Notes

Synthesis, *in Vitro* Binding Profile, and Autoradiographic Analysis of [^{3}H]-*cis*-3-[(2-Methoxybenzyl)amino]-2-phenylpiperidine, a Highly Potent and Selective Nonpeptide Substance P Receptor Antagonist Radioligand

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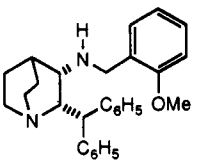
The synthesis of a highly potent and selective NK_1 receptor antagonist radioligand, [^{3}H]-*cis*-3-[(2-methoxybenzyl)amino]-2-phenylpiperidine (**6a**) is described. The *in vitro* binding pharmacology and autoradiographic distribution of **6a** in guinea pig brain following peripheral administration are also reported.

Introduction

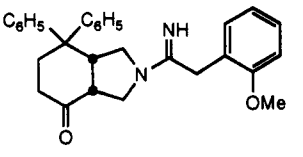
Substance P (SP, 1) is an undecapeptide first isolated by von Euler and Gaddum in 1931.[1] Its structure was determined by Chang and Leeman in 1970.[2] This neurotransmitter is a member of a class of structurally related peptides collectively referred to as tachykinins. Three tachykinin receptors have been identified and, on the basis of the corresponding affinities of various agonists, designated as NK_1, NK_2, and NK_3.[3] SP may be a mediator in the pathogenesis of several diverse disorders including asthma, arthritis, and schizophrenia.[4] However, a full understanding of the pharmacology of SP and its relationship to these and other disorders has been hampered by a lack of potent and selective antagonists for the receptor subtypes. Furthermore, the peptide nature of early antagonists generally renders them poor tools for *in vivo* studies due to their propensity to undergo proteolytic degradation. Recently, the first highly potent and selective nonpeptide antagonist for the NK_1 receptor, CP-96,345 (**2**), was described.[5] Subsequently, a structurally distinct antagonist, RP 67580 (**3**), was reported.[6] A potent and selective dipeptide NK_1 receptor antagonist, FK888 (**4**), has also been recently described.[7] SAR studies based upon the structure of CP-96,345 led to the discovery of (+)-(2*S*,3*S*)-3-[(2-methoxybenzyl)amino]-2-phenylpiperidine, CP-99,994 (**5**), a novel, highly potent, and selective NK_1 receptor antagonist.[8] While CP-96,345 has been shown to have significant affinity for the verapamil-sensitive calcium binding site,[9] compound **5** lacks this activity.[8b] In this paper, we describe the synthesis of a tritiated racemic derivative of **5** (compound **6a**) and its *in vitro* binding profile and autoradiographic distribution studies in guinea pig brain.

Arg-Pro-Lys-Pro-Gln-Gln-Phe-Phe-Gly-Leu-Met-NH_2

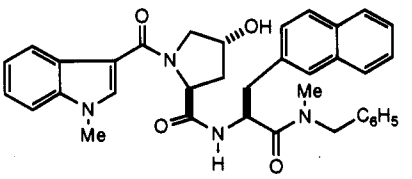
Substance P (**1**)



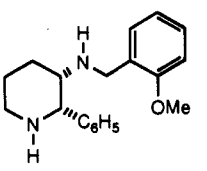
CP-96,345 (**2**)



RP 67580 (**3**)



FK888 (**4**)



CP-99,994 (**5**)

Chemistry

The synthesis of radioligand **6a** is summarized in Scheme I. All structures in this scheme are racemic. Metalation of 1,3,5-tribromobenzene followed by quenching with *N*,*N*-dimethylformamide (DMF), according to the known procedure,[10] provides 3,5-dibromobenzaldehyde. Treatment of this aldehyde with methyl 4-nitrobutyrate in the presence of ammonium acetate affords *trans*-disubstituted piperidine **7**.[11] Conversion of the *trans*-nitro substituent to a *cis*-(*o*-methoxybenzyl)amino group is accomplished using a four-step sequence analogous to that employed for the synthesis of **6b**.[12] Oxidative cleavage of the nitronate anion derived from **7** and oxime formation, without purification of the intermediate ketone, furnishes **8**. Oxime reduction proceeds stereoselectively with hydrogen delivery from the less hindered face of the molecule to give, after reductive amination of *o*-methoxybenzaldehyde, *cis*-substituted piperidinone **9**. No cleavage of the aromatic bromine substituents was observed. Borane reduction of the lactam function provides tritiation precursor **10**. Model studies on the hydrogenolysis of **10** indicated that exposure to hydrogen at atmospheric pressure in the presence of 20 mass % of 10% Pd/C proceeds cleanly to afford **6b**. The bromine atoms are cleaved selectively without loss of the methoxybenzyl moiety. Similar

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Scheme I

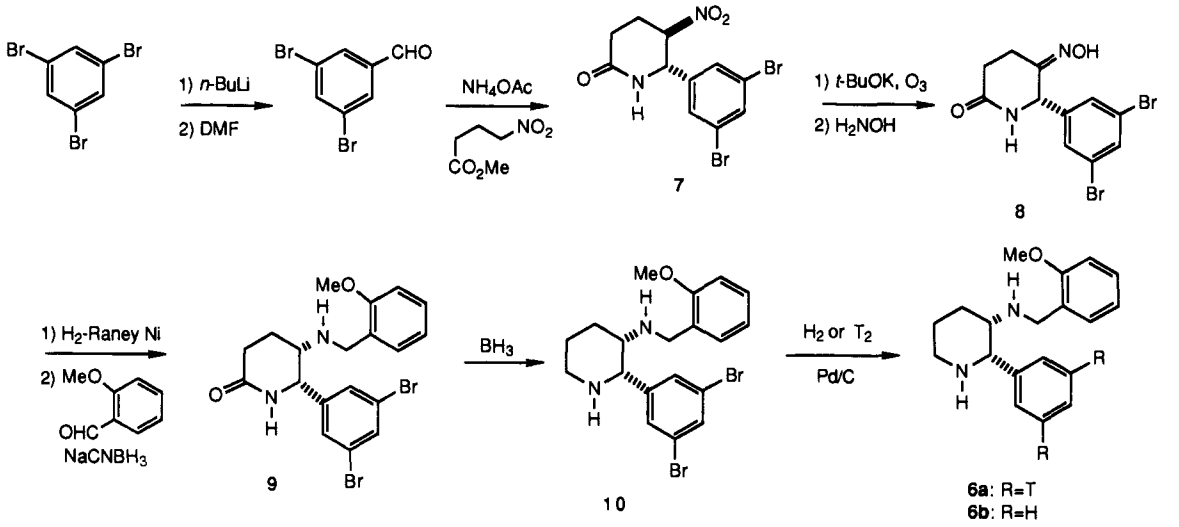


Table I. Pharmacological Characterization of the Binding of Compound **6a** to Guinea Pig Striatal Membranes

compound	N^a	K_i (nM) or % inhibition at highest dose tested
5	2	0.17 ± 0.045
6b	2	0.27 ± 0.055
11	1	50% at 10 μM
substance P	3	50.5 ± 1.4
Sar^9-Met(O_2)-SP (NK_1 receptor agonist)	3	20.0 ± 8.9
Neurokinin A (NKA, NK_2 receptor agonist)	1	3.7 μM
Eledoisin (NK_3 receptor agonist)	3	15% at 1 μM

[a] Number of determinations.

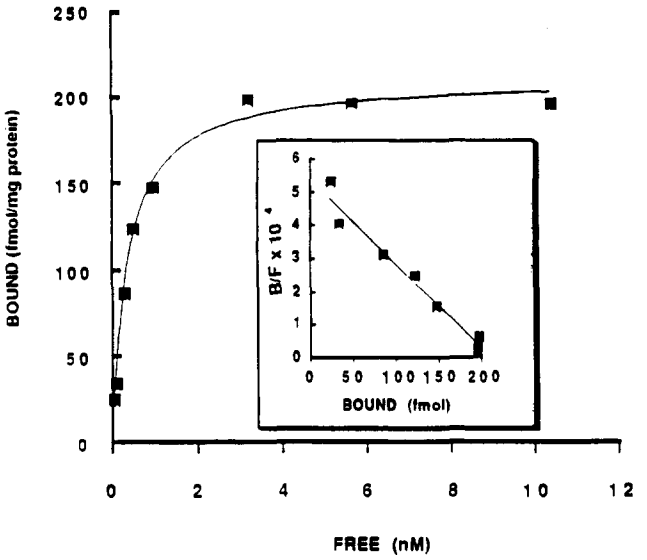
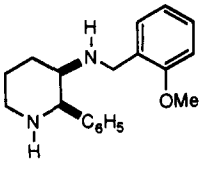


Figure 1. Saturation curve of the binding of compound **6a** to guinea pig brain.

conditions were employed in the tritiation procedure to obtain **6a** (specific activity 23.6 Ci/mmol, radiopurity 98.5%).

Characterization of the Binding of Compound 6a. Incubation of a fixed amount of neuronal membranes prepared from guinea pig caudate with increasing concentrations of **6a** (0.01–10 nM) revealed high affinity binding, K_d = 0.32 ± 0.06 nM, that was saturable and to a single site as reflected by the linear Scatchard plot (Figure 1). To confirm that the compound was labeling the NK_1 receptor, competition studies against **6a** (1–2 nM) were carried out using the NK_1 receptor agonists substance P and Sar^9-Met(O_2)-substance P; the NK_2 receptor agonist, neurokinin A (NKA); and the NK_3 receptor agonist, eledoisin (Table I). The rank order of K_i values obtained (Table I), SP ≥ Sar^9-Met(O_2)-SP ≫ NKA = eledoisin, is consistent with the conclusion that **6a** selectively binds to NK_1 receptors in guinea pig brain. As discussed previously,[5b] the absolute K_i values for SP and Sar^9-Met(O_2)-SP are greater than observed when binding is carried out *versus* an agonist radioligand. Drugs with high affinity for the noradrenergic transporter (desipramine), muscarinic (carbachol), benzodiazepine (clonazepam), dopamine (haloperidol), opiate (naloxone), or α-adrenergic (prazosin) receptors were without effect on the binding of **6a**. Thus, **6a** binds with high affinity to a single site consistent with labeling the NK_1 receptor. The enantiomer of compound **5**, compound **11**,[8a] exhibits significantly reduced NK_1 receptor affinity.



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Autoradiographic Distribution of 6a. Receptor autoradiography is a method for visualizing and measuring patterns of receptor distribution which provides information on the sites of action of a drug or neurotransmitter. Using receptor autoradiography, the distribution of binding sites labeled by **6a** in the guinea pig was mapped and compared qualitatively to the distribution of [^{3}H]SP binding sites in serial sections cut from the same brains. The two binding maps were found to be very similar (Figure 2a,b) and to agree in large part with the distribution of

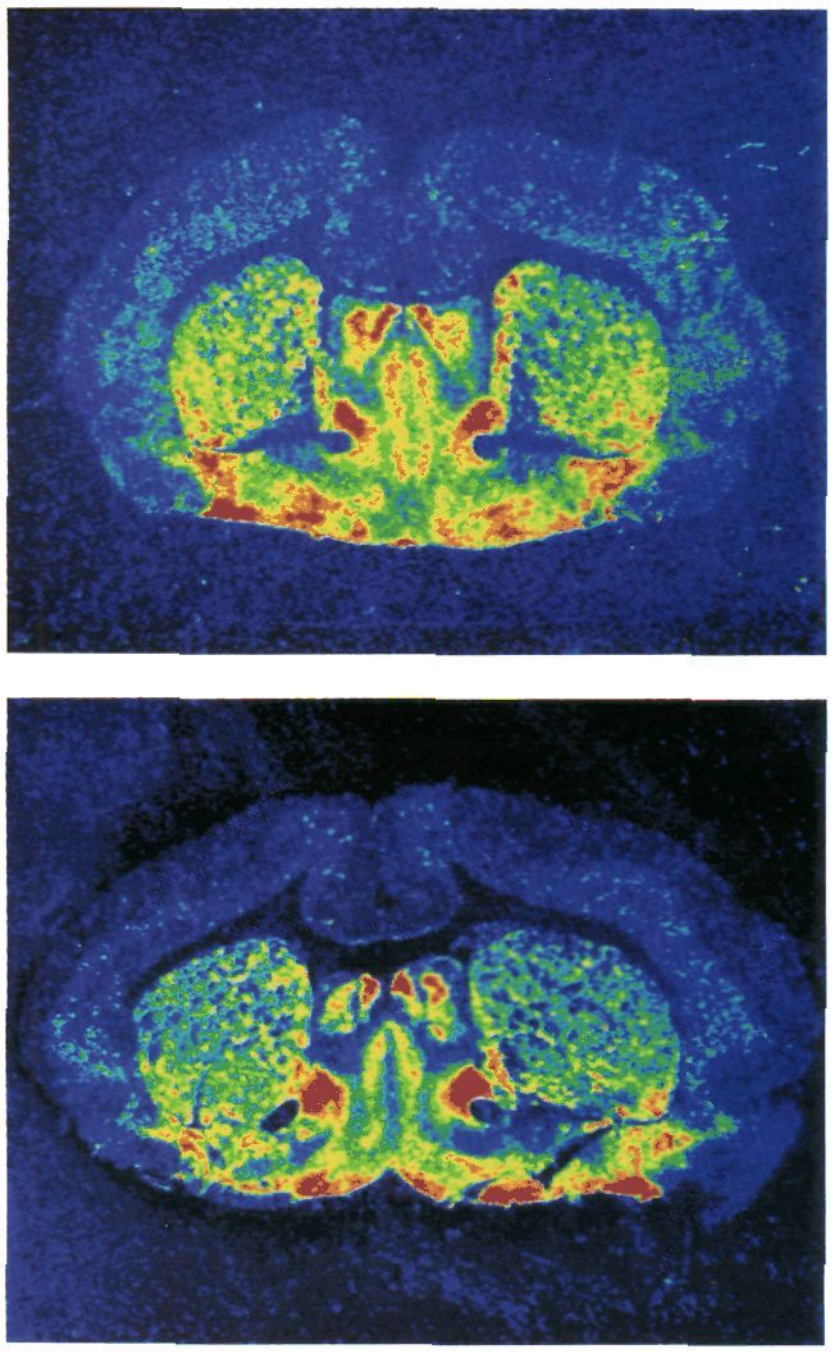

Figure 2. Distribution of the binding of (a) compound **6a** and (b) [^{3}H]SP in serial sections from the same guinea pig brain. (c) Displacement of **6a** with unlabeled SP. The density sequence follows the natural spectrum, i.e., red corresponds to regions of highest binding and blue corresponds to regions of low binding.

[^{3}H]SP binding sites reported previously for the guinea pig brain.[13] The specific binding component of **6a**, as defined by displacement with 5 μM **6b**, could be completely blocked by SP (5 μM), but not by the NK_3-selective tachykinin, eledoisin (5 μM).

Very dense specific binding (Table II) was found in discrete brain areas, including the amygdalohippocampal area, the diagonal band, anterior amygdala, medial habenula, septofimbrial nucleus, subiculum, and several nuclei surrounding the third ventricle. Moderate binding levels were found in several larger structures, including the striatum, nucleus accumbens, bed nucleus of the stria terminalis, superior colliculus, and the central gray, while specific binding was largely absent from the cerebral cortex, hippocampus, thalamus, and cerebellum. Overall, binding was most prominent in nuclei comprising the limbic system and in structures adjacent to the cerebral ventricles.

Table II. Autoradiographic Distribution of **6a** at Representative Sites in Guinea Pig Brain

structure	density (fmol/mg protein)
Forebrain	
nucleus accumbens	360 ± 25
lateral septal nucleus	76 ± 8
medial caudate	217 ± 4
globus pallidus	92 ± 6
anterior amygdala	614 ± 51
Midbrain	
dorsal hippocampus	65 ± 4
substantia nigra (reticulata)	85 ± 5
Hindbrain	
superior colliculus (zonal layer)	344 ± 15
central gray	184 ± 7

Summary of Results

Compound **5** is a highly potent antagonist at the NK_1 receptor. An efficient protocol for synthesis of a corresponding racemic tritium-labeled radioligand (**6a**) has been developed. The *in vitro* binding profile of **6a** in guinea pig striatum indicates that this radiolabel binds with high affinity and selectivity to NK_1 receptors. Autoradiographic studies show that it binds in brain with a distribution analogous to that of [^{3}H]SP. As such, compound **6a** is likely to be a useful tool in further defining biochemical, distribution and metabolic properties of **5** and other related compounds.

Experimental Section

General Methods. Unless otherwise noted, materials were obtained from commercial suppliers and used without further purification. Melting points are uncorrected. ^{1}H NMR spectra were determined on a Varian XL-300 spectrometer operating at 299.9 MHz. Significant ^{1}H NMR data are tabulated in the following order: multiplicity (s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet), number of protons, coupling constant(s) in hertz. Mass spectra were obtained with a Kratos Profile (EI Resolution 3500; source 200 °C; DIP) mass spectrometer. Column chromatography was done with J. T. Baker silica gel for flash column chromatography (40-μm average particle diameter) or Silica Woelm 32–63 (particle size 32–63 μm). Elemental analyses were performed by the Microanalytical Laboratory, operated by the Analytical Department, Pfizer Central Research, Groton, CT.

***trans*-6-(3,5-Dibromophenyl)-5-nitropiperidin-2-one (7).**[11] Under a nitrogen atmosphere in a round-bottom flask equipped with a reflux condenser were placed 19.4 g (74 mmol) of 3,5-dibromobenzaldehyde and 150 mL of ethanol. To the system were added 11.3 mL (74 mmol) of methyl 4-nitrobutyrate and 11.5 g (148 mmol) of ammonium acetate, and the reaction mixture was stirred at 80 °C for 6 h. The mixture was allowed to stand at room temperature overnight and was concentrated on a rotary evaporator. The residual oil was triturated with a small amount of ethanol and ether. Scratching and cooling resulted in crystal formation, and the brown solid (10.5 g) was collected by suction filtration. The solid was dissolved in dichloromethane, and the solution was washed with two portions of saturated aqueous sodium bicarbonate, dried (Na_2SO_4), and concentrated to obtain 8.08 g of **7** as a tan solid. The initial ether/ethanol filtrate was concentrated, and the residue was partitioned between dichlo-

romethane and saturated aqueous sodium bicarbonate. The mixture was washed with two portions of saturated aqueous sodium bicarbonate, dried (Na_2SO_4), and concentrated. Trituration (ethanol/ether) and filtration afforded an additional 3.9 g of **7**. The two isolates were combined (total yield 43%) and used without further purification in the subsequent transformation: ^{1}H NMR ($CDCl_3$) δ 2.25 (m, 1H), 2.52 (m, 3H), 4.62 (m, 1H), 5.18 (d, 1H, J = 6), 6.41 (br s, 1H) 7.35 (d, 2H, J = 2), 7.62 (t, 1H, J = 2); HRMS calcd for $C_{11}H_{10}Br_2N_2O_3$ 375.9058, found 375.9075.

***cis*-6-(3,5-Dibromophenyl)-5-[(2-methoxybenzyl)amino]-piperidin-2-one (9).** In a round-bottom flask were placed 10.0 g (26 mmol) of **7** and 50 mL of dichloromethane. To this stirring suspension was added 2.97 g (112 mmol) of potassium *tert*-butoxide, and the mixture was stirred for 15 min. To the system was added 50 mL of methanol. The mixture was cooled to −78 °C, and ozone was bubbled through the solution for 3 h. N_2 was bubbled through the solution for 10 min, 3 mL of dimethyl sulfide was added, and the N_2 bubbling was continued overnight. To the system were added water and dichloromethane, and 6.35 g of 6-(3,5-dibromophenyl)-5-oxopiperidin-2-one was isolated by filtration as a white solid: mp 222–223 °C; ^{1}H NMR (DMSO-d_6) δ 2.58 (m, 2H), 2.70 (m, 2H), 5.06 (s, 1H), 7.51 (br s, 2H), 8.21 (br s, 1H); HRMS calcd for $C_{11}H_9Br_2NO_2$ 344.9000, found 344.8911.

In a round-bottom flask was dissolved the ketone obtained above (6.3 g) in 75 mL of ethanol, and a solution prepared from 3.61 g (52 mmol) of hydroxylamine hydrochloride, 7.10 g (87 mmol) of sodium acetate, and 25 mL of water was added. The reaction mixture was stirred at room temperature for 3 days and partially concentrated, and the mixture was filtered to afford, after rinsing with ether, 6.03 g (64% yield from **7**) of oxime **8** as a white solid: mp 209–210 °C; ^{1}H NMR (DMSO-d_6) δ 2.30 (m, 2H), 2.62 (m, 1H), 3.44 (m, 1H), 5.14, 5.68 (2 br s, 1H), 7.46, 7.56 (2 br s, 2H), 8.10, 8.39 (2 br s, 1H); HRMS calcd for $C_{11}H_{10}Br_2N_2O_2$ 359.9109, found 359.9100.

Raney nickel (~12 g) was washed with H_2O until the pH was neutral and then washed with three portions of ethanol. In a bottle were placed 6.0 g of oxime **8** and ethanol, and the nickel catalyst (slurried in *ca.* 200 mL of ethanol) was added. The mixture was shaken under a hydrogen atmosphere on a Parr apparatus (*ca.* 40 psi) overnight. Additional washed Raney nickel (~6 g) was added to the system, and the mixture was shaken under hydrogen for 5 h. The crude reaction mixture was filtered through a pad of diatomaceous earth, and the filtrate was concentrated with a rotary evaporator to obtain 2.56 g of 5-amino-6-(3,5-dibromophenyl)piperidin-2-one as an oil. The product may be solidified by trituration with dichloromethane/ether: mp 202–209 °C; HRMS calcd for $C_{11}H_{12}Br_2N_2O$ 345.9136, found 345.9291.

Under a nitrogen atmosphere, in a round-bottom flask were placed the amine obtained above (2.56 g, 7.4 mmol), 20 mL of methanol, and *ca.* 4 g of 3-Å molecular sieves. The pH of the reaction mixture was adjusted to *ca.* 5 using methanol saturated with HCl, and 477 mg (7.4 mmol) of sodium cyanoborohydride was added to the system. The pH of the reaction mixture was adjusted to 5, and 1.21 g (8.9 mmol) of 2-methoxybenzaldehyde was added. The mixture was stirred at room temperature overnight and filtered through a pad of diatomaceous earth. The filtrate was concentrated, and the residue was partitioned between saturated aqueous sodium bicarbonate and chloroform. The layers were separated, the aqueous phase was washed with two portions of chloroform, and the combined organic fractions were dried (Na_2SO_4) and concentrated to afford 3.2 g of a yellow oil. The crude product was purified by flash column chromatography using 3:97 methanol/chloroform as the eluant to obtain 670 mg (9% yield from **8**) of **9**: mp 139–140 °C; ^{1}H NMR ($CDCl_3$) δ 1.70 (m, 1H), 1.86 (m, 1H), 2.26 (m, 1H), 2.55 (m, 1H), 2.92 (m, 1H), 3.46 (d, 1H, J = 12), 3.55 (s, 3H), 3.70 (d, 1H, J = 12), 4.54 (d, 1H, J = 3), 6.68 (d, 1H, J = 6), 6.78 (m, 1H), 6.98 (d, 1H, J = 6), 7.13 (t, 1H, J = 6), 7.23 (br s, 2H), 7.52 (br s, 1H); HRMS calcd for $C_{19}H_{20}Br_2N_2O_2$ 465.9892, found 465.9865. Anal. ($C_{19}H_{20}Br_2N_2O_2 \cdot {}^1/_4H_2O$) C, H, N.

***cis*-2-(3,5-Dibromophenyl)-3-[(2-methoxybenzyl)amino]-piperidine (10).** Under a nitrogen atmosphere, in a round-bottom flask equipped with a reflux condenser were placed 670 mg (1.4 mmol) of lactam **9** and 40 mL of THF. To this stirring solution was cautiously added 3.6 mL (7.2 mmol) of 2 M borane dimethyl sulfide in THF, and the reaction mixture was heated at 60 °C overnight. The mixture was cooled to room temperature, and 2 mL of methanol was slowly and cautiously added to the system. The mixture was stirred for 1.5 h and concentrated. To the system was added 386 mg (2.8 mmol) of potassium carbonate and 20 mL of ethanol, and the mixture was heated at reflux for 2.5 h and concentrated. The residue was partitioned between dichloromethane and water, the layers were separated, the aqueous phase was extracted with two portions of dichloromethane, and the combined organic fractions were dried (Na_2SO_4) and concentrated to afford 670 mg of crude material. This material was purified by flash column chromatography (30 g of silica gel) using 1:19 methanol/chloroform as the eluant to obtain 240 mg (38% yield) of pure **10** (mp 53–56 °C) and an additional 47 mg (7% yield) of slightly impure material: ^{1}H NMR ($CDCl_3$) δ 1.46 (m, 1H), 1.54 (m, 1H), 1.82 (m, 7H), 2.07 (m, 1H), 2.70 (m, 2H), 3.20 (m, 1H), 3.34 (d, 1H, J = 12), 3.54 (s, 3H), 3.68 (d, 1H, J = 12), 3.74 (s, 1H), 6.49 (d, 1H, J = 6), 6.79 (t, 1H, J = 6), 6.94 (d, 1H, J = 6), 7.14 (t, 1H, J = 6), 7.31 (s, 2H), 7.48 (s, 1H); HRMS calcd for $C_{19}H_{22}Br_2N_2O$ 454.0078, found 454.0143. Anal. ($C_{19}H_{22}Br_2N_2O$) C, H, N.

***cis*-3-[(2-Methoxybenzyl)amino]-2-phenylpiperidine (6b).** In a round-bottom flask were placed 10 mg of 10% Pd/C and 0.7 mL of THF, and the mixture was stirred under hydrogen (atmospheric hydrogenation apparatus) for 0.5 h. To the system were added 50 mg (0.11 mmol) of **10** and 0.061 mL (0.44 mmol) of triethylamine in 0.7 mL of THF (0.6 mL rinse), and the mixture was stirred under hydrogen for 22.5 h. The catalyst was removed by suction filtration through a pad of Celite, and the filter pad was rinsed well with methanol. The filtrate was concentrated, and the residue was partitioned between chloroform and saturated aqueous sodium bicarbonate. The layers were separated, and the aqueous phase was extracted with two portions of chloroform. The combined organic fractions were dried (Na_2SO_4) and concentrated to afford 25 mg (77% yield) of **6b** which had ^{1}H NMR spectral properties identical to those of material prepared previously.[8a,12]

Tritiation of Compound 10 and Purification of Compound 6a. A suspension of 10% Pd/C catalyst (10 mg) in THF (7 mL) was pretreated by stirring under 8 Ci of tritium gas for 1 h. The gas was removed, and a solution of **10** (50 mg) in THF (1.3 mL) and triethylamine (60 μL) were added to the system. The resulting mixture was stirred under 20 Ci of tritium gas for 16 h while maintaining the pressure at approximately 1 atm. The catalyst was removed by filtration through diatomaceous earth, and labile tritium was removed from the filtrate by codistillation with methanol *in vacuo*. Radio TLC analysis of the resulting crude product (1.98 Ci) indicated a purity of approximately 80%. A 265-mCi portion of the crude product was purified by preparative TLC on five 5- × 20-cm Whatman LK6F analytical TLC plates developed with $CHCl_3$/MeOH (9:1). The product band was removed and extracted with methanol. The resulting material was purified two additional times employing the same method using four plates each time to finally give 39.5 mCi of high purity material. The radiochemical purity was determined to be 98.5% by TLC [Whatman LK6F silica gel, $CHCl_3$/MeOH/HOAc (70:20:10), R_f = 0.52, predeveloped before spotting]. The specific activity was determined to be 23.6 Ci/mmol [spectrophotometric method using ϵ_{278} = 1.81 × 10^3 (95% ethanol)].

Binding Assay. Membranes were prepared from the striata of male Hartley guinea pigs. Binding studies were initiated by the addition of 25 μL of tissue (75 μg of protein) to buffer containing increasing concentrations of **6a** (0.01–6 nM). For competition studies, various concentrations of inhibitors were added to buffer containing 1.0 nM of **6a**. Following a 10-min incubation, the assay was terminated by rapid filtration over filter paper soaked in 0.01% polyethylenimine and then washed with 5 mL of ice-cold buffer. The filter disks were placed into 15- × 60-mm scintillation vials, and 4 mL of Ecolume was added. Radioactivity bound to the membranes was determined by counting the samples on a LKB Liquid Scintillation counter, 2–3 min per sample. Data were analyzed using the Lundon1 software program.

Autoradiography. Guinea pig brains (three animals) were frozen in isopentane (−20 °C), sectioned at 20-μm thickness, thaw

mounted onto gelatin coated slides, and stored at −65 °C. Slide-mounted sections were preincubated for 15 min at room temperature in 50 mM Tris HCl, 5 mM $MnCl_2$, and 0.02% BSA, adjusted to pH 7.3, followed by a 30-min incubation at 4 °C in buffer containing 2 nM **6a** or the same buffer but with the addition of chymostatin (2 μg/mL), bacitracin (40 μg/mL), leupeptin (4 μg/mL), and 2 nM [^{3}H]SP. Nonspecific binding was defined by the addition of 5 μM SP. The sections were washed in two successive washes in ice-cold buffer for 5 min each, dipped briefly in distilled water, and dried under a stream of air. All sections were placed in standard X-ray cassettes, along with tritium brain paste standards, apposed to LKB tritium-sensitive Ultrofilm for 1 month and developed by standard photographic procedures. Autoradiographic film density was quantitated using an Imaging Systems MCID image analysis system.

References

(1) von Euler, V. S.; Gaddum, J. H. An unidentified depressor substance in certain tissue extracts. *J. Physiol.* **1931**, *72*, 577–583.

(2) Chang, M.; Leeman, S. E. Isolation of a sialogogic peptide from bovine hypothalamic tissue and its characterization as substance P. *J. Biol. Chem.* **1970**, *245*, 4784–4790.

(3) Regoli, D.; Drapeau, G.; Dion, S.; Couture, R. New selective agonists for neurokinin receptors: pharmacological tools for receptor characterization. *Trends Pharmacol. Sci.* **1988**, *9*, 290–295.

(4) For a discussion of the physiological significance of the tachykinins, see: Helke, C. J.; Krause, J. E.; Mantyh, P. W.; Couture, R.; Bannon, M. J. Diversity in mammalian tachykinin peptidergic neurons: multiple peptides, receptors, and regulatory mechanisms. *FASEB* **1990**, *4*, 1606–1615 and references therein.

(5) (a) Snider, R. M.; Constantine, J. W.; Lowe, J. A.; Longo, K. P.; Lebel, W. S.; Woody, H. A.; Drozda, S. E.; Desai, M. C.; Vinick, F. J.; Spencer, R. W.; Hess, H.-J. A potent nonpeptide antagonist of the substance P (NK_1) receptor. *Science* **1991**, *251*, 435–437. (b) McLean, S.; Ganong, A.; Seeger, T.; Bryce, D.; Pratt, K.; Reynolds, L.; Siok, C.; Lowe, J.; Heym, J. Characterization of the activity and distribution of binding sites in brain of a nonpeptide substance P (NK_1) receptor antagonist. *Science* **1991**, *251*, 437–439.

(6) Garret, C.; Caruette, A.; Fardin, V.; Moussaoui, S.; Peyronel, J.-F.; Blanchard, J.-C.; Laudron, P. M. Pharmacological properties of a potent and selective nonpeptide substance P antagonist. *Proc. Natl. Acad. Sci. (U.S.A.)* **1991**, *88*, 10208–10212.

(7) Fujii, T.; Murai, M.; Morimoto, H.; Maeda, Y.; Yamaoka, M.; Hagiwara, D.; Miyake, H.; Ikari, N.; Matsuo, M. Pharmacological profile of a high affinity dipeptide NK_1 receptor antagonist, FK888. *Br. J. Pharmacol.* **1992**, *107*, 785–789.

(8) (a) Desai, M. C.; Lefkowitz, S. L.; Thadeio, P. F. Discovery of a potent substance P antagonist: recognition of the key molecular determinant. *J. Med. Chem.* **1992**, *35*, 4911. (b) McLean, S.; Ganong, A.; Seymour, P. A.; Snider, R. M.; Desai, M. C.; Rosen, T.; Bryce, D. K.; Longo, K. P.; Reynolds, L. S.; Robinson, G.; Schmidt, A. W.; Siok, C.; Heym, J. Pharmacology of CP-99,994: a nonpeptide antagonist of the tachykinin NK_1 receptor. *J. Pharm. Exp. Ther.*, in press.

(9) Schmidt, A. W.; McLean, S.; Heym, J. Substance P receptor antagonist, CP-96,345: interaction with calcium channels. *Eur. J. Pharmacol.* **1992**, *219*, 491–492.

(10) Chen, L. S.; Chen, G. J.; Tamborski, C. 1,3,5-Trisubstituted benzenes I. Synthesis and reactions of 3,5-dibromophenyllithium. *J. Organomet. Chem.* **1981**, *215*, 281–291.

(11) Bhagwatheeswaran, H.; Gaur, S. P.; Jain, P. A novel synthesis of substituted 2-oxopiperidines. *Synthesis* **1976**, 615–616.

(12) (a) Desai, M. C.; Rosen, T. J. WO 91/09844. (b) Desai, M. C.; Thadeio, P. F.; Lefkowitz, S. L. Synthesis of (±) - CP-99,994: A Highly Potent Substance P Antagonist. *Tetrahedron Lett.*, in press.

(13) Dam, T.-V.; Quirion, R. Pharmacological characterization and autoradiographic localization of substance P receptors in guinea pig brain. *Peptides* **1986**, *7*, 855–864.