) was added, and the agitation was continued at room temperature. At the end of the reaction (HPLC and TLC control), the resin was filtered off under nitrogen and washed with CH_2Cl_2 (3 × 10 mL), and the filtrates were combined and concentrated in vacuo to give **19** (318 mg) as an off-white solid after concentration from toluene (3×). A portion of this product was used in the next step without further purification. MS *m*/*z* calcd for $C_{32}H_{44}N_4O_5$, 564.33; found, 565.3 [M + H]$^+$.

{(*S*)-4-*tert*-Butoxycarbonylamino-6-[4-(9*H*-fluoren-9-ylmethoxycarbonyl)-piperazin-1-yl]-6-oxo-hexyl}-trimethyl-ammonium trifluoroacetate (20). Methyl iodide (160 μL, 2.57 mmol) was added to a solution of **19** (260 mg, 0.419 mmol) in CH_2Cl_2/MeOH (2:1; 6 mL) and stirring at room temperature was continued for 2 days. At the end of the reaction (HPLC control), the solvents were removed in vacuo and the residue was triturated with Et_2O. The resulting off-white solid was filtered under nitrogen, washed with Et_2O (3 × 6 mL), and dried under a stream of nitrogen to give crude **20**. This crude product was purified by preparative HPLC to obtain pure **20** (118 mg, 40%) as the trifluoroacetate salt. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.78−7.74 (2H, m), 7.60−7.53 (2H, m), 7.42−7.37 (2H, m), 7.37−7.28 (2H, m), 5.88−5.71 (1H, br s), 4.49 (2H, d, J = 6.4 Hz), 4.29−4.20 (1H, t, J = 6.4 Hz), 4.02−3.86 (1H, br s), 3.64−3.29 (10H, m), 3.16 (9H, m), 2.70−2.47 (2H, m), 1.99−1.69 (3H, m), 1.68−1.49 (1H, m), 1.41 (9H, s). MS *m*/*z* calcd for $C_{33}H_{47}N_4O_5$, 579.35; found, 579.2 [M + H]$^+$. HPLC purity: system B, 98.6%, t_R 16.90 min.

[(*S*)-4-*tert*-Butoxycarbonylamino-6-oxo-6-piperazin-1-yl-hexyl]-trimethyl-ammonium Trifluoroacetate (21). A solution of **20** (84 mg, 0.119 mmol) in 20% piperidine/DMF (2.4 mL) was stirred at room temperature. At the end of the deprotection reaction (HPLC control), the solvents were removed in vacuo and the residue was concentrated twice from DMF under reduced pressure. The resulting white solid was triturated with Et_2O and the supernatant was removed by pipet. The residue was dissolved in MeOH and concentrated again to obtain **21** in the form of the trifluoroacetate salt (59 mg), which was used as such without any further purification.

4-[(*S*)-6-Benzyloxycarbonylamino-3-*tert*-butoxycarbonylamino-hexanoyl]-piperazine-1-carboxylic Acid 9*H*-Fluoren-9-ylmethyl Ester (22). Commercially available Boc-β-Lys(*Z*)-OH DCHA (987 mg, 1.76 mmol) was partitioned between EtOAc (100 mL) and 1 N HCl (100 mL). The layers were separated, and the organic phase was washed with water (2 × 50 mL) and brine (2 × 50 mL), dried (Na_2SO_4), and concentrated under reduced pressure to give **15** as the free acid. A solution of acid **15** (447 mg, 1.17 mmol), HOBt (217 mg, 1.42 mmol), and EDAC (286 mg, 1.49 mmol) in CH_2Cl_2 (4 mL) was stirred at 0 °C for 30 min. Then Fmoc-piperazine hydrochloride (412 mg, 1.19 mmol) was added, followed by DIPEA (420 μL, 2.41 mmol), and stirring was continued at 0 °C for 30 min and then at room temperature. At the end of the reaction (HPLC control), the solvent was distilled off in vacuo, and the residue was partitioned between EtOAc (50 mL) and 1 N HCl (50 mL). The layers were separated and the organic phase was washed with 1 N HCl (50 mL). The combined acid washes were back-extracted with EtOAc (50 mL), then the organic extracts were combined and washed with 5% $NaHCO_3$ (2 × 50 mL), water (50 mL), and brine (50 mL), dried over Na_2SO_4, and concentrated in vacuo to afford crude **22** (694 mg, 88%) as a pale yellow foam. ^{1}H NMR (300

MHz, DMSO-d_6): δ 7.94−7.80 (2H, m), 7.68−7.60 (2H, m), 7.46−7.29 (9H, m), 7.26−7.19 (1H, br t, J = 5.5 Hz), 6.70−6.63 (1H, d, J = 8.8 Hz), 4.99 (2H, s), 4.46−4.37 (2H, d, J = 6.1 Hz), 4.33−4.25 (1H, t, J = 6.2 Hz), 3.77−3.65 (1H, m), 3.45−3.25 (8H, m), 3.02−2.91 (2H, m), 2.48−2.28 (2H, m), 1.48−1.28 (13H, m). HPLC purity: system B, 83.6%, t_R 23.26 min.

4-[(*S*)-3-Amino-6-benzyloxycarbonylamino-hexanoyl]-piperazine-1-carboxylic Acid 9*H*-Fluoren-9-ylmethyl Ester (23). A 4 N solution of HCl in dioxane (4 mL, 8 mmol) was added dropwise to a solution of **22** (232 mg, 0.346 mmol) in CH_2Cl_2 (4 mL), and stirring was continued for 40 min at room temperature. The solvents were removed in vacuo, and the residue was dissolved in MeOH, concentrated (2×), dissolved in toluene/MeOH (1:1), and concentrated again. The resulting oil was triturated with Et_2O to obtain a solid, which was filtered off under nitrogen to give **23** (198 mg, 94%) as the hydrochloride salt. ^{1}H NMR (300 MHz, DMSO-d_6): δ 7.92−7.87 (2H, m), 7.82−7.71 (3H, br s), 7.65−7.60 (2H, m), 7.46−7.24 (10H, m), 5.01 (2H, s), 4.45−4.38 (2H, d, J = 6.2 Hz), 4.31−4.25 (1H, t, J = 6.2 Hz), 3.50−3.31 (8H, m), 3.04−2.94 (2H, m), 2.77−2.68 (1H, m), 2.60−2.52 (1H, m), 1.64−1.42 (4H, m). HPLC purity: system B, 90.3%, t_R 17.72 min.

4-[(*S*)-6-Benzyloxycarbonylamino-3-dimethylamino-hexanoyl]-piperazine-1-carboxylic Acid 9*H*-Fluoren-9-ylmethyl Ester (24). A solution of **23** (82 mg, 0.135 mmol) and 37% aq HCHO (66 μL, 0.81 mmol) in MeOH (2 mL) was stirred at room temperature for 20 min. (Polystyryl)trimethylammonium cyanoborohydride (124 mg, 4.1 mmol) was added and stirring was continued at room temperature. At the end of the reaction (HPLC control), the resin was filtered off and washed with MeOH (3×), and the filtrates were combined and concentrated under reduced pressure to give crude **24** (83 mg, 97%) as a colorless oil, which was used, as such, without further purification. MS m/z calcd for $C_{35}H_{42}N_4O_5$, 598.32; found, 599.1 [M + H]$^+$.

((*S*)-4-Benzyloxycarbonylamino-1-{2-[4-(9*H*-fluoren-9-ylmethoxycarbonyl)-piperazin-1-yl]-2-oxo-ethyl}-butyl)-trimethyl-ammonium (25). A solution of crude **24** (77 mg, 0.12 mmol) and MeI (80 μL, 1.3 mmol) in MeOH (2 mL) was stirred at room temperature for five days. Because some starting amine still remained, the solvents were distilled off in vacuo, and the residue was dissolved in neat MeI (4 mL) and stirred at room temperature in the dark for an additional three days. The MeI was removed in vacuo, and the residue was purified by preparative HPLC to obtain **25** (54 mg, 62%), as a colorless oil, in the form of trifluoroacetate salt. MS m/z calcd for $C_{36}H_{45}N_4O_5$, 613.34; found, 613.20 [M]$^+$.

[(*S*)-4-Benzyloxycarbonylamino-1-(2-oxo-2-piperazin-1-yl-ethyl)-butyl]-trimethyl-ammonium Iodide (26). A solution of **25** (54 mg, 0.074 mmol) in 20% piperidine/DMF (4 mL) was stirred at room temperature for 30 min, then the solvents were removed under reduced pressure, and the residue was concentrated from DMF (2×). The resulting white crystalline solid was triturated with Et_2O (4 mL × 3) to remove the benzofulvene-piperidine adduct, and the residue was dried in vacuo to obtain **26** (39 mg, quantitative) as a white solid, which was used as such. MS m/z calcd for $C_{21}H_{35}N_4O_3$, 391.27; found, 391.3 [M]$^+$.

[(*R*)-5-*tert*-Butoxycarbonylamino-1-carboxy-pentyl]-trimethyl-ammonium (29). A solution of **27** (150 mg, 0.38 mmol) in MeOH (10 mL) was stirred under H_2 in the presence of 10% Pd/C (100 mg). When the deprotection reaction was complete, the mixture was filtered through Celite and the solution was concentrated in vacuo. The crude product was dissolved in 10% NaOH (4 mL), Me_2SO_4 (0.4 mL) was added, and stirring was continued in an ice bath for 30 min and then at room temperature for an additional 30 min. The reaction was neutralized by the dropwise addition of 5 N HCl, and the solution was concentrated in vacuo to obtain crude **29**, which was used as such without any further purification. MS m/z calcd for $C_{14}H_{30}N_2O_4$, 290.41; found, 290.5 [M]$^+$.

(*R*)-2,6-Bis-dimethylamino-hexanoic Acid (31). A solution of **30** (300 mg, 1.09 mmol) in CH_2Cl_2 (5 mL) and TFA (1 mL) was stirred at room temperature for 2 h. At the end of the reaction (TLC control), the solvents were distilled off in vacuo to obtain crude **31**. This was dissolved in dioxane (5 mL), and 37% aq HCHO (0.67 mL) was added, followed by a solution of $NaCNBH_3$ (164 mg, 2.6 mmol) in water. At the end of the reaction (TLC control, RP-18, AcCN/water 1:1), the solvents were distilled off under reduced pressure and the residue was purified by RP-flash chromatography to obtain **31** (127 mg, 60%). MS m/z calcd for $C_{10}H_{22}N_2O_2$, 202.30; found, 203.4 [M + H]$^+$.

(*R*)-2,6-Bis-trimethylammonium-hexanoic Acid (32). A mixture of **31** (70 mg, 0.3 mmol) in MeOH (3 mL), NaOH (70 mg, 1.75 mmol) in water (1.5 mL), and Me_2SO_4 (0.5 mL, 0.45 mmol) was stirred for 30 min in an ice bath and then for 30 min at room temperature. It was neutralized by the dropwise addition of 1 N HCl, and the solvents were distilled off in vacuo. The residue was washed with petroleum ether to leave the ammonium salts **32** as a mixture with NaCl, which was used, as such, without further purification. MS m/z calcd for $C_{12}H_{30}N_2O_2$, 232.2; found, 231.4 [M + H]$^+$.

General Synthesis of the ω Ammonium Salts 34 (Scheme 6). A solution of the commercially available Boc-protected derivatives **33** (0.43 mmol) in CH_2Cl_2 (10 mL) and TFA (1 mL) was stirred at room temperature for 1 h. The solvents were distilled off in vacuo, and the residue was dissolved in 10% NaOH (4 mL) and Me_2SO_4 (0.4 mL) was added. After 30 min, the mixture was neutralized by the dropwise addition of 1 N HCl. The solvents were distilled off in vacuo, and the residue was washed with petroleum ether to leave the ammonium salts **34** as a mixture with NaCl, which were used, as such, without further purification.

4-{*tert*-Butoxycarbonylamino-[(*E*)-*tert*-butoxycarbonylimino]-methyl}-piperazine-1-carboxylic Acid *tert*-Butyl Ester (36). A solution of *tert*-butyl 1-piperazinecarboxylate (252 mg, 1.35 mmol), DIPEA (290 μL, 1.66 mmol), and Goodman's reagent (446 mg, 1.14 mmol) in dry CH_2Cl_2 (2 mL) was stirred at room temperature overnight. At the end of the reaction (HPLC control), the solvent was removed in vacuo and the residue was partitioned between ethyl acetate (50 mL) and ice cooled 1 N HCl (50 mL). The layers were separated and the organic phase was washed with 1 N HCl (50 mL). The acid washes were combined and back-extracted with EtOAc (50 mL). The organic extracts were combined and washed with 5% $NaHCO_3$ (2 × 50 mL) and brine (50 mL), dried over Na_2SO_4, filtered, and concentrated in vacuo to obtain **36** (335 mg, 69%) as a colorless oil, which was used, as such, in the next reactions. ^{1}H NMR (300 MHz, DMSO-d_6): δ 8.95 (1H, br s), 3.34 (8H, br s), 1.45−1.34 (27H, m). HPLC purity: system B, 93.3%, t_R 16.39 min.

4-{[*tert*-Butoxycarbonyl-(3-dimethylamino-propyl)-amino]-[(*Z*)-*tert*-butoxycarbonylimino]-methyl}-piperazine-1-carboxylic Acid *tert*-Butyl Ester (37). DIAD (272 μL, 1.38 mmol) was added dropwise (at such a rate that the yellow coloration disappeared between each addition) to an ice-cold solution of **36** (198 mg, 0.462 mmol), triphenylphosphine (606 mg, 2.31 mmol), and 3-dimethylaminopropanol (163 μL, 1.38 mmol) in anhydrous THF (3 mL). When the addition was complete, the cooling bath was removed and the reaction was stirred at room temperature for 18 h. The solvent was removed in vacuo, and the residue was purified by flash chromatography, eluting with chloroform−MeOH (9:1), to give **37** (180 mg, 76%). MS m/z calcd for $C_{25}H_{47}N_5O_6$, 513.35; found, 514.2 [M + H]$^+$.

***N*-(3-Dimethylamino-propyl)-piperazine-1-carboxamidine (38).** A solution of 4 N HCl in dioxane (3 mL) was added to **37** (60 mg, 0.117 mmol) in MeOH (1 mL), and the reaction was stirred at room temperature overnight. The solvents were distilled off under reduced pressure to give **38** (40 mg, quantitative yield) as the hydrochloride salt, which was used without further purification. ^{1}H NMR (400 MHz, DMSO-d_6): δ 10.79 (1H, br s), 9.59 (2H, br s), 8.48 (1H, br s), 8.12 (2H, br s), 3.80−3.40 (8H, m), 3.38−3.29 (2H, m), 3.15−3.05 (2H, m), 2.75 (6H, s), 2.05−1.87 (2H, m).

[3-(*tert*-Butoxycarbonyl-{[(*E*)-*tert*-butoxycarbonylimino]-piperazin-1-yl-methyl}-amino)-propyl]-trimethyl-ammonium (39). A solution of **37** (50 mg, 0.097 mmol) and MeI (60 μL, 0.58 mmol) in Et_2O was stirred overnight at room temperature. A white precipitate formed, which was filtered off and dried in vacuo to give crude **39** (50 mg, 70%), which was used, as such, in the next reaction. MS *m/z* calcd for $C_9H_{21}N_5$, 199.30; found, 528.2 [M]$^+$.

Trimethyl-{3-[(piperazine-1-carboximidoyl)-amino]-propyl}-ammonium (40). A solution of 4 N HCl in dioxane (3 mL) was added to crude **39** (50 mg, 0.099 mmol) in MeOH (1 mL), and the reaction was stirred at room temperature overnight. The solvents were removed in vacuo, and the residue was triturated with Et_2O to give crude **40** as a highly hygroscopic solid, which was used without further purification. ^{1}H NMR (400 MHz, DMSO-d_6): δ 9.65 (2H, br s), 8.59 (1H, br s), 8.15 (2H, br s), 3.85−3.75 (4H, m), 3.43−3.38 (4H, m), 3.36−3.28 (4H, m), 3.11 (9H, s), 2.08−1.96 (2H, m).

***N*-{1-[4-((*S*)-2-Amino-6-dimethylamino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (44).** A solution of the Boc-Lys(Me)$_2$OH (13 mg, 0.046 mmol) in DMF (5 mL) was cooled in an ice bath. EDAC (9.0 mg, 0.046 mmol) and HOAt (6.3 mg, 0.046 mmol) were added and stirring was continued for 1 h. Amine **6a** (25 mg, 0.031 mmol) and DIPEA (8 μL, 0.046 mmol) were added and stirring was continued at room temperature overnight. At the end of the reaction (HPLC control), the solvent was distilled off in vacuo, the residue was dissolved in CH_2Cl_2 (5 mL), and TFA (1 mL) was added. At the end of the Boc-deprotection reaction (HPLC control), the solvents were distilled off again and the crude residue was purified by preparative HPLC to obtain **44** (30 mg, 88%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 9.44 (1H, br s), 8.64 (1H, s), 8.23−8.10 (3H, br s), 8.03 (1H, d), 7.83 (1H, d), 7.75 (1H, br s), 7.69−7.33 (8H, m), 5.59 (2H, s), 4.48 (1H, br s), 3.00 (1H, m), 2.78 (6H, s), 2.74−2.58 (4H, m), 2.60 (6H, s), 2.06−1.23 (14H, m). MS *m/z* calcd for $C_{36}H_{48}Cl_2N_6O_5S$, 746.28; found, 747.2 [M + H]$^+$. HPLC purity: System B, 98.7%, t_R = 8.96 min.

***N*-{1-[4-((*S*)-3-Amino-6-dimethylamino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (45).** A solution of acid **18** (9.0 mg, 0.032 mmol) in DMF (3 mL) was cooled in an ice bath. EDAC (6.0 mg, 0.032 mmol) and HOAt (4.0 mg, 0.032 mmol) were added, and the resulting mixture was stirred for an additional hour at 0 °C. Then amine **6a** (15 mg, 0.022 mmol) and DIPEA (5 μL, 0.032 mmol) were added and stirring was continued for 12 h at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, the residue was dissolved in CH_2Cl_2 (3.0 mL), and TFA (0.7 mL) was added. At the end of the Boc-deprotection reaction (HPLC control), the solvents were distilled off again and the residue was purified by preparative HPLC to obtain **45** (15 mg, 63% yield) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 9.47 (1H, br s), 8.61 (1H, s), 8.03 (1H, d), 7.82 (1H, d), 7.81 (3H, s), 7.68 (1H, d), 7.51 (1H, t), 7.39 (1H, d), 7.32 (1H, br s), 5.55 (2H, s), 3.52 (8H, m), 3.03−2.84 (2H, br s), 2.76 (3H, s), 2.63 (3H, s), 2.56 (3H, s), 2.03−1.34 (10H, m). MS *m/z* calcd for $C_{36}H_{48}Cl_2N_6O_5S$, 746.28; found, 747.3 [M + H]$^+$. HPLC purity: System B, 95%, t_R = 8.85 min.

***N*-{1-[4-((*S*)-2-Amino-6-guanidino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (46).** A solution of Boc-L-homoarginine hydrochloride (17 mg, 0.046 mmol) in DMF (3.0 mL) was cooled in an ice bath. EDAC (9.0 mg, 0.046 mmol) and HOAt (6.3 mg, 0.046 mmol) were added, and the resulting mixture was stirred for an additional hour at 0 °C. Then amine **6a** (25 mg, 0.031 mmol) and DIPEA (8 μL, 0.046 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, the residue was dissolved in CH_2Cl_2 (3.0 mL), and TFA (1.0 mL) was added. At the end of the Boc-deprotection reaction (HPLC control), the solvents were distilled off again and the residue was purified by preparative HPLC to afford **46** (27 mg, 79%). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.65 (1H, s), 8.14 (3H, br s), 8.04 (1H, d), 7.83 (1H, d), 7.81−7.44 (5H, m), 7.39−6.76 (3H, s), 5.50 (2H, s), 4.46 (1H, br s), 3.07 (2H, m), 2.72 (3H, s), 2.67 (3H, s), 2.03−1.91 (2H, m), 1.80−1.61 (4H, m), 1.53−1.25 (10H, m). MS *m/z* calcd for $C_{35}H_{46}Cl_2N_8O_5S$, 760.27; found, 761.1 [M + H]$^+$. HPLC purity: System B, 95%, t_R = 8.80 min.

2,4-Dichloro-*N*-{1-[4-((*S*)-2,6-diamino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (47). A solution of Boc-D-Lys(Boc)-OH (20 mg, 0.060 mmol) in DMF (3.0 mL) was cooled in an ice bath. EDAC (11.0 mg, 0.060 mmol) and HOAt (8.0 mg, 0.060 mmol) were added, and the resulting mixture was stirred for an additional hour at 0 °C. Amine **6a** (30 mg, 0.040 mmol) and DIPEA (9.0 μL, 0.05 mmol) were added, and stirring was continued at room temperature overnight. At the end of the reaction, the solvents were distilled off in vacuo, the residue was diluted with CH_2Cl_2 (2.0 mL), and TFA (0.5 mL) added. At the end of the Boc-deprotection reaction, solvents were distilled off in vacuo, and the residue was purified by preparative HPLC to obtain **47** (42 mg, 96%) in the form of trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.32 (1H, s), 8.10 (3H, br s), 8.02 (1H, d), 7.80 (3H, br s), 7.77 (1H, d), 7.76−7.33 (2H, m), 7.45 (1H, t), 7.30 (2H, m), 5.68 (2H, s), 4.48 (1H, br s), 2.81 (2H, br s), 2.79 (3H, s), 2.68 (3H, s), 2.02 (2H, s), 1.80 (4H, br s), 1.45−1.60 (8H, m). MS *m/z* calcd for $C_{34}H_{44}Cl_2N_6O_5S$, 718.25; found, 719.2 [M + H]$^+$. HPLC purity: System B, 98%, t_R = 8.51 min.

***N*-{1-[4-((*S*)-2,6-Bis-dimethylamino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (48).** A solution of (*S*)-2,6-bis-dimethylaminohexanoic acid (20 mg, 0.096 mmol), EDAC (28 mg, 0.096 mmol), and HOAt (20 mg, 0.096 mmol) in DMF (5.0 mL) was cooled in an ice bath. After 1 h, amine **6a** (50 mg, 0.072 mmol) and DIPEA (11 μL, 0.072 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, and the crude product was purified by preparative HPLC to obtain **48** (50.5 mg, 63%) as a trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 9.40 (1H, br s), 8.32 (1H, s), 8.02 (1H, d), 7.77 (1H, d), 7.76−7.33 (4H, m), 5.68 (2H, s), 4.48 (1H, br s), 3.89−3.45 (8H, m), 3.18−3.04 (2H, m), 2.81 (3H, s), 2.79 (3H, s), 2.68 (3H, s), 2.64 (3H, s), 2.09−1.28 (10H, m). MS *m/z* calcd for $C_{38}H_{52}Cl_2N_6O_5S$, 774.31; found, 775.3 [M + H]$^+$. HPLC purity: System B, >99%, t_R = 8.65 min.

[(*S*)-5-Amino-6-(4-{1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-6-oxo-hexyl]-trimethyl-ammonium Trifluoroacetate Salt (49). A solution of (*S*)-2-Fmoc-amino-6-trimethylammoniumhexanoic acid (20 mg, 0.045 mmol), EDAC (13 mg, 0.067 mmol), and HOAt (9 mg, 0.067 mmol) in DMF (5.0 mL) was cooled in an ice bath. After 1 h, amine **6a** (30 mg, 0.045 mmol) and DIPEA (7 μL, 0.045 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the residue was diluted with CH_2Cl_2 (2.0 mL) and piperidine (1 mL). At the end of the Fmoc-deprotection reaction, the crude product was purified by preparative HPLC to obtain **49** (15 mg, 29%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.66 (1H, s), 8.27−8.12 (3H, br s), 8.04 (1H, d), 7.84 (1H, d), 7.81−7.37 (4H, m), 5.60 (2H, s), 4.60−4.42 (1H, br s), 3.70−3.42 (8H, m), 3.24 (2H, m), 3.15 (9H, s), 2.75 (3H, s), 2.67 (3H, s), 2.04−1.93 (2H, m), 1.82−1.22 (14H, m). MS *m/z* calcd for $C_{37}H_{51}Cl_2N_6O_5S$, 761.3; found, 761.4 [M]$^+$. HPLC purity: System B, 98.8%, t_R = 8.61 min.

2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-*N*-{1-[4-(6-guanidino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-benzenesulfonamide Trifluoroacetate Salt (50). A solution of 6-Boc-amino-hexanoic acid (7.4 mg, 0.032 mmol) in DMF (2.0 mL) was cooled in an ice bath. EDAC (6.0 mg, 0.032 mmol) and HOAt (4.3 mg, 0.032 mmol) were added, and the resulting mixture was stirred for an additional hour at 0 °C. Amine **6a** (15 mg, 0.022 mmol) and DIPEA (5.0 μL, 0.032 mmol) were added and stirring was continued at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, and the residue was treated with CH_2Cl_2 (2 mL) and TFA (0.5 mL). At the end of the Boc-deprotection reaction (HPLC control), the solvents were distilled off in vacuo, and the residue was purified by preparative HPLC. The resulting amine was dissolved in CH_2Cl_2, Goodman's reagent (15 mg, 0.037 mmol) and DIPEA (7 mg, 0.055 mmol) were added, and the resulting mixture was stirred at room temperature overnight. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, and the residue was purified by preparative HPLC to obtain **50** (10 mg, 47%) as trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.44 (1H, s), 8.02 (1H, d), 7.79 (1H, d), 7.76−7.32 (5H, m), 7.05−6.84 (4H, br s), 5.55 (2H, s), 3.66−3.47 (8H, m), 3.10 (2H, m), 2.68 (3H, s), 2.52 (3H, s), 2.38−2.32 (2H, m), 2.08−1.23 (14H, m). MS *m/z* calcd for $C_{35}H_{45}Cl_2N_7O_5S$, 745.26; found, 746.5 $[M + H]^+$. HPLC purity: System B, 96.5%, $t_R = 10.17$ min.

[6-(4-{1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-6-oxo-hexyl]-trimethyl-ammonium Trifluoroacetate Salt (51). A solution of (5-carboxy-pentyl)-trimethyl-ammonium (80 mg, 0.17 mmol), EDAC (32 mg, 0.17 mmol), and HOAt (23 mg, 0.17 mmol) in DMF (5 mL) was cooled in an ice bath. After 1 h, amine **6a** (30 mg, 0.04 mmol) and DIPEA (6 μL, 0.04 mmol) were added, and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, and the residue was purified by preparative HPLC to give **51** (24 mg, 61%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.30 (1H, s), 8.07 (1H, d), 8.02−7.94 (1H, m), 7.91−7.76 (4H, m), 5.74 (2H, s), 3.67−3.50 (8H, m), 3.34−3.26 (2H, m), 3.07 (9H, s), 2.92 (3H, s), 2.68 (3H, s), 2.91 (3H, s), 2.43−2.36 (2H, m), 2.12−1.33 (14H, m). MS *m/z* calcd for $C_{37}H_{50}Cl_2N_5O_5S$, 746.29; found, 746.2 $[M]^+$. HPLC purity: System B, 99%, $t_R = 9.99$ min.

[(*S*)-1-(4-{1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazine-1-carbonyl)-5-guanidino-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (52). A solution of **54** (6.0 mg, 0.005 mmol) in CH_2Cl_2 (2.0 mL) was treated with Goodman's reagent (8.0 mg, 0.02 mmol) and DIPEA (0.04 mmol) and stirred at room temperature for 1 h. At the end of the reaction (HPLC control), TFA was added (0.5 mL) and stirring was continued until deprotection of Boc groups was complete (HPLC control). The solvents were distilled off, and the residue was purified by preparative HPLC to obtain **52** (5.0 mg, 87%) as trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.39 (1H, s), 8.02 (1H, d), 7.78 (1H, d), 7.76 (1H, d), 7.58 (1H, m), 7.35−7.30 (3H, m), 6.95 (4H, br s), 5.65 (2H, s), 4.61 (1H, d), 3.85−3.62 (10H, m), 3.05 (9H, s), 2.72 (3H, s), 2.68 (3H, s), 1.80−1.69 (3H, m), 1.60−1.53 (7H, m), 1.48−1.40 (4H, m). MS *m/z* calcd for $C_{38}H_{53}Cl_2N_8O_5S$, 803.32; found, 803.3 $[M + H]^+$. HPLC purity: System B, 97%, $t_R = 7.84$ min.

***N*-[1-[4-(2-(*S*)-trimethylammonium-6-trimethylammonium-hexanoyl)-piperazin-1-yl]-cyclopentyl]-2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (53).** A solution of acid **36** (10 mg, 0.04 mmol), EDAC (5.0 mg, 0.04 mmol), and HOAt (8.0 mg, 0.04 mmol) in DMF (5.0 mL) was cooled in an ice bath. After 1 h, amine **6a** (30 mg, 0.046 mmol) and DIPEA (7 μL, 0.046 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off and the crude residue was purified by preparative HPLC to obtain **53** (20.5 mg, 39%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.32 (1H, s), 8.02 (1H, d), 7.78 (1H, d), 7.73 (1H, d), 7.59−7.31 (3H, m), 5.68 (2H, s), 4.68−4,60 (1H, m), 4.01−3.56 (8H, m), 3.36−3.28 (2H, m), 3.22 (9H, s), 3.08 (9H, s), 2.68 (3H, s), 2.63 (3H, s), 2.13−1.43 (14H, m). MS *m/z* calcd for $C_{40}H_{58}Cl_2N_6O_5S$, 804.3; found, 803.3 $[M - H]^+$. HPLC purity: System B, 99.6%, $t_R = 8.80$ min.

[(*S*)-5-Amino-1-(4-{1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazine-1-carbonyl)-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (54). A solution of crude (*S*)-(5-*tert*-butoxycarbonylamino-1-carboxy-pentyl)-trimethyl-ammonium (50 mg, 0.17 mmol), EDAC (32 mg, 0.17 mmol), and HOAt (23 mg, 0.17 mmol) in DMF (5 mL) was cooled in an ice bath. After 1 h, amine **6a** (50 mg, 0.11 mmol) and DIPEA (25 μL, 0.11 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off and the residue was dissolved in CH_2Cl_2/TFA (3:1; 5.0 mL). After deprotection of the Boc group, the solvents were distilled off and the crude product was purified by preparative HPLC to afford **54** (21 mg, 17%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.35 (1H, s), 8.07 (1H, d), 8.01−7.94 (1H, m), 7.87 (2H, s), 7.80 (1H, d), 7.77−7.59 (3H, br s), 5.74 (2H, s), 4.65−4.58 (1H, m), 3.98−3.51 (8H, m), 3.22 (9H, s), 2.91 (6H, s), 2.82−2.80 (2H, m), 2.13−1.41 (14H, m). MS *m/z* calcd for $C_{37}H_{51}Cl_2N_6O_5S$, 761.3; found, 761.4 $[M]^+$. HPLC purity: System B, 99.2%, $t_R = 8.64$ min.

2,4-Dichloro-*N*-{1-[4-((*S*)-3,6-diamino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (55). A solution of β-Boc-lysine (22 mg, 0.064 mmol), HOAt (87 mg, 0.043 mmol), and EDAC (12.2 mg, 0.043 mmol) in DMF (3.0 mL) was stirred at room temperature for 1 h. Then a solution of **6a** (30 mg, 0.043 mmol) and DIPEA (10 μL, 0.043 mmol) in DMF (2.0 mL) was added and stirring was continued at room temperature overnight. At the end of the reaction (HPLC control), the solvent was distilled off and the residue was dissolved in TFA/CH_2Cl_2 (1:1; 3.0 mL). At the end of the reaction (HPLC control), the solvents were distilled off and the crude product was purified by preparative HPLC to obtain **55** (28 mg, 61%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.62 (1H, s), 8.04 (1H, d), 7.90−7.32 (12H, m), 5.59 (2H, s), 3.58−3.41 (8H, m), 2.86−2.56 (9H, m), 2.03−1.21 (12H, m). MS *m/z* calcd for $C_{34}H_{44}Cl_2N_6O_5S$, 718.25.72; found, 719.4 $[M + H]^+$. HPLC purity: System B, 99%, $t_R = 8.79$ min.

[(*S*)-4-Amino-6-(4-{1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-6-oxo-hexyl]-trimethyl-ammonium Trifluoroacetate Salt (56). A solution of acid **4a** (65 mg, 0.124 mmol), HOAt (22 mg, 0.16 mmol), and EDAC (32 mg, 0.17 mmol) in DMF (2 mL) was stirred in an ice bath for 30 min, then a solution of **21** (58 mg, 0.12 mmol) in DMF (2 mL) was added, followed by dropwise addition of DIPEA (42 μL, 0.24 mmol). Stirring was continued at 0 °C for 30 min, then at room temperature overnight. At the end of the reaction (HPLC control) the solvents were removed in vacuo. The residue was dissolved in CH_3CN/water (1:1) and purified by preparative HPLC to obtain the Boc-protected intermediate as trifluoroacetate salt. This compound was dissolved in TFA/CH_2Cl_2 (1:1; 4 mL) and stirred at room temperature overnight. The solvents were distilled off in vacuo, and the resulting product was purified by preparative HPLC to afford **56** (44 mg, 56%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.27 (1H, s), 8.08−7.99 (1H, m), 7.95−7.72 (4H, m), 7.59−7.51 (1H, m), 7.47−7.37 (2H, m), 5.68 (2H, m), 3.76−3.48 (8H, m), 3.37−3.24 (2H, m), 3.13−3.06 (9H, m), 2.92−2.79 (1H, m), 2.76−2.63 (7H, m), 2.13−1.99 (2H, m), 1.93−1.74 (3H, m), 1.73−1.60 (2H, m), 1.56−1.42 (4H, m). MS *m/z* calcd for $C_{37}H_{51}Cl_2N_6O_5S$, 761.30; found, 761.2 $[M]^+$. HPLC purity: System B, 98.6%, $t_R = 10.26$ min.

{(*S*)-4-Amino-1-[2-(4-{1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-2-oxo-ethyl]-butyl}-trimethyl-ammonium Trifluoroacetate Salt (57). A solution of acid **4a** (58 mg, 0.11 mmol), HOAt (19 mg, 0.14 mmol), and EDAC (32 mg, 0.17 mmol) in DMF (2.0 mL) was stirred in an ice bath for 30 min. Then a solution of amine **26** (39 mg, 0.073 mmol) in DMF (2.0 mL) was added while stirring, followed by the dropwise addition of DIPEA (13 μL, 0.075 mmol). Stirring was continued at 0 °C for an additional 30 min, then at room temperature overnight. At the end of the reaction (HPLC control), the solvents were removed in vacuo, and the residue was dissolved in CH_3CN/water + 0.1% TFA (1:1; 6.0 mL) and purified by preparative HPLC to obtain the Cbz-protected derivative (36.9 mg, 45%). A portion of this product (14.9 mg, 0.0133 mmol) was dissolved in TFA (1.5 mL), and triflic acid

(15 μL, 0.17 mmol) was added. At the end of the reaction (HPLC control), water (4.0 mL) was added, the resulting mixture was filtered, and the filtrate was purified by preparative HPLC to obtain **57** (15 mg, quantitative) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.25 (1H, s), 8.02 (1H, d), 7.79−7.69 (5H, m), 7.56−7.47 (1H, m), 7.44−7.32 (2H, m), 5.67 (2H, s), 3.74−3.54 (8H, m), 3.23−3.16 (1H, m), 3.09 (9H, s), 2.93−2.71 (4H, m), 2.68 (3H, s), 2.64 (3H, s), 2.14−2.00 (3H, m), 1.83−1.73 (2H, m), 1.72−1.55 (3H, m), 1.53−1.41 (4H, m). MS *m/z* calcd for $C_{37}H_{51}Cl_2N_6O_5S$, 761.30; found, 761.3 [M]$^+$. HPLC purity: System B, 97.9%, t_R = 10.02 min.

***N*-{1-[4-((*S*)-3-Amino-6-guanidino-hexanoyl)-piperazine-1-carbonyl]-cyclopentyl}-2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonamide Trifluoroacetate Salt (58).** A solution of acid **17** (18 mg, 0.036 mmol) in DMF (2.0 mL) was cooled in an ice bath. EDAC (8.0 mg, 0.036 mmol) and HOAt (5.0 mg, 0.036 mmol) were added, and stirring was continued for 1 h. Amine **6a** (20 mg, 0.024 mmol) and DIPEA (9.0 μL, 0.048 mmol) were added, and the resulting mixture was stirred at room temperature overnight. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo, and the residue was dissolved in CH_2Cl_2 (2.0 mL) and TFA (0.5 mL). After 12 h, the solvents were distilled off and the residue was purified by preparative HPLC to obtain **58** (6.0 mg, 23%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.44 (1H, s), 8.02 (1H, d), 7.79 (1H, d), 7.77−7.67 (3H, m), 7.50 (1H, t), 7.43 (1H, t), 7.37 (1H, d), 7.32 (1H, br s), 7.10−6.90 (4H, br s), 5.61 (2H, s), 3.77−3.41 (9H, m), 3.02 (2H, m), 2.79−2.68 (2H, m), 2.66 (3H, s), 2.59 (3H, s), 2.06−1.37 (12H, m). MS *m/z* calcd for $C_{35}H_{46}Cl_2N_8O_5S$, 760.27; found, 761.3 [M + H]$^+$. HPLC purity: System B, 91%, t_R = 9.02 min.

[2-(4-{1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-2-oxo-ethyl]-trimethyl-ammonium Trifluoroacetate Salt (59). A solution of carboxymethyl-trimethyl-ammonium chloride (6.24 mg, 0.06 mmol), EDAC (15.2 mg, 0.17 mmol), and HOAt (23 mg, 0.17 mmol) in DMF (5.0 mL) was stirred in an ice bath. After 1 h, amine **6a** (30 mg, 0.04 mmol) and DIPEA (11 μL, 0.04 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvent was distilled off in vacuo and the crude product was purified by preparative HPLC to obtain **59** (27 mg, 73%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.27 (1H, s), 8.03 (1H, d), 7.77 (1H, d), 7.74−7.37 (4H, m), 5.69 (2H, s), 4.51 (2H, s), 3.75−3.47 (8H, m), 3.29 (9H, s), 2.70 (3H, s), 2.66 (3H, s), 2.11−2.01 (2H, m), 1.84−1.73 (2H, m), 1.53−1.43 (4H, m). MS *m/z* calcd for $C_{32}H_{42}Cl_2N_5O_5S$, 690.23; found, 690.1 [M]$^+$. HPLC purity: System B, 98.1%, t_R = 9.64 min.

[4-(4-{1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-4-oxo-butyl]-trimethyl-ammonium Trifluoroacetate Salt (60). A solution of (3-carboxypropyl)trimethylammonium chloride (10 mg, 0.068 mmol), EDAC (16 mg, 0.085 mmol), and HOAt (10 mg, 0.073 mmol) in DMF (5.0 mL) was stirred in an ice bath. After 1 h, amine **6a** (30 mg, 0.04 mmol) and DIPEA (14 μL, 0.05 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the crude product was purified by preparative HPLC to obtain **60** (21 mg, 55%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.25 (1H, s), 8.02 (1H, d), 7.82−7.70 (2H, m), 7.58−7.33 (3H, m), 5.68 (2H, s), 3.64 (4H, br s), 3.55 (4H, br s), 3.37−3.28 (2H, m), 3.10 (9H, s), 2.69 (3H, s), 2.65 (3H, s), 2.50−2.44 (2H, m), 2.11−1.73 (6H, m), 1.55−1.42 (4H, br s). MS *m/z* calcd for $C_{35}H_{46}Cl_2N_5O_5S$, 718.24; found, 718.2 [M]$^+$. HPLC purity: System B, 97.6%, t_R = 9.71 min.

[(*S*)-3-Amino-4-(4-{1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-4-oxo-butyl]-trimethyl-ammonium Trifluoroacetate Salt (61). A solution of **10a** (13 mg, 0.05 mmol), EDAC (10 mg, 0.05 mmol), and HOAt (7.0 mg, 0.05 mmol) in DMF (5.0 mL) was stirred in an ice bath. After 1 h, amine **6a** (25 mg, 0.04 mmol) and DIPEA (11 μL, 0.04 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the residue was dissolved in CH_2Cl_2 (2.0 mL) and TFA (0.5 mL). After complete removal of the Boc group, the solvents were distilled off and the crude product was purified by preparative HPLC to obtain **61** (5 mg, 12%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.61 (1H, s), 8.32 (3H, br s), 8.02 (1H, d), 7.78 (1H, d), 7.81−7.35 (3H, m), 5.62 (2H, s), 4.58 (1H, br s), 3.85 (8H, m), 3.15 (9H, s), 2.72 (3H, s), 2.68 (3H, s), 2.22 (2H, m), 1.80 (2H, m), 1.60 (2H, m). MS *m/z* calcd for $C_{35}H_{47}Cl_2N_6O_5S$, 733.27; found, 733.2 [M]$^+$. HPLC purity: System B, 97%, t_R = 8.58 min.

[5-(4-{1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-5-oxo-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (62). A solution of (4-carboxybutyl)-trimethyl ammonium (7.0 mg, 0.04 mmol), EDAC (5.0 mg, 0.04 mmol), and HOAt (8.0 mg, 0.04 mmol) in DMF (5.0 mL) was stirred in an ice bath. After 1 h, amine **6a** (20 mg, 0.03 mmol) and DIPEA (8 μL, 0.03 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the crude product was purified by preparative HPLC to obtain **62** (20 mg, 69%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.39 (1H, s), 8.03 (1H, d), 7.78 (1H, d), 7.78−7.76 (1H, m), 7.60 (1H, t), 7.54−7.44 (2H, m), 5.65 (2H, s), 3.62 (4H, br s), 3.54 (4H, br s), 3.33−3.27 (2H, m), 3.05 (9H, s), 2.72 (3H, s), 2.68 (3H, s), 2.43 (2H, m), 2.06−1.99 (2H, m), 1.80−1.69 (4H, m), 1.60−1.53 (2H, m), 1.48−1.40 (4H, m). MS *m/z* calcd for $C_{36}H_{48}Cl_2N_5O_5S$, 732.27; found, 732.4 [M]$^+$. HPLC purity: System B, 97.2%, t_R = 9.27 min.

[(*S*)-4-Amino-5-(4-{1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazin-1-yl)-5-oxo-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (63). A solution of **10b** (19 mg, 0.07 mmol), EDAC (13 mg, 0.07 mmol), and HOAt (9.5 mg, 0.07 mmol) in DMF (5.0 mL) was stirred in an ice bath. After 1 h, amine **6a** (34 mg, 0.05 mmol) and DIPEA (24 μL, 0.09 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the residue was dissolved in CH_2Cl_2 (2.0 mL) and TFA (0.5 mL). When removal of the Boc group was complete, the solvents were distilled off and the crude product was purified by preparative HPLC to obtain **63** (15 mg, 27%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.33 (1H, s), 8.21 (3H, br s), 8.15 (1H, d), 7.78 (1H, d), 7.75 (1H, d), 7.58−7.50 (1H, t), 7.47−7.35 (2H, m), 5.64 (2H, s), 4.45 (1H, br s), 3.75 (8H, br s), 3.15 (9H, s), 2.72 (3H, s), 2.68 (2H, s), 1.77 (4H, m), 1.45 (4H, m). MS *m/z* calcd for $C_{36}H_{49}Cl_2N_6O_5S$, 747.8; found, 747.3 [M]$^+$. HPLC purity: System B, 97%, t_R = 8.37 min.

[5-(4-(1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl]-piperazin-1-yl)-4(*S*)-trimethylammonio-5-oxo-pentyl]trimethyl-ammonium Trifluoroacetate Salt (64). A solution of **14** (9.0 mg, 0.04 mmol), EDAC (5.0 mg, 0.04 mmol), and HOAt (8.0 mg, 0.04 mmol) in DMF (5.0 mL) was stirred in an ice bath. After 1 h, amine **6a** (20 mg, 0.03 mmol) and DIPEA (8 μL, 0.03 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the crude product was purified by preparative HPLC to obtain **64** (9.0 mg, 26%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.43 (1H, s), 8.03−8.00 (1H, d), 7.81−7.79 (1H, d), 7.75−7.73 (1H, d), 7.54 (1H, t), 7.44−7.37 (2H, m), 5.64 (2H, s), 4.65 (1H, d), 3.34 (2H, m), 3.20 (6H, s), 3.01 (9H, s), 2.81 (3H, s), 2.73 (3H, s), 2.68 (3H, s), 2.01 (2H, m), 1.77 (2H, m), 1.47 (4H, m). MS *m/z* calcd for $C_{35}H_{47}Cl_2N_6O_5S$, 790.34; found, 777.5 [M − CH_3]$^+$. HPLC purity: System B, >99%, t_R = 8.55 min.

[(*S*)-4-Amino-1-(4-{1-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazine-1-carbonyl)-butyl]-trimethyl-ammonium Trifluoro-

acetate Salt (65). A solution of **12** (11 mg, 0.04 mmol), EDAC (6.0 mg, 0.04 mmol), and HOAt (8.0 mg, 0.04 mmol) in DMF (5.0 mL) was stirred in an ice bath. After 1 h, amine **6a** (30 mg, 0.04 mmol) and DIPEA (11 μL 0.04 mmol) were added and stirring was continued overnight at room temperature. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the residue was dissolved in CH_2Cl_2 (2.0 mL) and TFA (0.5 mL). When removal of the Boc group was complete, the solvents were distilled off and the crude product was purified by preparative HPLC to obtain **65** (18 mg, 41%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.43 (1H, s), 8.03 (1H, d), 7.80−7.78 (2H, d), 7.74−7.72 (2H, d), 7.54 (1H, t), 7.43−7.38 (2H, m), 5.64 (2H, s), 4.69 (2H, d), 3.20 (9H, s), 2.85 (2H, s), 2.68 (3H, s), 2.63 (3H, s), 2.00 (2H, m), 1.76 (2H, m), 1.46 (4H, m). MS *m/z* calcd for $C_{36}H_{49}Cl_2N_6O_5S$, 747.28; found, 747.3 [M]$^+$. HPLC purity: System B, 98.7%, t_R = 8.64 min.

4-{1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-*N*-(3-dimethyl-amino-propyl)-piperazine-1-carboxamidine Trifluoroacetate Salt (66). A solution of acid **4a** (78 mg, 0.15 mmol), HOAt (20 mg, 0.15 mmol), and EDAC (28.5 mg, 0.15 mmol) in DMF (3.0 mL) was stirred at room temperature for 25 min. Then a solution of amine **38** (40 mg, 0.124 mmol) and DIPEA (65 μL, 0.37 mmol) in DMF (2.0 mL) was added and stirring was continued at room temperature overnight. At the end of the reaction (HPLC control), the solution was concentrated to 2.5 mL and purified by HPLC to give **66** (80 mg, 50%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 9.78−9.40 (1H, br s), 8.35−8.22 (1H, m), 8.06−7.94 (1H, m), 7.82−7.57 (5H, m), 7.57−7.46 (1H, m), 7.46−7.32 (2H, m), 5.71−5.63 (2H, m), 3.80−3.66 (4H, m), 3.59−3.48 (4H, m), 3.36−3.26 (2H, m), 3.15−3.06 (2H, m), 2.86−2.78 (6H, m), 2.73−2.58 (6H, m), 2.12−1.98 (2H, m), 1.98−1.87 (2H, m), 1.83−1.72 (2H, m), 1.54−1.41 (4H, m). MS *m/z* calcd for $C_{34}H_{45}Cl_2N_7O_4S$, 717.26; found, 718.2 [M + H]$^+$. HPLC purity: System B, 93.4%, t_R = 10.14 min.

{3-[(4-{1-[2,4-Dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-cyclopentanecarbonyl}-piperazine-1-carboximidoyl)-amino]-propyl}-trimethyl-ammonium Trifluoroacetate Salt (67). An ice-cold solution of **4a** (121 mg, 0.23 mmol), HOAt (31 mg, 0.23 mmol), and EDAC (44 mg, 0.23 mmol) in DMF (5.0 mL) was stirred for 20 min. Then a solution of the amine **40** (65 mg, 0.19 mmol) and DIPEA (19 μL, 0.57 mmol) in DMF (2.0 mL) was added. At the end of the reaction, the solvents were distilled off in vacuo and the crude product was purified by preparative HPLC to afford **67** (7.5 mg, 4%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.37−8.26 (1H, m), 8.07−7.98 (1H, m), 7.83−7.66 (4H, m), 7.56−7.46 (1H, m), 7.44−7.31 (2H, m), 5.73−5.64 (2H, m), 3.82−3.71 (4H, m), 3.62−3.52 (5H, m), 3.41−3.26 (4H, m), 3.18−3.06 (9H, m), 2.74−2.60 (6H, m), 2.14−1.96 (5H, m), 1.85−1.73 (2H, m), 1.56−1.43 (4H, m). MS *m/z* calcd for $C_{35}H_{48}Cl_2N_7O_4S$, 732.29; found, 732.1 [M]$^+$. HPLC purity: System B, 98.5%, t_R = 9.90 min.

[(*S*)-4-Amino-5-(4-{4-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-tetrahydro-thiopyran-4-carbonyl}-piperazin-1-yl)-5-oxo-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (68). A solution of the acid **10b** (36 mg, 0.13 mmol), EDAC (75 mg, 0.39 mmol), and HOAt (18 mg, 0.13 mmol) in DMF (3.0 mL) was stirred at room temperature for 1 h. Then a solution of amine **6d** (86 mg, 0.13 mmol) and DIPEA (44 μL, 0.26 mmol) in DMF (1.0 mL) was added and stirring was continued for 1 h. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the residue was dissolved in TFA (2.0 mL) and stirred at room temperature for 40 min. Et_2O was added with stirring and a product was precipitated that was filtered off. Purification by preparative HPLC afforded **68** (14 mg, 11%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.80 (1H, s), 8.0−7.40 (6H, m), 5.60 (1H, s), 4.55 (1H, s), 3.80−3.30 (8H, m), 3.0 (9H, s), 2.70 6H, s), 2.40−2.00 (8H, m), 1.75 (4H, d). MS *m/z* calcd for $C_{36}H_{49}Cl_2N_6O_5S_2$, 779.25; found, 779.2 [M]$^+$. HPLC purity: System A, >99%, t_R = 7.52 min.

[(*S*)-4-Amino-5-(4-{4-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-piperidine-4-carbonyl}-piperazin-1-yl)-5-oxo-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (69). A solution of the acid **10b** (10 mg, 0.036 mmol), EDAC (26 mg, 0.036 mmol), and HOAt (6.0 mg, 0.044 mmol) in DMF (3.0 mL) was stirred at room temperature for 1 h. Amine **6e** (26 mg, 0.036 mmol) and DIPEA (15 μL, 0.088 mmol) in DMF (1.0 mL) were added and stirring was continued for 1.5 h. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the residue was dissolved in TFA/CH_2Cl_2 (1:1; 4.0 mL) and stirred at room temperature for 40 min. The solvents were distilled off in vacuo, and the residue was purified by preparative HPLC to afford **69** (14.9 mg) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6): δ 9.20 (1H, s), 8.50 (2H, s), 8.25 (3H, s), 8.10−7.30 (6H, m), 5.60 (1H, s), 4.55 (1H, s), 4.00−3.30 (8H, m), 3.30−2.90 (13H, m), 2.80−2.55 (9H, m), 2.25−1.70 (8H, m). MS *m/z* calcd for $C_{36}H_{50}Cl_2N_7O_5S$, 762.29; found, 762.3 [M]$^+$. HPLC purity: System A, >99%, t_R = 5.17 min.

[(*S*)-4-Amino-5-(4-{4-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-1-methyl-piperidine-4-carbonyl}-piperazin-1-yl)-5-oxo-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (70). Crude **6e** (114 mg, 0.072 mmol) was dissolved in dry DMF (1 mL), and DIPEA was added until the pH was about 7. Acid **10** (22 mg, 0.079 mmol), HOAt (10.8 mg, 0.079 mmol), EDAC (15.2 mg, 0.079 mmol), and DIPEA (13.5 μL, 0.079 mmol) were added, and the mixture was stirred at room temperature overnight. The solvents were distilled off under reduced pressure, the residue was dissolved in dry DCM (1.5 mL), and TFA (1.5 mL) was added dropwise while stirring, After 30 min, the solvents were distilled off and the residue was purified by preparative HPLC to obtain **70** (15 mg, 21%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6, 353 °K): δ 8.90 (1H, s), 8.19 (2H, br s), 8.05 (1H, d, J = 8.7 Hz), 7.80 (1H, d, J = 8.7 Hz), 7.74 (1H, d, J = 8.3 Hz), 7.52 (1H, t, J = 8.1 Hz), 7.41 (1H, d, J = 7.7 Hz), 7.33 (1H, s), 5.66 (2H, s), 4.47 (1H, s), 4.13−3.38 (12H, covered by water), 3.38−3.27 (2H, m), 3.07 (9H, s), 2.69−2.64 (6H, m), 2.61 (3H, s), 2.38−1.95 (4H, m), 1.89−1.70 (4H, m). MS *m/z* calcd for $C_{37}H_{52}Cl_2N_7O_5S$, 776.31; found, 776.3 [M]$^+$. HPLC purity: System A, >99%, t_R = 8.20 min.

[(*S*)-5-(4-{1-Acetyl-4-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-piperidine-4-carbonyl}-piperazin-1-yl)-4-amino-5-oxo-pentyl]-trimethyl-ammonium Trifluoroacetate Salt (71). A solution of acid **10** (8.0 mg, 0.026 mmol), HOAt (4.0 mg, 0.026 mmol), EDAC (5.6 mg, 0.026 mmol), and DIPEA (4.5 μL, 0.026 mmol) in DMF (2 mL) was stirred at room temperature for 1 h and then added to crude **6f** (18 mg, 0.025 mmol) dissolved in dry DMF (1 mL). At the end of the reaction (HPLC control), the solvents were distilled off under reduced pressure and the residue was dissolved in DCM/TFA (1:1; 2 mL) and stirred for 2 h. The solvents were distilled off, and the crude product was purified by preparative HPLC to obtain **71** (10 mg, 38%) as the trifluoroacetate salt. ^{1}H NMR (400 MHz, DMSO-d_6, 353 °K): δ 8.55 (1H, s), 8.15 (2H, br s), 8.02 (1H, d, J = 8.7 Hz), 7.77 (1H, d, J = 8.7 Hz), 7.70 (1H, d, J = 8.2 Hz), 7.47 (1H, t, J = 8.2 Hz), 7.36−7.29 (2H, m), 5.67 (2H, s), 4.46 (1H, s), 3.93−3.56 (8H, m), 3.55−3.06 (6H, m, covered by water), 3.07 (9H, s), 2.65 (3H, s), 2.61 (3H, s), 1.89 (3H, s), 1.85−1.71 (8H, m). MS *m/z* calcd for $C_{38}H_{52}Cl_2N_7O_6S$, 804.3; found, 804.2 [M]$^+$. HPLC purity: System B, 97%, t_R = 14.23 min.

[(*S*)-5-Amino-6-(4-{4-[2,4-dichloro-3-(2,4-dimethyl-quinolin-8-yloxymethyl)-benzenesulfonylamino]-tetrahydro-pyran-4-carbonyl}-piperazin-1-yl)-6-oxo-pentyl]-trimethyl-ammonium Hydrochloride Salt (72). A solution of the acid **10** (75 mg, 0.27 mmol), EDAC (62 mg, 0.32 mmol), and HOAt (44 mg, 0.32 mmol) in DMF (3.0 mL) was stirred at room temperature for 1 h. Amine **6b** (150 mg, 0.18 mmol) and DIPEA (110 μL, 0.6 mmol) in DMF (1.0 mL) were added, and stirring was continued for 20 h. At the end of the reaction (HPLC control), the solvents were distilled off in vacuo and the residue was dissolved in CH_2Cl_2, washed with 5% $NaHCO_3$ and satd NaCl, dried over Na_2SO_4, and concentrated under reduced pressure. The residual oil was dissolved in a small volume of CH_2Cl_2 (1.0 mL) and precipitated with Et_2O. Filtration gave the crude Boc-protected derivative (145 mg, 0.17 mmol, 94%). This was dissolved in CH_2Cl_2 (3.0 mL), and 4 N HCl in dioxane (3.0 mL) was added. After stirring at room temperature for 30 min, the solvents were removed in vacuo. Trituration of the residue with Et_2O afforded **72** (**MEN 16132**; 110 mg, 73%) as the hydrochloride salt. ^1H NMR (400 MHz, DMSO-d_6): δ 8.85 (1H, s), 8.50 (3H, s), 8.02 (1H, d), 7.90−7.60 (4H, m), 5.59 (2H, s), 4.57−4.45 (1H, m), 3.70−3.18 (12H, m), 3.08 (9H, s), 2.81 (6H, m), 1.95−1.60 (8H, m). MS *m/z* calcd for $C_{36}H_{49}Cl_2N_6O_6S$, 763.2; found, 763.1 $[M]^+$. HPLC purity: System B, 97%, t_R = 7.40 min.

(B) Biology. The receptor binding assays and the measurement of inositol monophosphate accumulation were carried out as described in ref 11. The measurement of the antagonist potency (pA_2) in the BK-induced contraction of the GPI was performed as described in ref 13.

In vivo experiments were performed in male Dunkin Hartley guinea pigs weighing 350−400 g (Charles River, Italy) in accordance with the European Union and the local ethical commitee regulations. Evaluation of bronchoconstriction and hypotension induced by iv bradykinin and the antagonist effects of the compounds were performed according to the methods reported in ref 16.

Acknowledgment. We are grateful to Mr. Ricci and Mr. Fabbri for technical assistance, to Dr. A. Cartoni, Dr. A. Triolo, and Dr. S. Mauro for analytical assistance, and to Dr. A. Ettorre and Dr. G. Balacco for the NMR spectra. This work was supported by grants from the Italian Ministry of University and Research (RIF.506/DSPAR 98).

References

(1) Mak, J. C. W.; Barnes, P. J. Autoradiographic visualization of bradykinin receptors in human and guinea-pig lung. *Eur. J. Pharmacol.* **1991**, *194*, 37−43.

(2) Dear, J. W.; Wirth, K.; Scadding, G. K.; Foreman, J. C. Characterization of the bradykinin receptor in the human nasal airway using the binding of [^{125}I]-Hoe 140. *Br. J. Pharmacol.* **1996**, *119*, 1054−1062.

(3) Polosa, R.; Holgate, S. T. Comparative airway response to inhaled bradykinin, kallidin, and [des-Arg^9]bradykinin in normal and asthmatic subjects. *Eur. J. Pharmacol.* **1989**, *163*, 195−197.

(4) Churchill, L.; Pongracic, J. A.; Reynolds, C. J.; Naclerio, R. M.; Proud, D. Pharmacology of nasal provocation with bradykinin: Studies of tachyphylaxis, cyclooxygenase inhibition, α-adrenergic stimulation and receptor subtype. *Int. Arch. Allergy Appl. Immunol.* **1991**, *95*, 322−331.

(5) Shibayama, Y; Skoner, D.; Suehiro, S.; Konishi, J. E.; Fireman, P.; Kaplan, A. P. Bradykinin levels during experimental nasal infection with rhinovirus and attenuated influenza virus. *Immunopharmacology* **1996**, *33*, 311−313.

(6) Svensson, C.; Andresson, M.; Persson, C. G.; Venge, P.; Alkner, U.; Pipkorn, U. Albumin, bradykinins, and eosinophil cationic protein on the nasal mucosal surface in patients with hay fever during natural allergen exposure. *J. Allergy Clin. Immunol.* **1990**, *85*, 828−833.

(7) Akbary, A. M.; Wirth, K. J.; Scholkens, B. A. Efficacy and tolerability of Icatibant (Hoe 140) in patients with moderately severe chronic bronchial asthma. *Immunopharmacology* **1996**, *33*, 238−242.

(8) Austin, C. E.; Foreman, J. C.; Scadding, G. K. Reduction by Hoe 140, the B$_2$ kinin receptor antagonist, of antigen-induced nasal blockage. *Br. J. Pharmacol.* **1994**, *111*, 969−971.

(9) Kuoppala, A.; Lindsted, K. A.; Saarinen, J.; Kovanen, P. T.; Kokkonen, J. O. Inactivation of bradykinin by angiotensin-converting enzyme and by carboxypeptidase N in human plasma. *Am. J. Physiol.* **2000**, *278* (4), H1069−1074.

(10) Campbell, D. J.; Krum, H.; Esler, M. D. Losartan increases bradykinin levels in hypertensive humans. *Circulation* **2005**, *111*, 315−320.

(11) Fattori, D.; Rossi, C.; Fincham, C. I.; Berettoni, M.; Calvani, F.; Catrambone, F.; Felicetti, P.; Gensini, M.; Terracciano, R.; Altamura, M.; Bressan, A.; Giuliani, S.; Maggi, C. A.; Meini, S.; Valenti, C.; Quartara, L. Design and synthesis of novel sulfonamide-containing bradykinin hB$_2$ receptor antagonists. 1. Synthesis and SAR of α,α-dimethylglycine sulfonamides. *J. Med. Chem.* **2006**, *49*, 3602−3613.

(12) Sum of residual BK-induced response calculated in percentage with respect to the basal response and normalized in comparison to the control registered at the various times of observation (5−30−60−90−120−180−210 min), further calculated in percentages with respect to the theoretical maximum. As a consequence, the maximal inhibition that may be obtained corresponds to $\Sigma\% = 0$, while the absence of inhibition corresponds to $\Sigma\% = 100$.

(13) Cucchi, P.; Meini, S.; Bressan, A.; Catalani, C.; Bellucci, F.; Santicioli, P.; Lecci, A.; Faiella, A.; Rotondaro, L.; Giuliani, S.; Giolitti, A.; Quartara, L.; Maggi, C. A. MEN16132, a novel potent and selective nonpeptide antagonist for the human bradykinin B2 receptor. In vitro pharmacology and molecular characterization. *Eur. J. Pharmacol.* **2005**, *528*, 7−16.

(14) (a) McAllistar, S. M.; Alpar, H. O.; Teitelbaum, Z.; Bennet, D. B. Do interactions with phospholipids contribute to the prolonged retention of polypeptides within lungs? *Adv. Drug Delivery Rev.* **1996**, *19*, 89−110. (b) Upton, R. N.; Doolette, D. J. Kinetic aspects of drug disposition in the lungs. *Clin. Exp. Pharmacol. Physiol.* **1999**, *26*, 381−391.

(15) Barnes, P. G. Tiotropium Bromide. *Exp. Opin. Inv. Drugs* **2001**, *10*, 733−740.

(16) Valenti, C.; Cialdai, C.; Giuliani, S.; Lecci, A.; Tramontana, M.; Meini, S.; Quartara, L.; Maggi, C. A. MEN 16132, a novel, potent and selective non-peptide kinin B$_2$ receptor antagonist. In vivo activity on bradykinin-induced bronchoconstriction and nasal mucosa microvascular leakage in anesthetized guinea pigs. *J. Pharmacol. Exp. Ther.* **2005**, 616−623.

JM061143K