Structure-activity Relationships for Development of Neurokinin-3 Receptor Antagonists with Reduced Environmental Impact

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Koki Yamamoto

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Koki Yamamoto

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Preface

Pharmaceuticals include one or multiple bioactive ingredients that cure and/or alleviate symptoms in human or veterinary diseases. Many of these ingredients are excreted from the body in an unmetabolized form and/or as active metabolites via urine and/or feces. The fate of each ingredient after excretion depends on its chemical properties as well as the methods used to treat water it is excreted into (Figure 1).[1] Even after wastewater treatment plant operations, bioactive ingredients and their metabolites often survive to become ground and surface water contaminants in downstream areas.[2]

The high chemical stability of the ingredients is crucial for the quality control of pharmaceuticals during long-term storage before administration. However, in terms of the impacts on the natural environment and ecosystems, stable bioactive substances with high bioactivity may have unfavorable effects on non-target species after excretion from patients or treated animals. Indeed, there have been cases of environmental contamination by bioactive ingredients derived from human and animal drugs. Additionally, exposure of pathogenic microorganisms to antibacterial and antiviral agents in residential and industrial effluent has led to the emergence of the drug-resistant strains.[3,4] For example, oxytetracycline-resistant bacteria were identified in wastewater streams including high levels of oxytetracycline.[5] In aquatic environments, a synthetic estrogen used for hormone therapy, 17α-ethynylestradiol (EE2), has been found to influence sex determination of aquatic fauna and flora and to accumulate in benthic invertebrates from the sediments.[6,7] Additionally, antidepressants often accumulate in organisms higher up in the food chain.[8] For example, venlafaxine was detected in aquatic invertebrates, as well as in the riparian spiders that preyed upon them.[9] Moreover, bioaccumulation of antidepressants affects animal behaviors such as predator avoidance.[10] To overcome these disadvantages of highly stable and potent bioactive substances, drug design to turn off bioactivity after

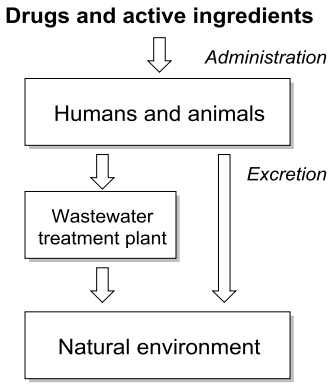


Figure 1. Fate of bioactive substances in pharmaceuticals.

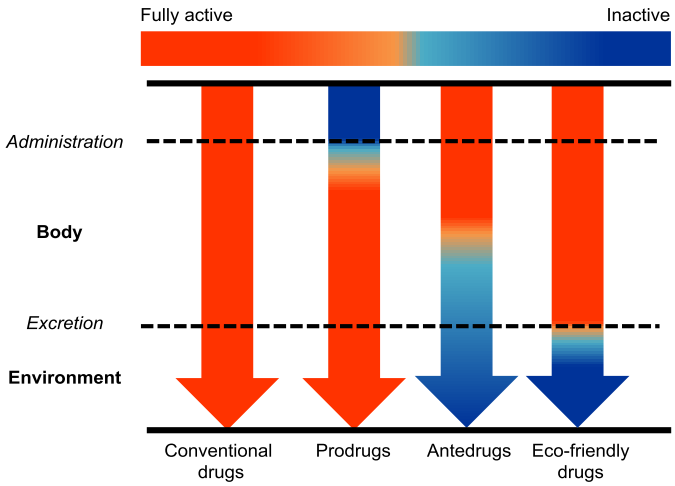
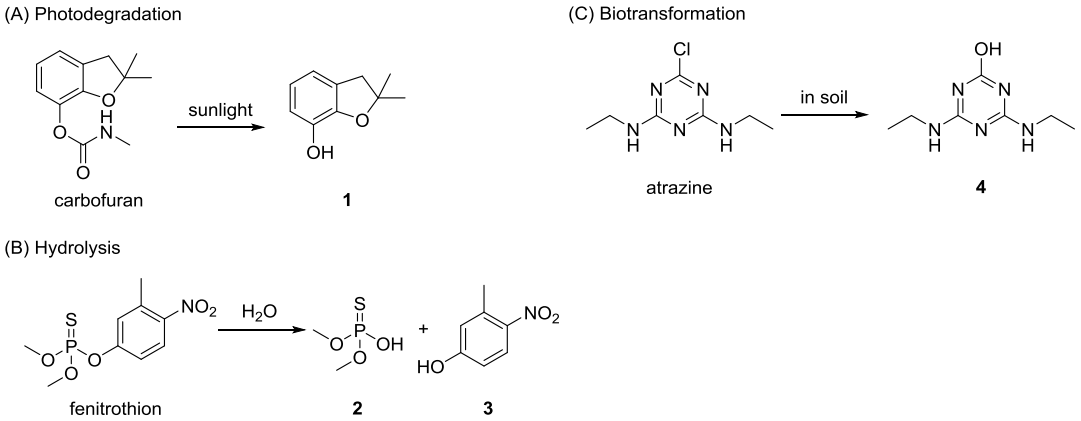


Figure 2. Biological activities of conventional drugs and eco-friendly drugs over time and space.

release into the environment is needed.

To date, several chemistry-based approaches have been developed to control the bioactivities and properties of pharmaceutical agents to ensure appropriate drug delivery and distribution in time and space (Figure 2).[11] Prodrugs are drugs that have been inactivated by various modifications that are converted into an active form by metabolism after administration. For example, Fortin et al. developed an antimicrotubule agent that is activated by CYP1A1 for tumor-selective bioactivity.[12] Moreover, several ester derivatives of ibuprofen have been designed to reduce gastrointestinal side effects.[13] Prodrugs are mainly designed to improve oral bioavailability, while decreasing the incidence of adverse effects during the administration and distribution processes.[14] The antedrug concept is an alternative approach to controlling bioactivity that is converted to an inactive form before excretion in order to avoid the possible toxicity during the excretion process. For example, budiodarone (antiarrhythmic) is administered as a bioactive form, which is subjected to gradual (bio)transformation into the inactive form by plasma and tissue esterases to reduce pulmonary toxicity.[15] Eco-friendly drugs are a similar concept, in which the bioactivity is offset after the active ingredient is released from the drug target. Whereas the antedrug bioactivities can be controled by metabolism process in the body, the eco-friendly drugs should be spontaneously degraded under the natural environmental conditions.

Previously, a number of pesticides caused serious adverse effects on the environment as well as animal bodies. One of the problems is that insecticides exhibit the toxicity to non-target species. For example, carbaryl (carbamate insecticide) and malathion (organophosphate insecticide) have negative effects on plant growth.[16] In addition, some



Scheme 1. Pesticides with the decomposition under the natural environmental conditions.

organophosphate insecticides remain in the environment to have adverse effects even on humans through the food chains. For example, methyl parathion detected in vegetables poses some risk to human health such as chromosomal aberrations and cardiovascular abnormalities.[17] To overcome these disadvantages, pesticides with decomposable property under the natural environmental conditions such as photodegradation, hydrolysis, and biotransformation by microorganisms have been developed. Some examples are described below. Carbofuran is an insecticide, which is converted into low toxic compound **1** by sunlight irradiation (Scheme 1A).[18] Hydrolysis of fenitrothion (insecticide) in the basic buffer leads to transformation into the compounds **2** and **3** (Scheme 1B).[19] Atrazine (herbicide) is degraded by microorganisms to give a nontoxic form **4** in the soil (Scheme 1C).[20]

Neurokinin-3 receptor (NK3R) is a class A GPCR that is preferentially activated by neurokinin B (NKB).[21] Early studies indicated that NKB–NK3R systems play a role in control of dopaminergic functions in the brain.[22] Therefore, NK3R antagonists were previously developed as antipsychotics for schizophrenia, which is caused by excessive dopaminergic function.[23] Recent studies have revealed that NKB–NK3R signaling plays an important role in controlling mammalian reproduction via regulation of the hypothalamic–pituitary–gonadal (HPG) axis.[24,25] NK3R with respect to reproductive function is expressed on the kisspeptin–NKB–dynorphin A neurons (KNDy neurons) located in the hypothalamic arcuate nucleus. NKB positively regulates the reproductive hormone cascade via activation of the gonadotropin-releasing hormone (GnRH) neuron in the hypothalamus, leading to the pulsatile secretion of luteinizing hormone (LH) from the pituitary gland (Figure 3).[25] Administration of senktide, an NK3R-selective agonist, stimulated LH secretion in sheep,[26] rats,[27] and monkey.[28] Through the structure–activity

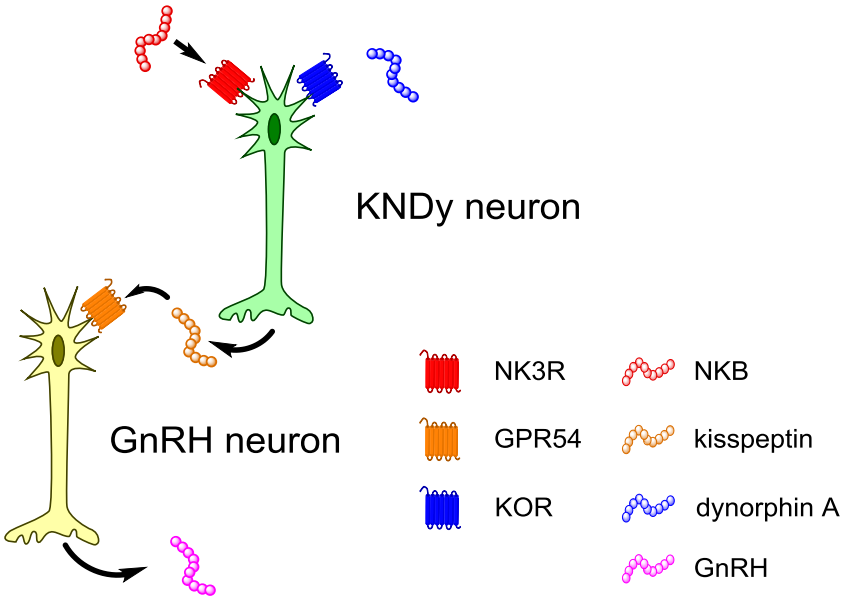


Figure 3. Regulation of GnRH secretion by NKB–NK3R signaling.

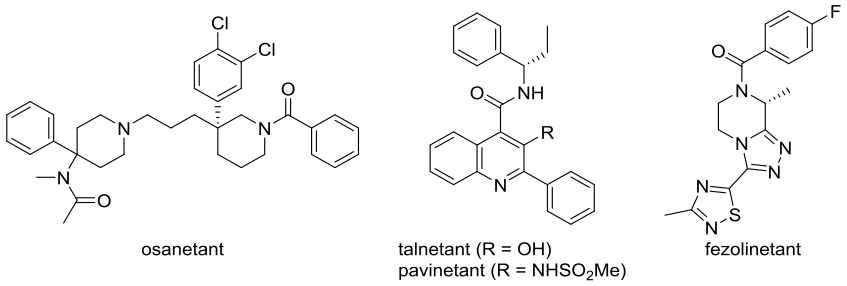


Figure 4. Structures of NK3R antagonists.

relationship studies of senktide, the author's group identified novel NK3R selective agonists with high potency and resistance to proteolytic degradation.[29,30]

A number of NK3R antagonists have been exploited over the last two decades. In early studies, osanetant[31] and talnetant[32] were investigated for the treatment of schizophrenia (Figure 4).[33] A recent clinical study revealed that oral administration of pavinetant, which is a talenetant derivative, effectively decreased testosterone levels of healthy volunteers.[34] Pavinetant is expected to be applicable to treatment of polycystic ovarian syndrome (PCOS).[35] Fezolinetant was recently identified as a novel NK3R antagonist with a unique triazolopiperazine scaffold.[36] Oral administration of fezolinetant in monkeys led to reduction of gonadotropin and ovarian hormone levels throughout the menstrual cycle.[37] A clinical investigation also revealed that fezolinetant was an effective inhibitor of the HPG axis to decrease gonadotropin secretion.[38] NK3R antagonists also have the potential for use as veterinary medicine to regulate the secretion of reproductive hormones in companion animals and zoo animals.[39] Because their reproductive functions

are only suppressed during the medication period, NK3R antagonists can be useful as antifertility agents and sedating agents. Additionally, NK3R antagonists can be used as anticancer agents for androgen-dependent prostate carcinoma via inhibition of GnRH secretion from the hypothalamus.[40] Thus, NK3R antagonists are expected to be therapeutic agents for various disorders of reproductive functions.

After excretion from treated humans and animals, NK3R antagonists and their bioactive metabolite(s) may affect the reproductive functions of non-target species via water pollution and/or soil contamination. To minimize the possibility of these adverse effects on non-target species, structures with potent NK3R antagonistic activity need to be converted into inactive form(s) by drug metabolizing systems in the body before excretion and/or by spontaneous degradation in the environment soon after excretion. Particularly in the case of oral drugs, significant proportion of the active ingredient(s) is excreted without being absorbed into the body. Because these ingredient(s) with less bioavailability cannot be metabolized by liver, kidney and/or other processes, the direct degradation under environmental conditions should be considered.

For the development of eco-friendly NK3R antagonists, the compounds should satisfy two requirements. First, the compounds should have potent NK3R inhibitory activity, which is comparable to that of the parent compound(s). Second, the compounds should be converted into the inactive form(s) by possible reaction(s) in the natural environment such as photoreaction, hydrolysis, biodegradation and/or air oxidation. Because available reactions for degradation under mild conditions in the natural environment are limited, it is challenging to design the expected active ingredient structures, which could be converted into the inactive form in appropriate time and space.

In this thesis, the author describes structure-activity relationship studies of NK3R antagonists with reduced environmental impact to enable development of potential pharmaceutical agents for treatment of reproductive disorders.

In Chapter 1, the author describes a structure-activity relationship study of talnetant derivatives having a labile functional group, which can assist their conversion into less active species in the natural environment.

In Chapter 2, Section 1, the author describes the development of facile synthetic protocols of fused piperazine scaffolds to investigate the structure-activity relationships of fezolinetant derivatives. The applications of the resulting oxadiazolopiperazines to scaffolds in bioactive compounds were also investigated.

In Chapter 2, Section 2, the author describes the scaffold hopping from fezolinetant using a series of fused piperidine heterocycles for identification of novel NK3R antagonists.

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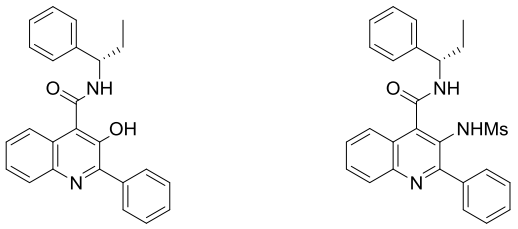
Chapter 1. Development of NK3R Antagonists with a Labile Functional Group in the Natural Environment

Summary

For development of novel NK3R antagonists with reduced environmental toxicity, a structure-activity relationship study of an NK3R antagonist, talnetant, was conducted. Among several talnetant derivatives with labile functional groups in the natural environment, a 3-mercaptoquinoline derivative exhibited comparable biological activity to that of the parent talnetant. Additionally, 3-mercaptoquinoline was converted into the disulfide or isothiazolone form by air-oxidation, both of which showed no binding affinity to NK3R.

Talnetant **1a** and pavinetant **1b** are quinoline-based NK3R antagonists (Figure 1). The quinoline core motif in talnetant was identified from structure-activity relationship studies of NK1R antagonists. Subsequently, improvement of the binding affinity and selectivity toward NK3R led to the development of talnetant as a second class non-peptide NK3R antagonist that was more potent and selective than a first class non-peptide NK3R antagonist, osanetant.[1] In early investigations, clinical studies of talnetant were conducted to investigate it as an agent for treatment of schizophrenia and irritable bowel syndrome.[2,3] Recently, it was revealed that NK3R antagonists could be used for the treatment of reproductive disorders by inhibiting the release of reproductive hormones.[4] To develop novel NK3R antagonists with reduced environmental impact, the author conducted a structure-activity relationship study of talenetant derivatives by introduction of functional group(s) that could be converted into inactive form(s) in the natural environment.

In the natural environment, chemical substances can be degraded by oxygen- and sunlight-mediated reactions. The author designed novel NK3R antagonists whose



talnetant (**1a**) pavinetatnt (**1b**)

Figure 1. Structures of NK3R antagonists with a quinoline scaffold.

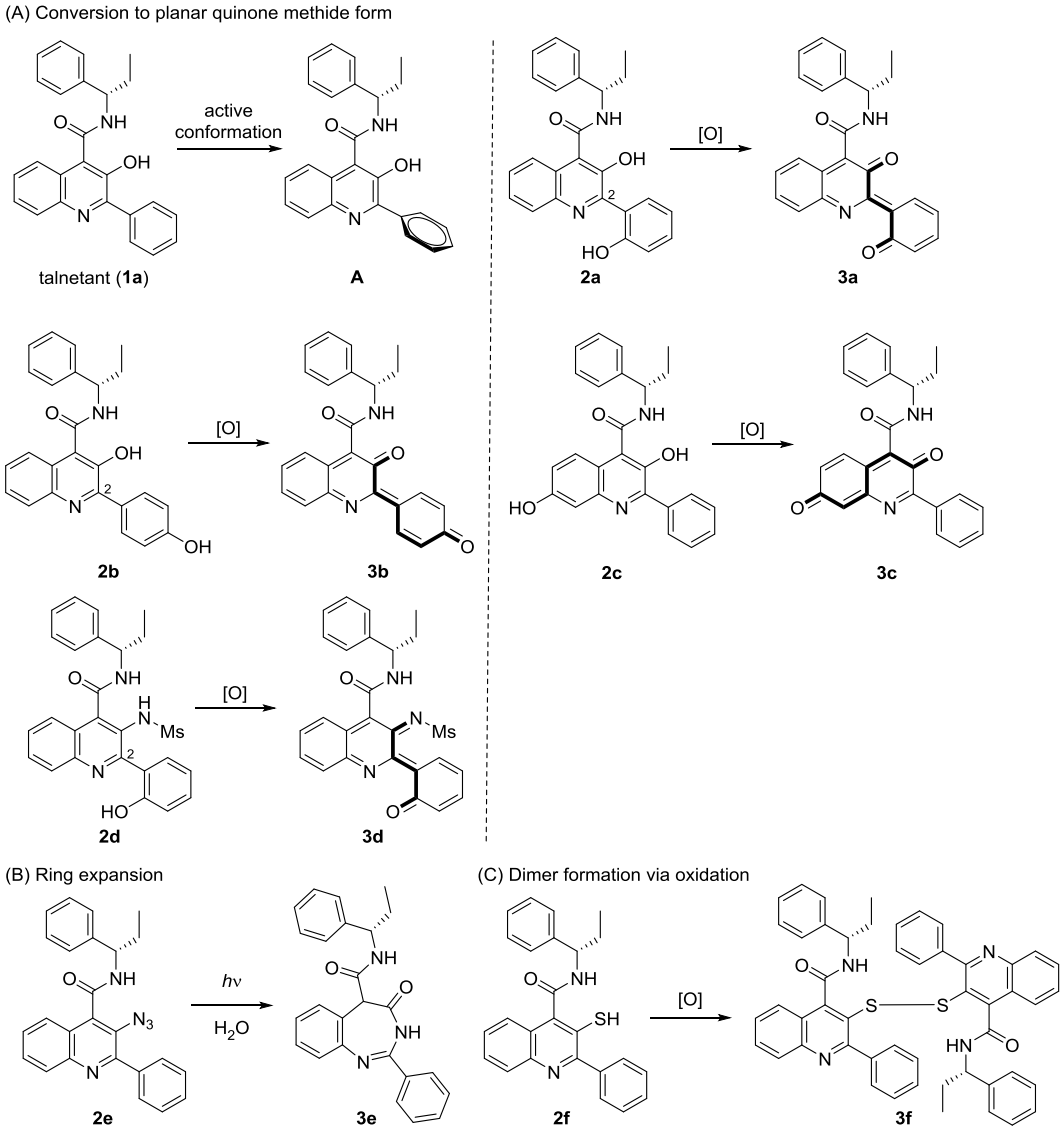
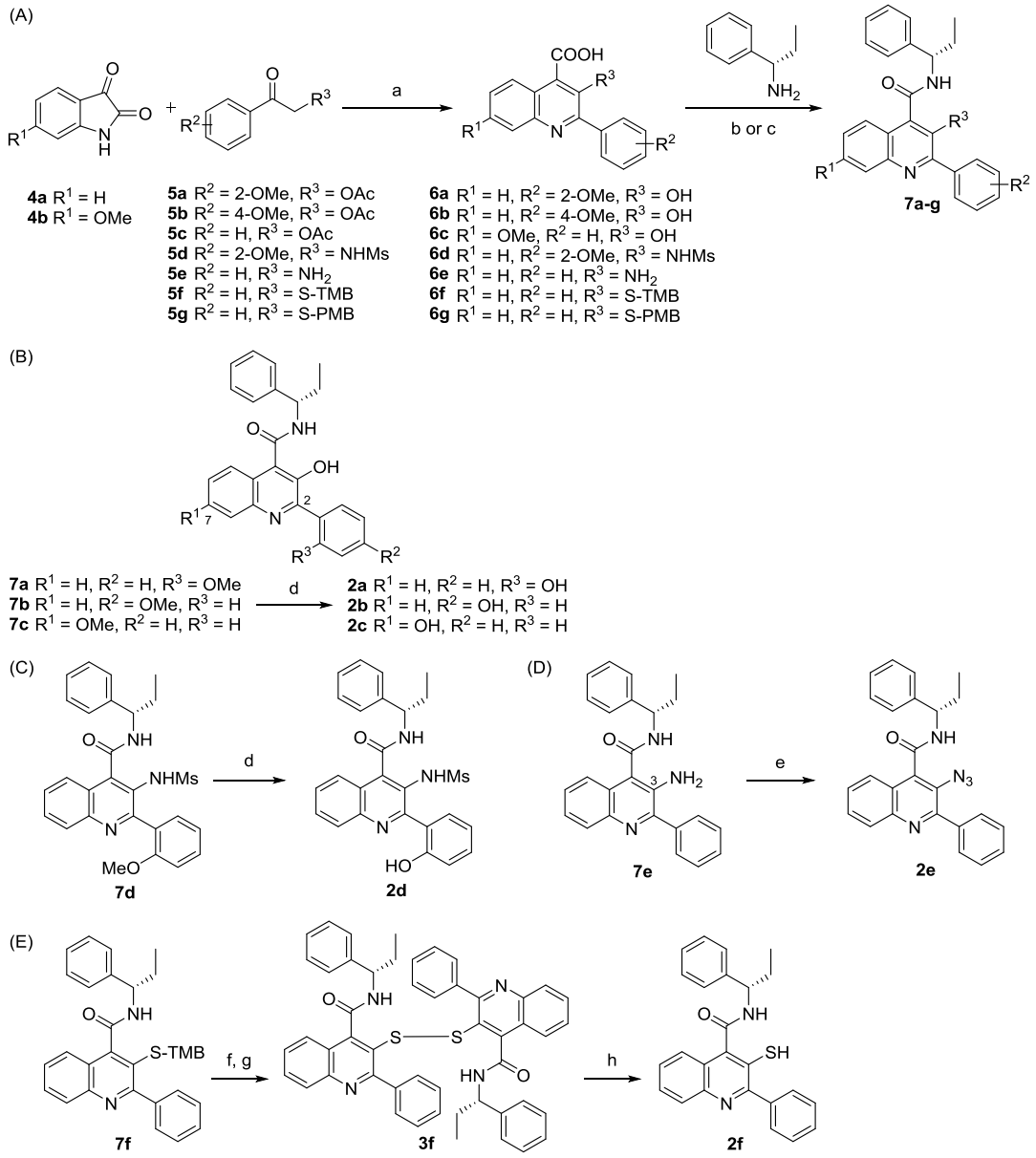


Figure 2. Design of NK3R antagonists with reduced environmental toxicity.

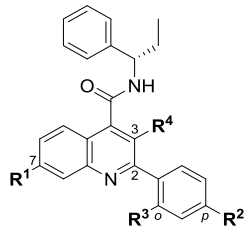
biological activities could be altered via three possible conversions including air-oxidation and photoreaction. Compound **2a** having an *o*-hydroxyphenyl group at the quinoline 2-position was designed to be converted under oxidative conditions to provide the quinone methide form **3a** (Figure 2A). It was expected that compound **3a** with a planar conformation would show decreased potency compared with the parent **2a** because talnetant (**1a**) binds to NK3R in the twisted biaryl conformation **A**.[5] Compounds **2b** and **2d** could also be converted into the similar planar quinone methide-type products under oxidative conditions. It was also expected that compound **2c** could be decomposed via the formation of a labile quinone methide form. For the second class of NK3R antagonists, 3-azidoquinoline **2e** was designed so that it could be subjected to



Scheme 1. Synthesis of talnetant derivatives **2a**–**f**. *Reagents and conditions*: (a) NaOH, EtOH, 85 °C; (b) CDI, TEA, MeCN, 50 °C; (c) HATU, DIPEA, DMF, rt; (d) BBr_3, CH_2Cl_2, 0 °C to rt; (e)$TMSN_3$, *t*-$BuNO_2$, MeCN, 0 °C to rt; (f) TFA, thioanisole, rt; (g) O_2, MeCN, rt; (h) L-cysteine, MeOH/H_2O, 0 °C to rt. *Abbreviations*: Ms: methanesulfonyl. TMB: 2,4,6-trimethoxybenzyl. PMB: 4-methoxybenzyl.

photoactivation to provide the ring-expanded benzodiazepine **3e** via nitrene formation. Because the quinoline ring is essential to the biological activity,[1a] ring expansion could lead to inactivation (Figure 2B). Compound **2f** with a 3-mercapto group was also designed to be dimerized to form a disulfide **3f** under oxidative conditions. The dimerization via

Table 1. Biological evaluation of talnetant derivatives.



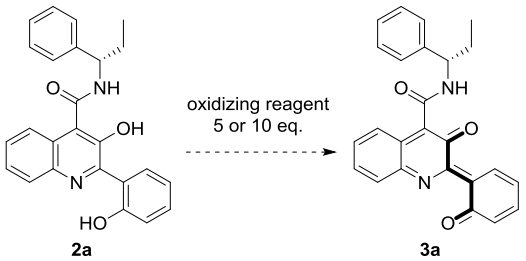
compound	**R^1**	**R^2**	**R^3**	**R^4**	IC$_{50}$ (nM)[a]
1a	H	H	H	OH	8.4 ± 2.0
1b	H	H	H	NH-Ms[b]	1.6 ± 0.7
7a	H	H	OMe	OH	1527 ± 234
7b	H	OMe	H	OH	965 ± 133
7c	OMe	H	H	OH	44 ± 8.0
2a	H	H	OH	OH	6.0 ± 2.0
2b	H	OH	H	OH	390 ± 68
2c	OH	H	H	OH	245 ± 45
2d	H	H	OH	NH-Ms[b]	3.9 ± 0.8
2e	H	H	H	N_3	24 ± 12
2f	H	H	H	SH	5.6 ± 1.8
7g	H	H	H	S-PMB[c]	154 ± 65

[a]IC$_{50}$ values are the concentrations required for 50% inhibition of the ([^{125}I]His^3, MePhe^7)-NKB binding to the NK3R (n = 6). [b]Ms: methanesulfonyl. [c]PMB: 4-methoxybenzyl.

air-oxidation could lead to a loss or decrease in the biological activity (Figure 2C).

The author synthesized the derivatives with the potential to be inactivated via the above-mentioned three pathways. The Pfitzinger reaction of isatins **4a** and **4b** with the appropriate aryl ketones **5a**–**g** provided quinoline-4-carboxylic acids **6a**–**g** in 20–74% yield. For the preparation of amides **7a**–**g**, carboxylic acids **6a**–**g** were coupled with (*S*)-1-phenylpropan-1-amine using carbonyldiimidazole (CDI) in acetonitrile or *O*-(7-azabenzotriazol-1-yl)-*N*,*N*,*N*′,*N*′-tetramethyluronium hexafluorophosphate (HATU) in DMF (Scheme 1A).[6] The talnetant derivatives **2a**–**d** with a hydroxy group were obtained by treatment of the methoxy precursors **7a**–**d** with BBr_3 (Schemes 1B and 1C). For the preparation of compound **2e** with an azide group at the quinoline 3-position, compound **7e** was treated with $TMSN_3$ and *t*-$BuNO_2$ under Sandmeyer conditions (Scheme 1D). The derivative **2f** with a 3-mercapto group was prepared from **7f** in three steps via deprotection of the trimethoxybenzyl group, formation of a temporary dimer by air-oxidation, and

Table 2. Conversion of diol **2a** under oxidative conditions.



2a **3a**

entry	oxidizing agent[a]	solvent	time	result
1	Air (K_2CO_3)	acetone	24 h	no reaction
2	Air (DBU)	MeOH	24 h	no reaction
3	H_2O_2/$NaHCO_3$	acetone	24 h	no reaction
4	$FeCl_3$	MeOH	24 h	no reaction
5	DDQ	MeOH	24 h	no reaction
6	MnO_2	acetone	24 h	no reaction
7	$KMnO_4$	acetone	24 h	no reaction
8	PCC	MeOH	5 min	decomp.
9	CAN	MeCN	5 min	decomp.
10	O_3	MeOH	30 min	decomp.
11	NBS	MeOH	3 h	decomp.
12	Ag_2O	acetone	3 h	decomp.

[a]DBU: 1,8-diazabicyclo[5.4.0]undec-7-ene; DDQ: 2,3-dichloro-5,6-dicyano-*p*-benzoquinone; CAN: ammonium cerium (IV) nitrate; PCC: pyridinium chlorochromate; NBS: *N*-bromosuccinimide.

subsequent reduction with L-cysteine (Scheme 1E). Of note is that the deprotection of the *p*-methoxybenzyl group in **7g** by treatment with TFA, TfOH, and thioanisole for the synthesis of **2f** was unsuccessful.

The biological activities of talnetant derivatives **2a**–**f** along with the synthetic intermediates **7a**–**c** and **7g** was evaluated by a competitive binding assay using ([^{125}I]-His^3, MePhe^7)-NKB for human NK3R.[7] The *o*-hydroxy modification at the 2-phenyl group on the quinoline (**2a** and **2d**) led to slightly more potent NK3R inhibition [IC_{50} (**2a**): 6.0 nM, IC_{50} (**2d**): 3.9 nM] compared with talnetant (IC_{50}: 8.4 nM). The 3-mercapto modification (**2f**) also inhibited the NKB binding to NK3R with high potency [IC_{50} (**2f**): 5.6 nM]. In contrast, the 7-methoxy, 7-hydroxy, 3-azido and 3-(*p*-methoxybenzyl)thio modifications of the quinoline ring (**7c**, **2c**, **2e**, and **7g**) decreased NK3R inhibition [IC_{50} (**7c**): 44 nM, IC_{50} (**2c**): 245 nM, IC_{50} (**2e**): 24 nM, IC_{50} (**7g**): 154 nM]. The *o*-methoxy (**7a**), *p*-methoxy (**7b**), or *p*-hydroxy (**2b**) modifications at the 2-phenyl group were also

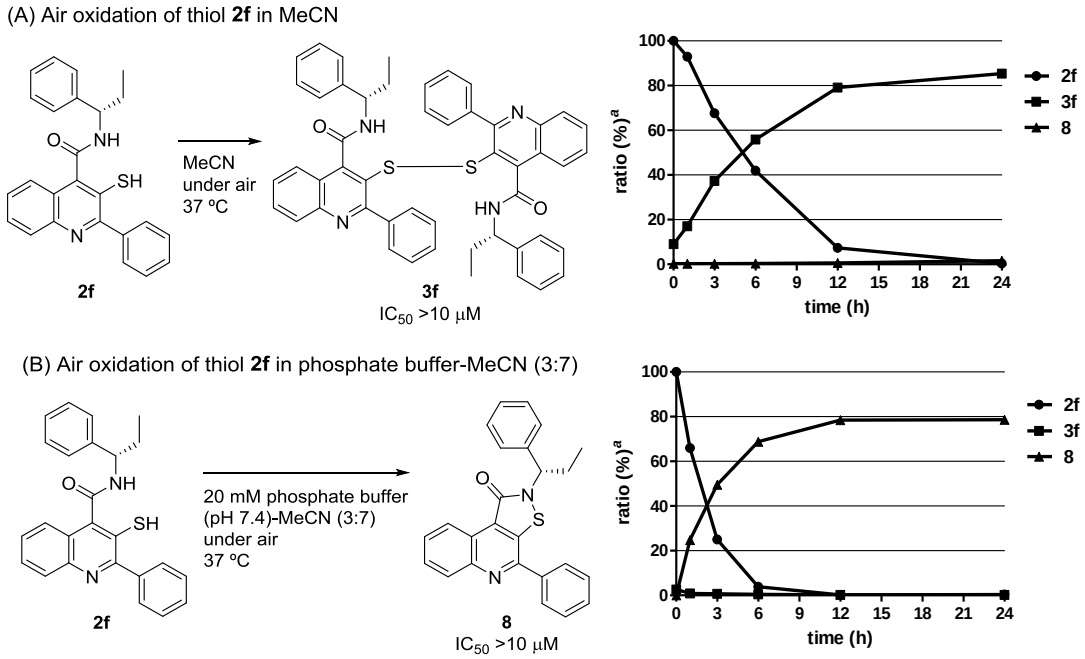
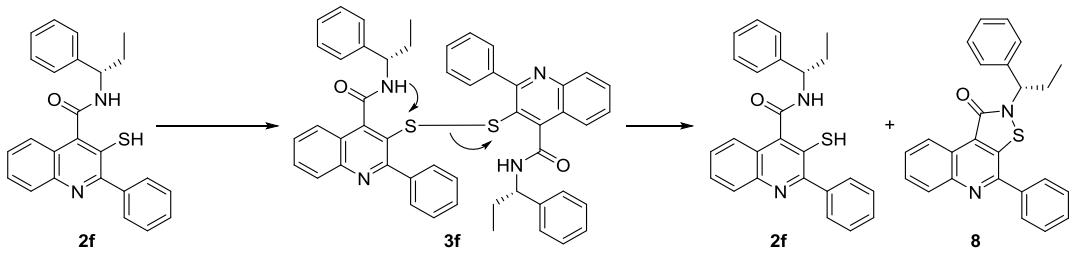


Figure 3. Investigation of the oxidation process of thiol **2f**. Compounds were dissolved in solvents at 1 mM. *a*The ratios of monomer units were calculated by HPLC analysis using calibration curves.

not favorable.

The structural conversion or degradation from the highly potent diol **2a** and thiol **2f** under oxidative conditions was tested (Table 2 and Figure 3). When compound **2a** was treated with mild oxidizing agents including hydrogen peroxide (H_2O_2), no reaction occurred to give the possible quinone methide form **3a** with the expected planar conformation. Treatment of **2a** with highly reactive oxidizing agents such as pyridinium chlorochromate (PCC) led to degradation without formation of **3a**. These results that a severe oxidizing condition is needed for decomposition of **2a**. In contrast, quinolinethiol **2f** was easily converted to the disulfide form **3f** in MeCN under air for 24 h at 37 °C (Figure 3A). In a solution of 20 mM phosphate buffer (pH 7.4)-MeCN, thiol **2f** was converted to isothiazolone **8** by the same treatment for 12 h at 37 °C (Figure 3B). Both oxidation products (**3f** and **8**) did not inhibit NK3R binding (IC_{50} >10 μM), suggesting that the thiol **2f** has an appropriate chemical property to offset the pharmacological effects under the conditions of the natural environment.

To investigate the oxidation process of thiol **2f**, transformations were investigated in solutions of various pH buffer (pH 5.0–10.0). For complete dissolution of the material, 70% MeCN was included for the analysis. In a solution of acetate buffer (pH 5.0), thiol **2f** was converted to the disulfide form **3f**. In contrast, thiol **2f** was converted to



Scheme 2. Plausible oxidation process of thiol **2f**.

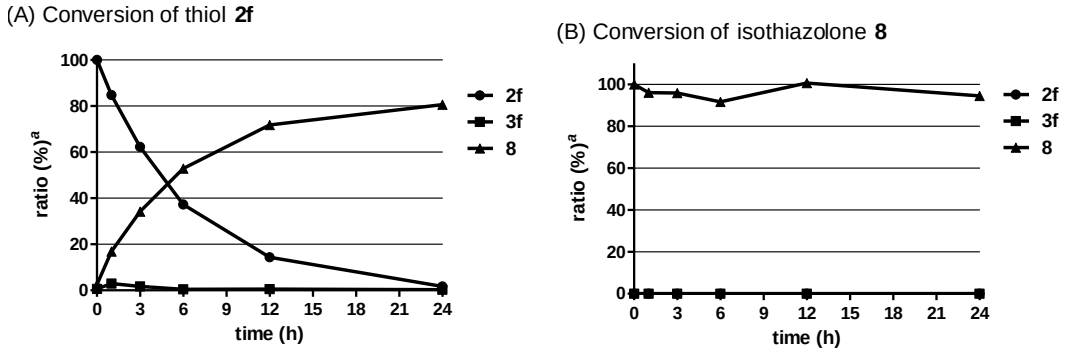


Figure 4. Stability of thiol **2f** and isothiazolone **8** in pig serum under air at 37 °C. Compounds were dissolved in pig serum at 0.1 mM. *a*The ratios of monomer units were calculated by HPLC analysis using calibration curves.

isothiazolone **8** in a solution of neutral or basic buffer (pH 7.0–10.0). Additionally, disulfide **3f** was disproportionated into thiol **2f** and isothiazolone **8** in a solution of phosphate buffer (pH 8.0). These results implied that thiol **2f** was initially converted to disulfide **3f** followed by the subsequent transformation into isothiazolone form **8** by the plausible mechanism (Scheme 2).

The stability of thiol **2f** in pig serum was investigated by HPLC analysis. Compound **2f** was slowly converted to isothiazolone **8** via air-oxidation in pig serum (Figure 4A), while no change from **8** was observed under the identical conditions over 24 h (Figure 4B). Thus, compound **2f** could be a novel NK3R antagonist, which is irreversibly converted into the inactive form **8** in the body after administration as well as under aerobic conditions after excretion from the body. The inactive compound **8** would be the predominant form present if compound **2f**-derived substances were uptaken from the environment to other organisms. Of note, it cannot be ruled out that the reverse reactions of isothiazolone **8** may occur under reductive conditions such as degradation by intestinal bacteria. Additionally, gradual inactivation of thiol **2f** in pig serum might be unfavorable for the prolonged in vivo biological activity. To overcome these disadvantages, further optimization including SAR studies and drug formulation may be needed.

In conclusion, the author revealed that talnetant derivatives **2a**, **2d** and **2f** with hydroxy- or mercapto-group modifications exhibited comparable NK3R binding inhibition to that of the parent compound talnetant. Among these potent derivatives, thiol **2f** was spontaneously converted to the inactive disulfide (**3f**) or isothiazolone (**8**) forms under aerobic conditions. Accordingly, the thiol **2f** may be a novel NK3R antagonist for the treatment of sex-hormone disorders with decreased environmental impact.

Experimental Section

General methods: ^{1}H NMR spectra were recorded using a JEOL ECA-500 spectrometer. Chemical shifts are reported in δ (ppm) relative to Me_4Si as an internal standard. ^{13}C NMR spectra were recorded using a JEOL ECA-500 spectrometer and referenced to the residual solvent signal. Exact mass (HRMS) spectra were recorded on a JMS-HX/HX 110A mass spectrometer or Shimadzu LC-ESI-IT-TOF-MS equipment. IR spectra were obtained on a JASCO FT/IR-4100 spectrometer. Optical rotations were measured with a JASCO P-1020 polarimeter. Melting points were measured by a hot stage melting points apparatus (uncorrected). For flash chromatography, Wakogel C-300E (Wako Pure Chemical Industries, Ltd) was employed. Compounds **4a**, **4b**, **5a-e** and **5g** were commercially available. Compounds **6e**,[1b] **6g**,[8] **7e**[1b] and **7g**[8] were prepared according to the literature.

1-Phenyl-2-[(2,4,6-trimethoxybenzyl)thio]ethan-1-one (5f). $NaBH_4$ (1.90 g, 51.0 mmol) was added to a solution of 2,4,6-trimethoxybenzaldehyde (5.00 g, 25.5 mmol) in MeOH (125 mL) at 0 °C. After being stirred at room temperature for 10 min, the reaction mixture was quenched with saturated NH_4Cl and concentrated. After the residue was extracted with EtOAc, the extract was washed with H_2O, and dried over $MgSO_4$. After concentration, the residue was dissolved in MeCN (51.0 mL). Thiourea (3.90 g, 51.0 mmol) and *p*-toluenesulfonic acid (8.80 g, 51.0 mmol) were added to the solution at 0 °C. After the mixture was stirred at room temperature for 30 min, NaOH (3.00 g, 76.5 mmol) was added at 0 °C, and the reaction was continued for 30 min at room temperature. The reaction mixture was acidified to pH 1 with 1 M HCl. After concentration, the residue was extracted with EtOAc. The extract was washed with H_2O and brine, and dried over $MgSO_4$. The filtrate was concentrated and the residue was dissolved in $CHCl_3$ (25.5 mL). To the solution were added phenacyl bromide (5.10 g, 25.5 mmol) and TEA (7.01 mL, 51.0 mmol) at 0 °C. After the mixture was stirred at room temperature for 2 h, 1 M HCl was added. The whole was washed with H_2O and brine, and dried over $MgSO_4$. The filtrate was concentrated, and the resulting residue was purified by flash chromatography over silica gel with *n*-hexane–EtOAc (5:1) to give the title compound **5f** (4.80 g, 57%): colorless solid; mp 71-73 °C; IR (neat) 1684 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 3.67 (s, 6H), 3.78 (s, 3H), 3.82 (s, 2H), 3.88 (s, 2H), 6.07 (s, 2H), 7.42-7.45 (m, 2H), 7.52-7.56 (m, 1H), 7.94 (d, *J* = 7.4 Hz, 2H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 24.1, 38.0, 55.2, 55.5 (2C), 90.3 (2C), 107.0, 128.3 (2C), 128.4 (2C), 132.9, 135.9, 158.8 (2C), 160.3, 194.6; *Anal.* calcd for $C_{18}H_{20}O_4S$: C, 65.04; H, 6.06. Found: C, 64.92; H, 5.90.

3-Hydroxy-2-(2-methoxyphenyl)quinoline-4-carboxylic acid (6a). 2-Acetoxy-2'-methoxyacetophenone (**5a**) (920 mg, 4.12 mmol) in EtOH (5.20 mL) were added to a solution of isatin (**4a**) (500 mg, 3.40 mmol) in 10 M NaOH (3.00 mL) and EtOH (800 μL) at 85 °C. After the mixture was stirred at 85 °C for 1 h, H_2O (9.00 mL) was added to the reaction mixture at 0 °C. The mixture was acidified to pH 1 with 1 M HCl. The precipitate was collected by filtration and washed with H_2O and EtOH to give the title compound **6a** (583 mg, 58%): pale yellow solid; mp 227-229 °C; IR (neat) 1654 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 3.77 (s, 3H), 7.13-7.14 (m, 1H), 7.21 (d, J = 8.0 Hz, 1H), 7.47-7.49 (m, 1H), 7.52-7.56 (m, 1H), 7.61-7.64 (m, 1H), 7.68-7.71 (m, 1H), 8.00-8.02 (m, 1H), 8.90-8.92 (m, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 55.7, 111.5 (2C), 120.3 (2C), 124.8, 126.0, 126.5 (2C), 128.6, 130.8 (2C), 131.1, 151.6, 157.3 (2C), 170.4; HRMS (ESI) calcd for $C_{17}H_{14}NO_4$ $[M+H]^+$: 296.0917, found: 296.0916.

3-Hydroxy-2-(4-methoxyphenyl)quinoline-4-carboxylic acid (6b). By use of a procedure similar to that described for the preparation of the compound **6a** from **4a** and **5a**, compounds **4a** (500 mg, 3.40 mmol) and **5b** (779 mg, 3.74 mmol) were converted to the title compound **6b** (742 mg, 74%): yellow solid; mp 217-219 °C; IR (neat) 1647 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 3.87 (s, 3H), 7.14 (d, J = 8.6 Hz, 2H), 7.59-7.65 (m, 2H), 8.03-8.05 (m, 1H), 8.07-8.09 (m, 2H), 8.92 (br s, 1H); ^{13}C NMR (125 MHz, 0.2 M NaOD in CD_3OD-D_2O (9:1)) δ 53.7, 111.9 (3C), 122.0, 123.0, 124.4, 126.1, 126.4, 130.0 (2C), 131.4, 138.3, 154.3, 154.7, 158.7, 175.5; HRMS (ESI) calcd for $C_{17}H_{14}NO_4$ $[M+H]^+$: 296.0917; found: 296.0908.

3-Hydroxy-7-methoxy-2-phenylquinoline-4-carboxylic acid (6c). By use of a procedure similar to that described for the preparation of the compound **6a** from **4a** and **5a**, compounds **4b** (1.00 g, 5.65 mmol) and **5c** (1.30 g, 7.35 mmol) were converted to the title compound **6c** (1.20 g, 74%): orange solid; mp 237-239 °C; IR (neat) 1629 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 3.91 (s, 3H), 7.27-7.30 (m, 1H), 7.43 (d, J = 2.9 Hz, 1H), 7.50-7.51 (m, 3H), 8.03-8.04 (m, 2H), 8.69-8.71 (m, 1H); ^{13}C NMR (125 MHz, 0.2 M NaOD in CD_3OD-D_2O (9:1)) δ 53.7, 105.6, 118.1, 118.9, 121.3, 125.8, 126.8 (2C), 127.5, 128.5 (2C), 137.1, 142.0, 151.6, 151.7, 156.7, 173.0; HRMS (ESI) calcd for $C_{17}H_{14}NO_4$ $[M+H]^+$: 296.0917; found: 296.0912.

2-(2-Methoxyphenyl)-3-(methylsulfonamido)-quinoline-4-carboxylic acid (6d). By use of a procedure similar to that described for the preparation of the compound **6a** from **4a** and **5a**, compounds **4a** (474 mg, 3.22 mmol) and **5d** (783 mg, 3.22 mmol) were converted to the title compound **6d** (241 mg, 20%): colorless solid; mp 260-262 °C; IR (neat) 1725 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 2.32 (s, 3H), 3.73 (s, 3H),

7.08-7.14 (m, 2H), 7.36-7.39 (m, 1H), 7.47-7.49 (m, 1H), 7.71-7.74 (m, 1H), 7.84-7.86 (m, 1H), 7.92 (d, J = 8.0 Hz, 1H), 8.07 (d, J = 8.0 Hz, 1H), 9.07 (s, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 41.8, 55.7, 111.0, 120.2, 123.2, 125.1, 125.6, 127.9, 128.1, 129.1, 130.3, 130.4, 131.3, 140.3, 146.0, 156.7, 158.9, 166.9; *Anal.* calcd for $C_{18}H_{16}N_2O_5S \cdot 0.5EtOH$: C, 57.71; H, 4.84; N, 7.08. Found: C, 57.62; H, 4.75; N, 7.04.

2-Phenyl-3-[(2,4,6-trimethoxybenzyl)thio]quinoline-4-carboxylic acid (6f). By use of a procedure similar to that described for the preparation of the compound **6a** from **4a** and **5a**, compound **4a** (2.80 g, 19.0 mmol) and **5f** (4.80 g, 14.6 mmol) were converted to the title compound **6f** (5.10 g, 51%): pale yellow solid; mp 241-243 °C; IR (neat) 1720 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 3.31 (s, 6H), 3.67 (s, 2H), 3.72 (s, 3H), 5.97 (s, 2H), 7.41-7.42 (m, 3H), 7.56-7.57 (m, 2H), 7.70-7.72 (m, 1H), 7.76-7.77 (m, 1H), 7.84-7.86 (m, 1H), 8.06 (d, J = 8.6 Hz, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 29.2, 55.2 (3C), 90.5 (2C), 105.1, 122.6, 123.0, 124.7, 127.3 (2C), 127.8, 128.0, 129.2, 129.6 (2C), 130.8, 139.8, 146.4, 148.8, 158.4 (2C), 160.4, 162.0, 167.5; HRMS (ESI) calcd for $C_{26}H_{24}NO_5S$ $[M+H]^+$: 462.1370; found: 462.1365.

(*S*)-3-Hydroxy-2-(2-methoxyphenyl)-*N*-(1-phenylpropyl)quinoline-4-carboxamide (7a). CDI (243 mg, 1.50 mmol) were added to a solution of compound **6a** (400 mg, 1.36 mmol) and TEA (247 μL, 1.77 mmol) in MeCN (1.60 mL). After the mixture was stirred at 50 °C for 5 h, (*S*)-1-phenylpropan-1-amine (218 μL, 1.50 mmol) was added to the reaction mixture at 50 °C. After being stirred at 50 °C for 5 h, the reaction mixture was stirred at room temperature overnight. After the reaction mixture was concentrated, the residue was dissolved in EtOAc. The extract was washed with 10% $NaHCO_3$, 1 M HCl, H_2O, and brine, and dried over $MgSO_4$. The filtrate was concentrated, and the residue was recrystallized from *n*-hexane–*n*-PrOAc to give the title compound **7a** (137 mg, 25%): colorless solid; $[\alpha]^{28}_D$ –22.4 (*c* 0.93, $CHCl_3$); mp 156-158 °C; IR (neat) 3349-3112 (OH), 1638 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.94 (t, J = 7.4 Hz, 3H), 1.73-1.83 (m, 2H), 3.73 (s, 3H), 5.01-5.05 (m, 1H), 7.06-7.09 (m, 1H), 7.12-7.14 (m, 1H), 7.25-7.27 (m, 1H), 7.29-7.31 (m, 1H), 7.34-7.37 (m, 2H), 7.43-7.49 (m, 4H), 7.53-7.55 (m, 2H), 7.91-7.93 (m, 1H), 9.08 (d, J = 8.0 Hz, 1H), 9.18 (br s, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.2, 29.3, 54.7, 55.4, 111.4, 120.2, 123.5, 125.7, 125.9, 126.3, 126.7 (3C), 126.8, 127.0, 128.2 (2C), 128.8, 130.1, 130.8, 141.9, 143.6, 145.1, 152.0, 157.3, 164.7; *Anal.* calcd for $C_{26}H_{24}N_2O_3$: C, 75.71; H, 5.86; N, 6.79. Found: C, 75.68; H, 5.91; N, 6.82.

(*S*)-3-Hydroxy-2-(4-methoxyphenyl)-*N*-(1-phenylpropyl)quinoline-4-carboxamide (7b). By use of a procedure similar to that described for the preparation of the compound **7a** from **6a**, compound **6b** (300 mg, 1.02 mmol) was converted to the title compound **7b**

(169 mg, 40%): yellow solid; $[\alpha]^{28}_{D}$ –57.5 (*c* 1.21, $CHCl_3$); mp 75-77 °C; IR (neat) 1605 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.95 (t, *J* = 7.2 Hz, 3H), 1.73-1.87 (m, 2H), 3.84 (s, 3H), 5.02-5.07 (m, 1H), 7.06-7.08 (m, 2H), 7.26-7.29 (m, 1H), 7.36-7.38 (m, 2H), 7.44-7.46 (m, 3H), 7.50-7.51 (m, 1H), 7.54-7.57 (m, 1H), 7.95-8.00 (m, 3H), 9.14 (d, *J* = 8.0 Hz, 1H), 9.78 (br s, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.2, 29.3, 54.8, 55.2, 113.4 (2C), 123.6, 125.2, 126.6, 126.7 (4C), 128.0, 128.2 (2C), 128.9, 130.2, 130.9 (2C), 142.4, 143.5, 144.5, 151.8, 159.8, 164.8; *Anal.* calcd for $C_{26}H_{24}N_2O_3$: C, 75.71; H, 5.86; N, 6.79. Found: C, 75.49; H, 6.02; N, 6.72.

(*S*)-3-Hydroxy-2-phenyl-*N*-(1-phenylpropyl)-7-methoxyquinoline-4-carboxamide (7c). By use of a procedure similar to that described for the preparation of the compound **7a** from **6a**, compound **6c** (1.00 g, 3.39 mmol) was converted to the title compound **7c** (850 mg, 61%): pale yellow solid; $[\alpha]^{28}_{D}$ –23.2 (*c* 1.10, DMSO); mp 190-192 °C; IR (neat) 1617 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.95 (t, *J* = 7.4 Hz, 3H), 1.74-1.84 (m, 2H), 3.89 (s, 3H), 5.01-5.06 (m, 1H), 7.16-7.18 (m, 1H), 7.26-7.28 (m, 1H), 7.35-7.39 (m, 3H), 7.42-7.48 (m, 4H), 7.50-7.52 (m, 2H), 7.95-7.97 (m, 2H), 9.12 (d, *J* = 8.6 Hz, 1H), 9.46 (br s, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.2, 29.3, 54.8, 55.4, 107.5, 119.6, 120.2, 124.8, 126.7 (3C), 127.9 (2C), 128.2 (2C), 128.6, 129.3, 129.4 (2C), 138.0, 142.9, 143.5, 143.9, 152.2, 158.3, 164.7; *Anal.* calcd for $C_{26}H_{24}N_2O_3$: C, 75.71; H, 5.86; N, 6.79. Found: C, 75.57; H, 5.97; N, 6.75.

(*S*)-2-(2-Methoxyphenyl)-3-(methylsulfonamido)-*N*-(1-phenylpropyl)quinoline-4-carboxamide (7d). By use of a procedure similar to that described for the preparation of the compound **7a** from **6a**, compound **6d** (100 mg, 0.269 mmol) was converted to the title compound **7d** (80.2 mg, 30%): colorless solid; $[\alpha]^{28}_{D}$ +47.7 (*c* 1.01, $CHCl_3$); mp 107-109 °C; IR (neat) 1647 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.92 (t, *J* = 7.4 Hz, 3H), 1.78-1.98 (m, 2H), 2.35 (s, 3H), 3.77 (s, 3H), 5.00-5.04 (m, 1H), 7.06-7.09 (m, 1H), 7.13 (d, *J* = 8.0 Hz, 1H), 7.26-7.29 (m, 1H), 7.36-7.38 (m, 3H), 7.43-7.47 (m, 3H), 7.54-7.56 (m, 1H), 7.62-7.64 (m, 1H), 7.75-7.78 (m, 1H), 8.01-8.02 (m, 1H), 8.23 (br s, 1H), 8.92 (d, *J* = 8.0 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 9.8, 28.2, 40.2, 55.4, 55.8, 111.3, 121.6, 122.2, 124.2, 124.8 (2C), 126.1 (2C), 126.5, 126.9, 127.5, 127.6 (2C), 128.6, 129.6, 130.5, 131.6 (2C), 146.6, 155.0, 155.2, 163.5; *Anal.* calcd for $C_{27}H_{27}N_3O_4S \cdot 0.3H_2O$: C, 65.44; H, 5.63; N, 8.48. Found: C, 65.60; H, 5.66; N, 8.15.

(*S*)-2-Phenyl-*N*-(1-phenylpropyl)-3-[(2,4,6-trimethoxybenzyl)thio]quinoline-4-carboxamide (7f). HATU (6.34 g, 16.7 mmol), (*S*)-1-phenylpropylamine (3.20 mL, 22.2 mmol) and DIPEA (7.70 mL, 44.5 mmol) were added to a solution of 2-phenyl-3-[(2,4,6-trimethoxybenzyl)thio]quinoline-4-carboxylic acid (**6f**) (5.10 g, 11.1 mmol) in DMF

(16.7 mL). After being stirred at room temperature for 4 h, the mixture was concentrated. After the residue was dissolved in EtOAc, the whole was washed with 1 M HCl, H_2O, and brine, and dried over $MgSO_4$. The filtrate was concentrated, and the resulting residue was purified by flash chromatography over silica gel with *n*-hexane–EtOAc (2:1) to give the title compound **7f** (5.80 g, 91%): brown amorphous solid; $[\alpha]^{28}_D$ +55.2 (*c* 0.53, $CHCl_3$); IR (neat) 1647 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 0.99 (t, *J* = 7.2 Hz, 3H), 1.78-1.85 (m, 2H), 3.35 (s, 6H), 3.71-3.72 (m, 5H), 5.07-5.11 (m, 1H), 6.00 (s, 2H), 7.26-7.29 (m, 1H), 7.33-7.36 (m, 2H), 7.39-7.41 (m, 3H), 7.46-7.47 (m, 2H), 7.55-7.56 (m, 4H), 7.76-7.80 (m, 1H), 8.03 (d, *J* = 8.6 Hz, 1H), 8.97 (d, *J* = 8.6 Hz, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.2, 29.3, 29.7, 54.8, 55.2, 55.3 (2C), 90.5 (2C), 105.3, 124.1, 124.8, 124.9, 126.7 (2C), 126.8, 127.2 (3C), 127.9, 128.1 (2C), 129.0, 129.6 (2C), 130.3, 140.3, 143.2, 146.3, 150.3, 158.4 (2C), 160.3, 161.9, 165.3; HRMS (ESI) calcd for $C_{35}H_{35}N_2O_4S$ $[M + H]^+$: 579.2312, found: 579.2309.

(*S*)-3-Hydroxy-2-(2-hydroxyphenyl)-*N*-(1-phenylpropyl)quinoline-4-carboxamide (2a). BBr_3 (17% solution in CH_2Cl_2; 1.21 mL, 2.17 mmol) was added to compound **7a** (50.0 mg, 0.121 mmol) at 0 °C. After being stirred at room temperature for 5 h, the mixture was quenched with H_2O and extracted with EtOAc. The extract was washed with brine and dried over $MgSO_4$. After concentration, the residue was purified by flash chromatography over silica gel with *n*-hexane–EtOAc (2:1) to give the title compound **2a** (39.6 mg, 82%): colorless solid; $[\alpha]^{28}_D$ –21.7 (*c* 1.10, DMSO); mp 185-187 °C; IR (neat) 1636 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 0.96 (t, *J* = 7.2 Hz, 3H), 1.76-1.82 (m, 2H), 5.03-5.07 (m, 1H), 6.96-7.01 (m, 2H), 7.25-7.28 (m, 1H), 7.33-7.38 (m, 3H), 7.44-7.45 (m, 2H), 7.49-7.52 (m, 1H), 7.55-7.61 (m, 2H), 7.83-7.85 (m, 1H), 7.97 (d, *J* = 8.0 Hz, 1H), 9.15-9.16 (m, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.2, 29.4, 54.8, 116.5, 119.0, 123.1, 123.6, 125.4, 126.7 (3C), 126.8, 127.0, 127.1, 128.2 (3C), 128.3, 130.7, 131.4, 143.5 (2C), 151.6, 156.4, 164.5; *Anal*. calcd for $C_{25}H_{22}N_2O_3$: C, 75.36; H, 5.57; N, 7.03. Found: C, 75.27; H, 5.70; N, 7.00.

(*S*)-3-Hydroxy-2-(4-hydroxyphenyl)-*N*-(1-phenylpropyl)quinoline-4-carboxamide (2b). By use of a procedure similar to that described for the preparation of the compound **2a** from **7a**, compound **7b** (50.0 mg, 0.121 mmol) was converted to the title compound **2b** (27.6 mg, 57%): yellow solid; $[\alpha]^{28}_D$ –20.5 (*c* 1.03, MeOH); mp 109-111 °C; IR (neat) 3388-3125 (OH), 1650 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 0.95 (t, *J* = 7.2 Hz, 3H), 1.73-1.86 (m, 2H), 5.02-5.06 (m, 1H), 6.91-6.93 (m, 2H), 7.26-7.29 (m, 1H), 7.35-7.38 (m, 2H), 7.43-7.44 (m, 2H), 7.48-7.53 (m, 2H), 7.59-7.61 (m, 1H), 7.87-7.90 (m, 2H), 7.99-8.00 (br m, 1H), 9.17-9.18 (m, 1H), 9.91 (br s, 1H); ^{13}C NMR (125 MHz,

DMSO-d_6) δ 11.2, 29.3, 54.8, 114.8 (2C), 123.6, 125.1, 126.5, 126.6, 126.7 (3C), 127.8, 128.2 (2C), 128.6, 128.8, 131.0 (2C), 142.4, 143.5, 144.5, 152.1, 158.3, 164.9; *Anal.* calcd for $C_{25}H_{22}N_2O_3 \cdot 0.7H_2O$: C, 73.15; H, 5.73; N, 6.82. Found: C, 72.99; H, 5.62; N, 6.70.

(*S*)-3,7-Dihydroxy-2-phenyl-*N*-(1-phenylpropyl)quinoline-4-carboxamide (2c). By use of a procedure similar to that described for the preparation of the compound **2a** from **7a**, compound **7c** (500 mg, 1.21 mmol) was converted to the title compound **2c** (297 mg, 62%): pale yellow solid; $[\alpha]^{28}_D$ –16.2 (*c* 0.96, DMSO); mp 265-267 °C; IR (neat) 3177-2973 (OH), 1605 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.95 (t, *J* = 7.2 Hz, 3H), 1.76-1.82 (m, 2H), 5.01-5.05 (m, 1H), 7.05-7.07 (m, 1H), 7.23-7.28 (m, 2H), 7.35-7.38 (m, 3H), 7.43-7.51 (m, 5H), 7.92-7.94 (m, 2H), 9.10 (d, *J* = 8.0 Hz, 1H), 9.29 (br s, 1H), 9.91 (br s, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.2, 29.3, 54.8, 110.3, 119.4, 119.6, 124.8, 126.7 (3C), 127.9 (2C), 128.2 (2C), 128.5, 129.2, 129.4 (2C), 138.2, 142.3, 143.6, 144.2, 152.2, 156.4, 165.0; *Anal.* calcd for $C_{25}H_{22}N_2O_3$: C, 75.36; H, 5.57; N, 7.03. Found: C, 75.22; H, 5.70; N, 6.98.

(*S*)-2-(2-Hydroxyphenyl)-3-(methanesulfonylamino)-*N*-(1-phenylpropyl)quinoline-4-carboxamide (2d). By use of a procedure similar to that described for the preparation of the compound **2a** from **7a**, compound **7d** (30.0 mg, 0.0613 mmol) was converted to the title compound **2d** (35.3 mg, 73%): pale yellow solid; $[\alpha]^{28}_D$ –1.22 (*c* 1.17, MeOH); mp 129-131 °C; IR (neat) 1641 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.94 (t, *J* = 7.2 Hz, 3H), 1.73-1.90 (m, 2H), 2.38 (s, 3H), 5.01-5.02 (m, 1H), 6.97-7.01 (m, 2H), 7.28-7.35 (m, 2H), 7.38-7.39 (m, 2H), 7.47-7.50 (m, 3H), 7.59-7.65 (m, 2H), 7.80-7.84 (m, 1H), 8.07 (d, *J* = 8.6 Hz, 1H), 9.34 (d, *J* = 8.0 Hz, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.2, 29.2, 41.8, 55.4, 116.0, 119.4, 124.4, 125.2, 125.8, 126.6, 127.0 (3C), 127.4, 128.3 (2C), 129.0, 130.2, 130.5, 132.4, 143.0, 143.5, 146.1, 154.4, 159.0, 164.6; HRMS (FAB) calcd for $C_{26}H_{26}N_3O_4S$ $[M+H]^+$: 476.1639; found: 476.1631.

(*S*)-3-Azido-2-phenyl-*N*-(1-phenylpropyl)quinoline-4-carboxamide (2e). Trimethylsilyl azide (39.0 μL, 0.296 mmol) and *tert*-butyl nitrite (35.4 μL, 0.296 mmol) were added to a solution of (*S*)-3-amino-2-phenyl-*N*-(1-phenylpropyl)quinoline-4-carboxamide (**7e**)[1b] (50.0 mg, 0.197 mmol) at 0 °C. After being stirred at room temperature for 5 h, the reaction mixture was concentrated. Crude product was purified by flash chromatography over silica gel with EtOAc–*n*-hexane (2:1) to give the title compound **2e** (35.0 mg, 44%.): pale yellow solid; $[\alpha]^{28}_D$ –20.3 (*c* 0.67, $CHCl_3$); mp 135-137 °C; IR (neat) 1632 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.96 (t, *J* = 7.2 Hz, 3H), 1.78-1.83 (m, 2H), 5.03-5.08 (m, 1H), 7.27-7.30 (m, 1H), 7.37-7.40 (m, 2H), 7.43-7.44 (m, 2H), 7.53-7.65 (m, 5H), 7.77-7.82 (m, 3H), 8.08 (d, *J* = 8.6 Hz, 1H), 9.44 (d, *J* = 8.6 Hz, 1H); ^{13}C NMR (125

MHz, DMSO-d_6) δ 11.0, 29.3, 55.0, 124.1, 124.3, 126.6 (2C), 126.9, 127.0, 128.1, 128.3 (2C), 128.5 (2C), 129.2 (3C), 129.3, 129.8, 135.8, 137.3, 143.0, 144.9, 153.6, 163.2; *Anal.* calcd for $C_{25}H_{21}N_5O$: C,73.69; H, 5.19; N, 17.19. Found: C, 73.53; H, 5.23; N, 17.25.

3,3'-Dithiobis{2-phenyl-*N*-[(*S*)-1-phenylpropyl]quinoline-4-carboxamide} (3f). Compound **7f** (5.70 g, 9.90 mmol) was added to a solution of thioanisole (11.6 mL, 99.0 mmol) in TFA (99.0 mL). After being stirred at room temperature for 2 h, the mixture was concentrated. MeCN (49.5 mL) was added to the residue. After being stirred at room temperature under O_2 for 4 days, the mixture was concentrated. Crude product was purified by flash chromatography over silica gel with EtOAc–*n*-hexane (2:1) to give the title compound **3f** (2.00 g, 51%.): pale yellow solid; $[\alpha]^{28}_D$ –98.7 (*c* 1.12, MeOH); mp 222-224 °C; IR (neat) 1630 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.72 (t, *J* = 7.4 Hz, 6H), 1.46-1.60 (m, 4H), 4.65-4.70 (m, 2H), 7.12-7.16 (m, 8H), 7.19-7.27 (m, 8H), 7.34-7.36 (m, 4H), 7.51-7.56 (m, 4H), 7.78-7.82 (m, 2H), 8.02 (d, *J* = 8.0 Hz, 2H), 8.54 (d, *J* = 7.4 Hz, 2H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 10.4 (2C), 28.4 (2C), 55.1 (2C), 124.0 (2C), 124.4 (2C), 125.3 (2C), 126.6 (2C), 126.7 (2C), 126.8 (2C), 127.1 (4C), 127.3 (2C), 127.7 (2C), 128.0 (2C), 128.1 (2C) 129.2 (2C), 129.4 (2C), 129.5 (2C), 130.9 (2C), 140.0 (2C), 142.1 (2C), 147.1 (2C), 149.5 (2C), 161.4 (2C), 164.5 (2C); *Anal.* calcd for $C_{50}H_{42}N_4O_2S_2 \cdot 1.5H_2O$: C, 73.05; H, 5.52; N, 6.82. Found: C, 72.80; H, 5.31; N, 6.70.

(*S*)-3-Mercapto-2-phenyl-*N*-(1-phenylpropyl)quinoline-4-carboxamide (2f). L-Cysteine hydrochloride (426 mg, 3.52 mmol) was added to a solution of compound **3f** (1.00 g, 1.26 mmol) in MeOH (50.0 mL) and H_2O (15.0 mL) at 0 °C. After being stirred at room temperature for 30 min, the mixture was filtrated. The filtrate was concentrated and the precipitate was collected by filtration to give the title compound **2f** (746 mg, 74%): pink solid; $[\alpha]^{28}_D$ –61.8 (*c* 0.75, MeOH); mp 87-89 °C; IR (neat) 3330-3111 (SH), 1629 (C=O) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.98 (t, *J* = 7.2 Hz, 3H), 1.75-1.87 (m, 2H), 5.04-5.09 (m, 1H), 7.28-7.30 (m, 1H), 7.37-7.40 (m, 2H), 7.44-7.45 (m, 2H), 7.52-7.58 (m, 4H), 7.62-7.63 (m, 3H), 7.72-7.75 (m, 1H), 8.02 (d, *J* = 8.6 Hz, 1H), 9.47 (d, *J* = 8.0 Hz, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.3, 29.2, 55.0, 123.9, 124.0, 126.7 (3C), 127.0, 127.8, 128.3 (4C), 128.9 (3C), 129.1, 129.4, 139.2, 142.3, 143.1, 144.5, 157.9, 165.4; *Anal.* calcd for $C_{25}H_{22}N_2OS$: C, 75.35; H, 5.56; N, 7.03. Found: C, 75.13; H, 5.72; N, 6.78.

(*S*)-4-Phenyl-2-(1-phenylpropyl)isothiazolo[5,4-*c*]quinolin-1(2*H*)-one (8) (Authentic Sample). *N*-Bromosuccinimide (756 mg, 4.25 mmol) was added to a solution of (*S*)-3-[(4-methoxybenzyl)thio]-2-phenyl-*N*-(1-phenylpropyl)quinoline-4-carboxamide (**7g**)[8] (500 mg, 0.965 mmol) in MeCN (20 mL). After being stirred at room temperature for 2

days, the reaction mixture was concentrated and filtered through amine silica gel. Crude product was purified by HPLC to give the title compound **8** (136 mg, 36%): yellow solid; $[\alpha]^{28}_{D}$ –224 (*c* 0.98, $CHCl_3$); IR (neat) 1649 (C=O) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 0.96 (t, *J* = 7.2 Hz, 3H), 2.26-2.32 (m, 2H), 5.78-5.81 (m, 1H), 7.32-7.33 (m, 1H), 7.39-7.42 (m, 2H), 7.51-7.52 (m, 2H), 7.63-7.67 (m, 3H), 7.80-7.85 (m, 2H), 7.96-7.97 (m, 2H), 8.23 (d, *J* = 8.6 Hz, 1H), 9.26 (d, *J* = 8.0 Hz, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 11.1, 26.5, 58.5, 121.6, 123.0, 124.1, 127.2 (2C), 127.5 (2C), 128.1, 128.6, 128.8 (2C), 129.4 (4C), 130.6, 134.4, 137.3, 139.4, 145.2, 150.9, 164.5; *Anal.* calcd for $C_{25}H_{20}N_2OS$: C, 75.73; H, 5.08; N, 7.07. Found: C, 75.71; H, 5.02; N, 7.03.

Inhibitory activity of talnetant derivatives against NKB binding to NK3R. Membranes from NK3R-expressing CHO cells were incubated with 50 μL solution of talnetant derivatives, 25 μL of radioactive ligand solution [([^{125}I]His^3, MePhe^7)-NKB, 0.4 nM, PerkinElmer Life Sciences], and 25 μL of NK3R membrane suspension (10 μg) in assay buffer [50 mM HEPES (pH 7.4), 5 mM $MgCl_2$, 1 mM $CaCl_2$, 0.1% BSA]. Reaction mixtures were filtered through GF/B filters, pretreated with 0.3% polyethyleneimine. Filters were washed with wash buffer [50 mM HEPES (pH 7.4), 500 mM NaCl, 0.1% BSA] and dried at 55 °C. Bound radioactivity was measured by TopCount (PerkinElmer Life Sciences) in the presence of MicroScint-O (30 μL) (PerkinElmer Life Sciences).

Quantitative analysis of air oxidation process from thiol 2f. Compound **2f** (1 mM) was incubated in MeCN or a mixture of 20 mM phosphate buffer (pH 7.4) and MeCN [30:70 (v/v)] under air at 37 °C. An aliquot of the sample was analyzed by HPLC at the indicated intervals and the peak area was recorded by UV detection at 254 nm. The ratios of the resulting compounds were calculated from the calibration curves.

Quantitative analysis of the conversion of thiol 2f to isothiazolone 8 in pig serum. Compound **2f** (0.1 mM) was incubated in pig serum (containing 0.5% DMSO) at 37 °C. A 20 μL aliquot was sampled at the indicated intervals, and extracted with MeCN (80 μL). An aliquot of the sample was analyzed by HPLC and the peak area was recorded by UV detection at 254 nm. The ratios of the resulting compounds were calculated from the calibration curves.

Preparation of animal sera. Pig serum was obtained from 5-month-old female hybrid pigs. After the collection, blood was incubated overnight at 4 °C, centrifuged (4 °C, 1,710 g, 15 min), and the supernatant was used as sera. The experiments were conducted in accordance with the guidelines of the Committee on the Care and Use of Experimental Animals at the Graduate School of Bioagricultural Sciences, Nagoya University.

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Chapter 2. Development of NK3R Antagonists with a Degradable Scaffold in the Natural Environment

Section 1. Synthesis and Application of Fused Piperazine Derivatives for Investigation of Degradable Core Motifs

Summary

For investigation of decomposable scaffolds for the fezolinetant triazopiperazine core in the natural environment, an efficient method for synthesis of [1,2,4]triazolo[4,3-a]piperazine derivatives was established based on gold(I)-catalyzed domino cyclization of an amidrazone substrate with a terminal alkyne. The amidoxime congeners were also converted into [1,2,4]oxadiazolo[4,5-a]piperazine derivatives in the presence of a gold catalyst.

Fezolinetant is an NK3 receptor (NK3R) antagonist with a unique triazolopiperazine scaffold, which is being developed for treatment of hot flashes.[1,2] Triazolopiperazine scaffolds that are also included in a variety of bioactive compounds such as sitagliptin (dipeptidyl peptidase-4 inhibitor for antidiabetic agent), have a high stability in the natural environment (Figure 1).[3-5] To develop novel NK3R antagonists with a degradable scaffold in the natural environment, the author planned a structure-activity relationship study of the triazolopiperazine part of fezolinetant by scaffold hopping for decomposable motifs. Scaffold hopping is an approach to improve the bioactivities and physicochemical properties of a lead compound by substitution of the core structures in the lead compounds with other motif(s).[6] The author expected that dearomatized derivatives of the triazolopiperazine could be easily decomposable in the natural environment. To accomplish this, the synthetic method of novel fused piperazine scaffolds was developed by gold(I)-catalyzed domino cyclization.

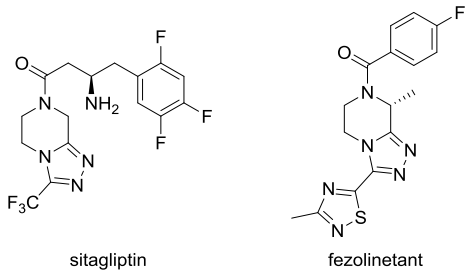
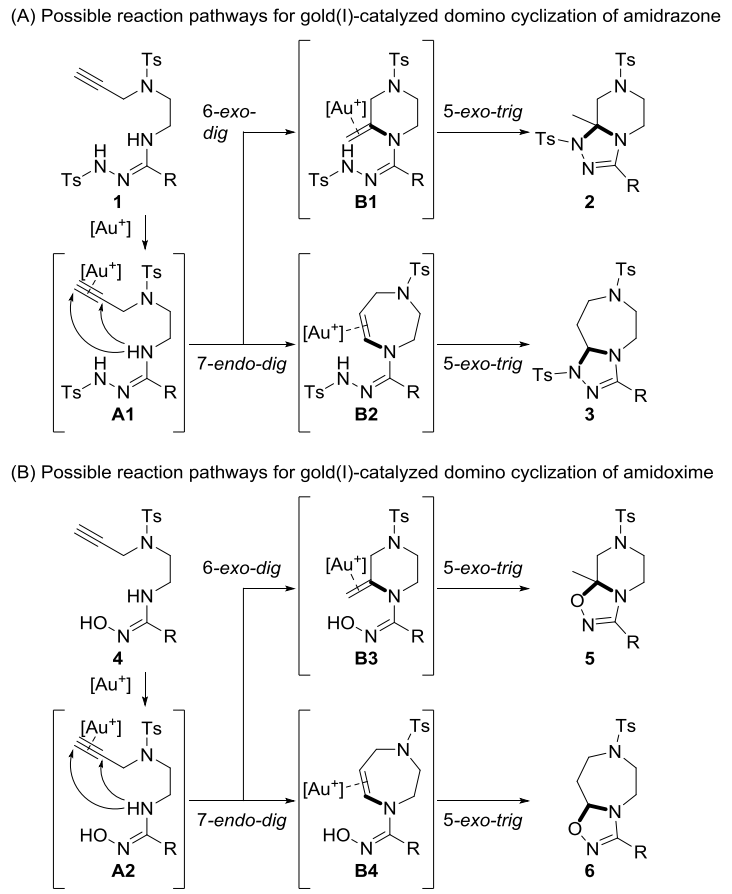
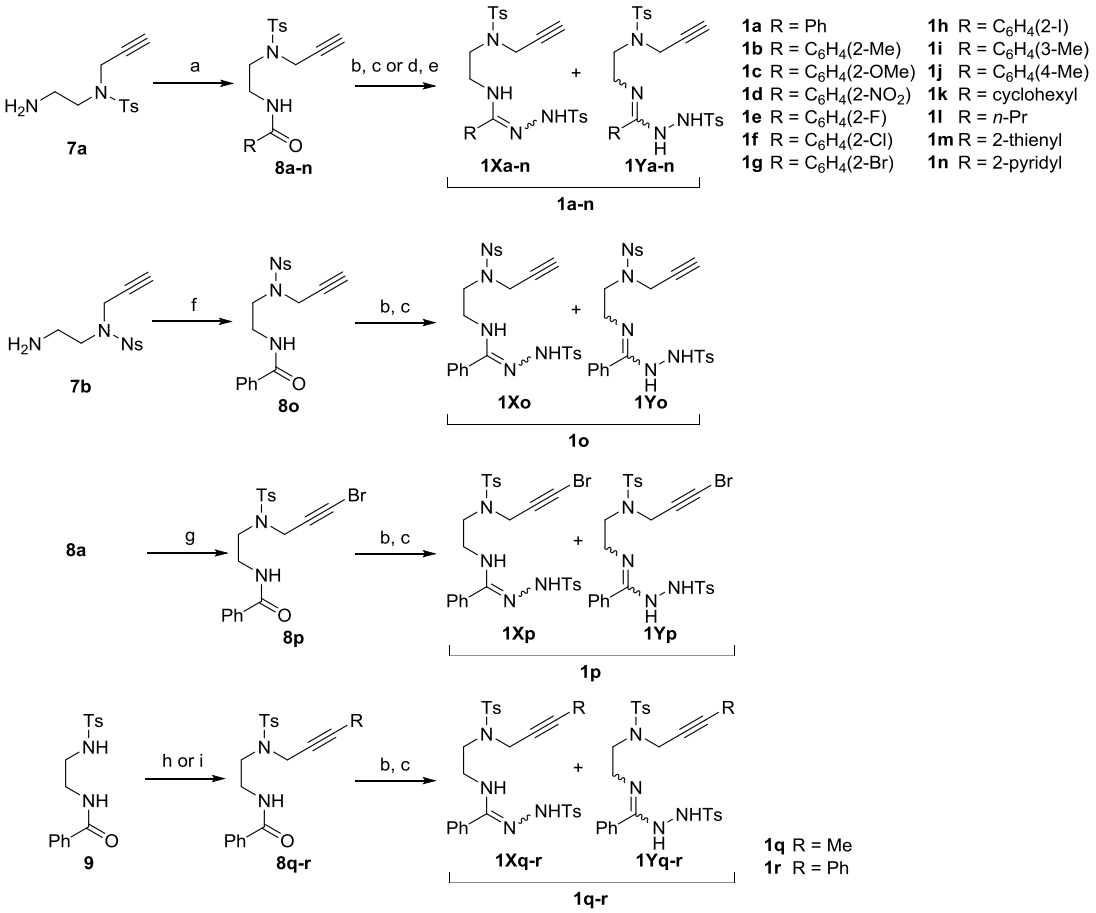


Figure 1. Structures of bioactive [1,2,4]triazolo[4,3-*a*]piperazine derivatives.



Scheme 1. Possible reaction pathways for gold(I)-catalyzed domino cyclization.

During the past decade, gold(I) catalysts have attracted considerable attention as effective π-acids for the activation of alkynes.[7] The gold-catalyzed hydroalkoxylation and hydroamination of alkynes and alkenes have been used extensively to synthesize a broad range of heterocycles.[8-10] The author envisaged that [1,2,4]triazolo[4,3-*a*]piperazine or related heterocycles would be provided by a gold-catalyzed intramolecular cascade reaction of open-chain precursors bearing an alkyne moiety. That is, the gold-catalyzed reaction of amidrazone **1** would lead to the formation of piperazine **B1** and tetrahydro-1,4-diazepine **B2** through a 6-*exo-dig* type and 7-*endo-dig* type hydroamination of **A1**, respectively (Scheme 1A). The piperazine (**B1**) and 1,4-diazepine (**B2**) intermediates could be converted into 1,2,4-triazole scaffolds (**2** and **3**, respectively) by the subsequent 5-*exo-trig* type hydroamination (aminal formation).[11] It was also expected that 1,2,4-oxadiazoles **5** and/or **6** could be obtained when using amidoxime **4** as a substrate (Scheme 1B). In this section, the author describes a novel synthetic approach for [1,2,4]triazolo[4,3-*a*]piperazine derivatives **2** and their 1,2,4-oxadiazole congeners by a gold-catalyzed intramolecular cascade reaction. The applications of the

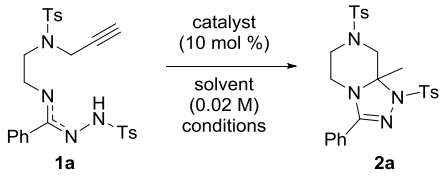


Scheme 2. Synthesis of amidrazone **1a**–**r**. *Reagents and conditions*: (a) R-CO_2H, WSC·HCl, HOBt, CH_2Cl_2, rt; (b) Tf_2O, 2-methoxypyridine, CH_2Cl_2, −20 °C; (c) $TsNHNH_2$, −20 °C to rt; (d) PCl_5, toluene, reflux; (e) $TsNHNH_2$, rt; (f) Ph-CO_2H, WSC·HCl, HOBt, CH_2Cl_2, rt; (g) NBS, $AgNO_3$, acetone, rt to 0 °C; (h) 1-bromo-2-butyne, K_2CO_3, MeCN, 60 °C; (i) 3-bromo-1-phenylpropyne, K_2CO_3, MeCN, 60 °C.

[1,2,4]oxadiazolo[4,5-*a*]piperazine scaffold **5** to medicinal chemistry approaches are also presented.

Initially, the author synthesized the substrate amidrazones (Scheme 2). To prepare benzamides **8a**–**o**, *N*-propargyl sulfonamides **7a** or **7b** were coupled with the corresponding benzoic acids using WSC·HCl and HOBt in CH_2Cl_2. The amidrazones **1a**–**m** and **1o** were obtained by the reaction of the benzamides **8a**–**m** and **8o** with trifluoromethanesulfonic anhydride (Tf_2O) followed by conjugation with *N*-tosylhydrazine[12] as an inseparable mixture of hydrazonamide form **1X** and imidohydrazine form **1Y**. The amidrazone **1n** was prepared by treatment of benzamide **8n** with phosphorus pentachloride and *N*-tosylhydrazine in toluene. For preparation

Table 1. Optimization of reaction conditions.



entry	catalyst	solvent[a]	conditions	yield (%)[b]
1	IPrAuCl/$AgNTf_2$	DCE	50 °C, 21 h	19
2	JohnPhosAuCl/ $AgNTf_2$	DCE	50 °C, 21 h	45
3	BrettPhosAuCl/ $AgNTf_2$	DCE	50 °C, 21 h	38
4	$Ph_3PAuCl/AgNTf_2$	DCE	50 °C, 21 h	64
5	*t*-$Bu_3PAuCl/AgNTf_2$	DCE	50 °C, 21 h	21
6	**L**$AuCl/AgNTf_2$	DCE	50 °C, 21 h	73
7	Tf_2NH	DCE	50 °C, 21 h	0
8	$(Ph_3C)B(C_6F_5)_4$	DCE	50 °C, 21 h	0
9	$Ph_3PAuCl/AgNTf_2$	DCE	80 °C, 3 h	74
10	$Ph_3PAuCl/AgNTf_2$	TCE	100 °C, 2 h	57
11	$Ph_3PAuCl/AgOTf$	DCE	80 °C, 98 h	38
12	$Ph_3PAuCl/AgSbF_6$	DCE	80 °C, 9.5 h	72
13	$Ph_3PAuCl/AgNTf_2$	MeCN	80 °C, 2 h	53
14	$Ph_3PAuCl/AgNTf_2$	*i*-PrOH	80 °C, 5 h	63
15	$Ph_3PAuCl/AgNTf_2$	1,4-dioxane	80 °C, 5 h	72
16	$Ph_3PAuCl/AgNTf_2$	toluene	80 °C, 2.5 h	74
17[c]	$Ph_3PAuCl/AgNTf_2$	toluene	80 °C, 9 h	60
18	**L**$AuCl/AgNTf_2$	toluene	80 °C, 2.5 h	78
19	Ph_3PAuCl	DCE	80 °C, 42 h	0
20	$AgNTf_2$	DCE	80 °C, 42 h	28
21	[$RuCl_2$(*p*-cymene)]/*n*-Bu_3P	DCE	80 °C, 42 h	0

[a]DCE: 1,2-dichloroethane; TCE: 1,1,2,2-tetrachloroethane. [b]Isolated yield. [c]5 mol % of the catalyst was used.

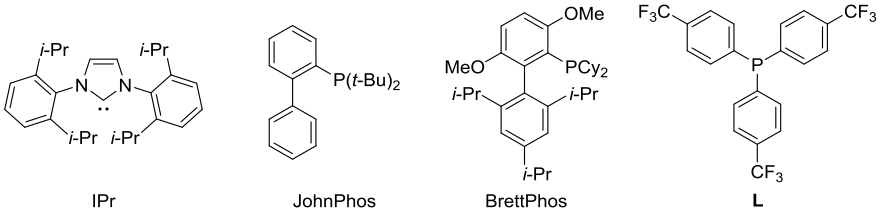
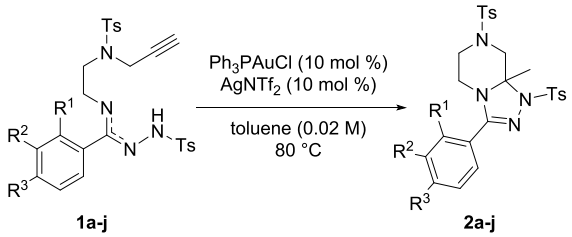


Table 2. Investigation of the substituent effect at the substrate phenyl group.



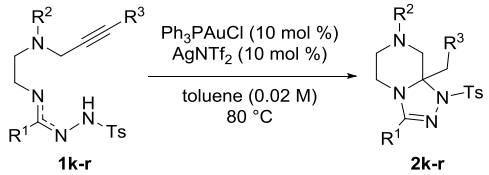
entry	substrate	R^1	R^2	R^3	yield (%)[a]
1	**1a**	H	H	H	74 (78[b]) (**2a**)
2	**1b**	Me	H	H	69 (**2b**)
3	**1c**	OMe	H	H	62 (86[b]) (**2c**)
4	**1d**	NO_2	H	H	69 (**2d**)
5	**1e**	F	H	H	63 (71[b]) (**2e**)
6	**1f**	Cl	H	H	58 (**2f**)
7	**1g**	Br	H	H	58 (**2g**)
8	**1h**	I	H	H	52 (**2h**)
9	**1i**	H	Me	H	78 (**2i**)
10	**1j**	H	H	Me	83 (**2j**)

[a]Isolated yield. [b]**L**AuCl was used instead of Ph_3PAuCl.

of benzamide **8p**, the benzamide **8a** was reacted with NBS and $AgNO_3$ in acetone. The benzamides **8q**–**r** were prepared by treatment of amide **9** with the corresponding substituted propargyl bromide and K_2CO_3 in MeCN. The amidrazones **1p**–**r** were obtained by the same manipulations of **1a**–**m**.

The optimization of the reaction conditions was carried out for the cascade cyclization of amidrazone **1a** (Table 1). The reaction of **1a** with 10 mol % of IPrAuCl/$AgNTf_2$ in 1,2-dichloroethane (DCE) at 50 °C gave [1,2,4]triazolo[4,3-*a*]piperazine **2a** as the isolable isomer, albeit in a low yield of 19% (entry 1). After screening several other phosphine ligands [JohnPhos, BrettPhos, Ph_3P, (t-Bu)$_3$P, and tris(4-trifluoromethylphenyl)phosphine (**L**); entries 2–6], the author found that use of Ph_3PAuCl and **L**AuCl provided **2a** in 64% and 73% yields, respectively (entries 4 and 6). No transformation was observed by treatment with Tf_2NH or $(Ph_3C)B(C_6F_5)_4$ (entries 7 and 8).[13] The higher temperature (80 °C) improved the reaction efficiency (entry 9). Among three silver salts ($AgNTf_2$, AgOTf and $AgSbF_6$) tested for the reactions (entries 9, 11 and 12), use of AgOTf led to a decrease in the yield (38%, entry 11). Screening of the reaction solvent (entries 13–16) revealed that toluene afforded the best result in terms

Table 3. Substrate scopes.



1k-r 2k-r

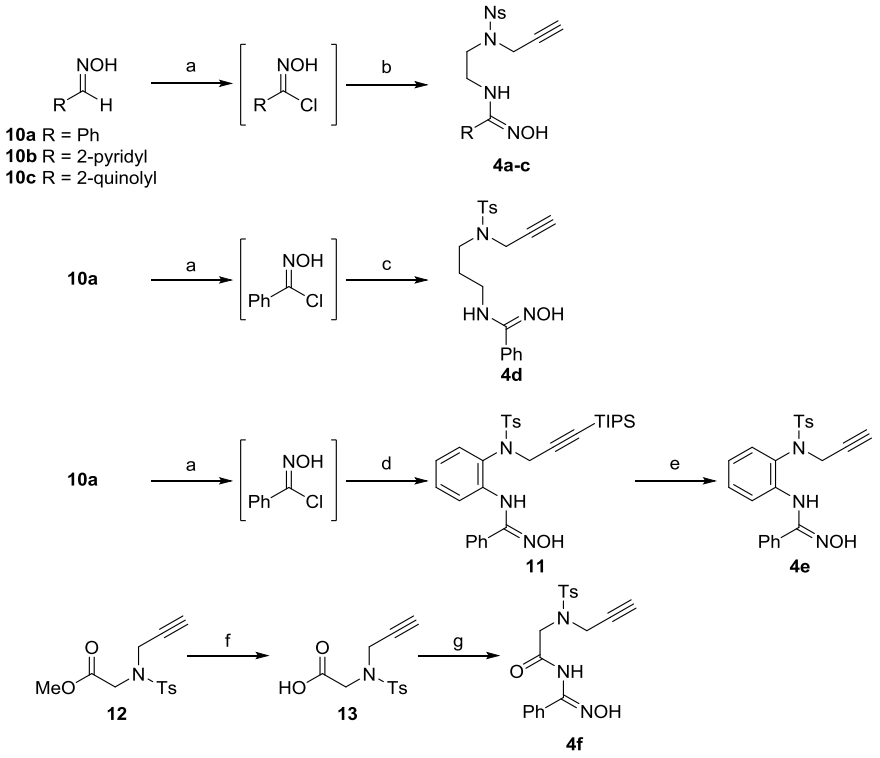
entry	substrate	R^1	R^2	R^3	yield (%)[a]
1	**1k**	cyclohexyl	Ts	H	48 (**2k**)
2	**1l**	*n*-Pr	Ts	H	68 (**2l**)
3	**1m**	2-thienyl	Ts	H	81 (**2m**)
4	**1n**	2-pyridyl	Ts	H	75 (**2n**)
5	**1o**	Ph	Ns	H	65 (68[b]) (**2o**)
6	**1p**	Ph	Ts	Br	30 (**2p**)
7	**1q**	Ph	Ts	Me	0 (**2q**)
8	**1r**	Ph	Ts	Ph	0 (**2r**)

[a]Isolated yield. [b]**L**AuCl was used instead of Ph_3PAuCl.

of the reaction time and yield (entry 16). Decreasing the loading of catalyst to 5 mol % led to a decrease in the yield of **2a** even after a prolonged reaction time (entry 17). The optimal reaction conditions were obtained in the presence of **L**AuCl/$AgNTf_2$ as a catalyst in toluene (entry 18). Use of Ph_3PAuCl alone or [$RuCl_2$(*p*-cymene)]/*n*-Bu_3P proved unsuccessful (entries 19 and 21).[14] The reaction with $AgNTf_2$ gave only 28% yield (entry 20).

Next, the author examined the substituent effect at the phenyl group of amidrazone **1** on the intramolecular cyclizations (Table 2). $Ph_3PAuCl/AgNTf_2$-mediated reaction of amidrazones **1b** and **1c** bearing an electron-donating methyl and methoxy group at the *ortho* position (R^1) proceeded smoothly to give the desired products **2b** and **2c** in 69% and 62% yields, respectively. Amidrazones **1d**–**h** bearing an electron-withdrawing nitro and halogen group at the same position were well tolerated (entries 4–8). Use of **L**AuCl instead of Ph_3PAuCl led to improvement in the product yields (entries 1, 3, and 5). The positions of the substituent in the reaction were investigated by using amidrazones **1i** and **1j** bearing a methyl group at the *meta* position (R^2) or *para* position (R^3). Both substrates reacted more efficiently to give the corresponding triazolopiperazines **2i** and **2j** in 78% and 83% yields, respectively (entries 9 and 10). These results implied that the substituents at the phenyl group were unlikely to significantly affect the cyclization mode and yield.

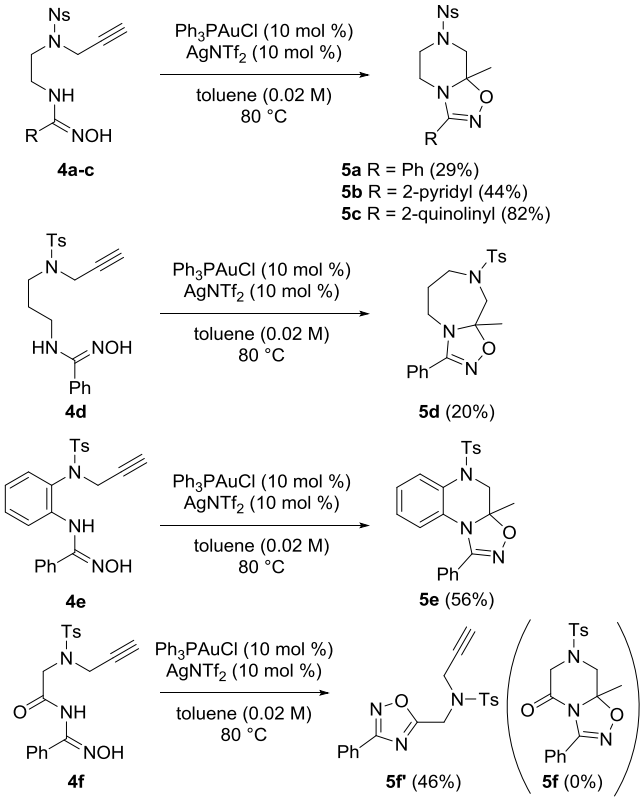
The author further investigated the scope of the reaction using a variety of substrates (Table 3). Amidrazone **1k** bearing a cyclohexyl group at the R^1 position reacted less



Scheme 3. Synthesis of amidoxime substrates **4a**–**f**. *Reagents and conditions*: (a) NCS, DMF, 40 °C; (b) **7b**, Et_3N, CH_2Cl_2, 40 °C; (c) **7c**, Et_3N, CH_2Cl_2, 40 °C; (d) **7d**, Et_3N, CH_2Cl_2, 40 °C; (e) TBAF, THF, 0 °C to rt; (f) $LiOH·H_2O$, THF/H_2O (1:1), rt; (g) *N*-hydroxybenzimidamide, HATU, *i*-Pr_2NEt, CH_2Cl_2, rt.

efficiently to give the corresponding triazole **2k** in 48% yield (entry 1). In contrast, amidrazone **1l** bearing a *n*-propyl group was well tolerated and gave 3-propyltriazole **2l** in 68% yield (entry 2). Thiophene **1m** and pyridine **1n** also reacted smoothly to provide the triazoles **2m** and **2n**, respectively (entries 3 and 4). The reaction of the substrate having an easily removable nosyl (Ns) group at the R^2 position also afforded an *N*-Ns protected piperazine derivative **2o** (entry 5). Amidrazone **1p** incorporating a bromoalkynyl group was also converted to the desired product **2p**, but in a lower yield (entry 6). Unfortunately, the reactions of amidrazone derivatives **1q** and **1r** bearing a methyl and phenyl group at the alkynyl terminus (R^3 position) did not proceed. This is probably due to the lower reactivity of the internal alkyne for the first step or instability of the possible product(s) via 7-*endo-dig* cyclization (entries 7 and 8).

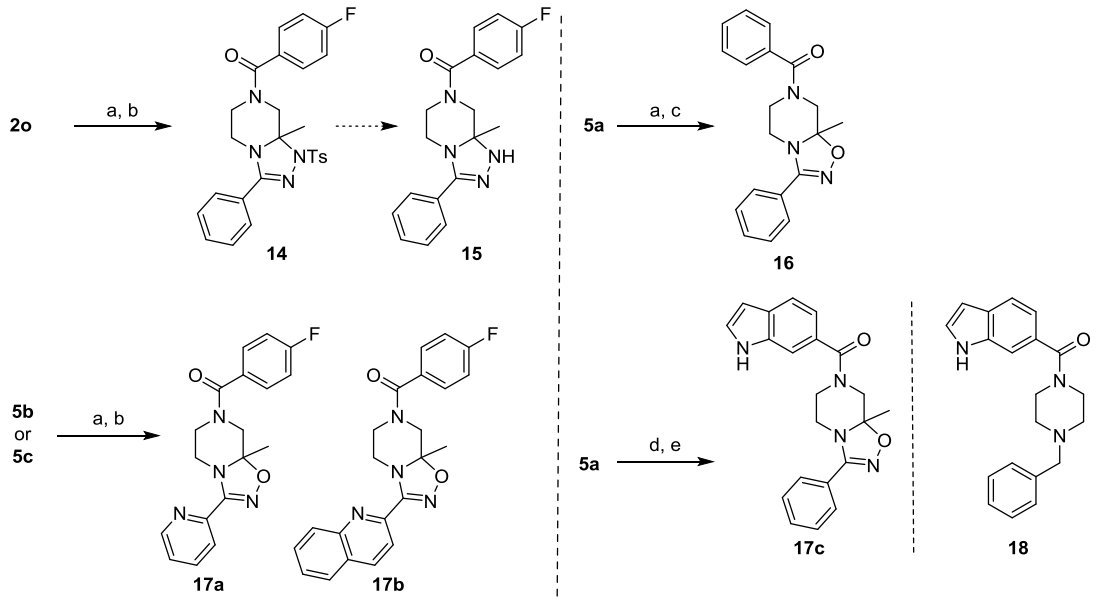
To expand the substrate scope, the author then moved on to investigate the reactions of amidoxime derivatives **4**, in which the oxime OH group was expected to work as the second nucleophilic group of the domino cyclization. The amidoxime substrates **4a**–**e** were prepared from oximes **10a**–**c** and *N*-propargyl sulfonamides **7b**–**d**. The amidoxime



Scheme 4. Reaction scope of amidoximes.

4f was obtained by coupling compound **13** and *N*-hydroxybenzimidamide using HATU and *i*-Pr_2NEt in CH_2Cl_2 (Scheme 3). The reaction under the same conditions using $Ph_3PAuCl/AgNTf_2$ for amidoxime **4a** provided a [1,2,4]oxadiazolo[4,5-*a*]piperazine **5a** in 29% yield (Scheme 4). The low yield was probably due to the decomposition of substrate amidoxime **4a**. Similarly, pyridyl and quinolyl derivatives (**4b** and **4c**) were converted to the oxadiazolopiperazines **5b** and **5c**, respectively. Amidoxime **5d** with a propylene diamine tether was also subjected to the cyclization conditions to provide the [1,2,4]oxadiazolo[4,5-*a*][1,4]diazepine **5d** via 7-*exo*-*dig*/5-*exo*-*trig* cyclizations, although in a lower yield (20%). The *o*-phenylenediamine derivative **4e** reacted smoothly to produce a unique oxadiazole-fused quinoxaline derivative **5e** in 56% yield. These observations imply that this reaction would be applicable to the synthesis of a wide variety of fused triazole and oxadiazole derivatives including piperazines, diazepines and quinoxalines. As an exception, the reaction of imide-derived amidoxime **4f** provided a monocyclic 1,2,4-oxadiazole **5f'** (46%), likely through the nucleophilic addition of the oxime OH group onto the intramolecular amide carbonyl group in **4f** followed by dehydration.[15]

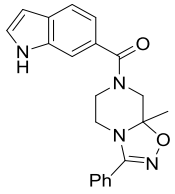
The resulting [1,2,4]triazolo- or [1,2,4]oxadiazolo[4,5-*a*]piperazine scaffold were

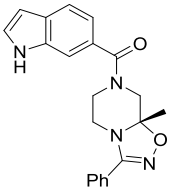


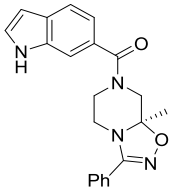
Scheme 5. Synthesis of fused piperazine derivatives and their biological activities. *Reagents and conditions*: (a) PhSH, K_2CO_3, MeCN, rt; (b) 4-fluorobenzoyl chloride, Et_3N, CH_2Cl_2, 0 °C to rt; (c) benzoyl chloride, Et_3N, CH_2Cl_2, rt; (d) SilliaMetS Thiol, Cs_2CO_3, MeCN, 65 °C; (e) indole-6-carboxylic acid, HATU, *i*-Pr_2NEt, DMF, rt.

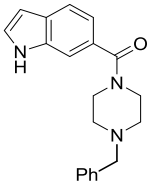
applied to the development of novel bioactive substances (Scheme 5). Initially, the author designed a triazolopiperazine derivative **15** and two oxadiazolopiperazine derivatives **17a** and **17b** of fezolinetant. For preparation of triazolopiperazine **14**, Ns deprotection of **2o** followed by acylation with 4-fluorobenzoyl chloride gave *N*-Ts protected triazolopiperazine **14**. However, Ts deprotection of triazolopiperazine **14** failed to provide triazolopiperazine **15**. Oxadiazolopiperazine derivatives **17a** and **17b** were obtained by the same manipulations of **14**. However, no inhibition of **17a** and **17b** against NK3R activation was observed by monitoring intracellular Ca^{2+} flux (data not shown).[16] Alternatively, to investigate the application of oxadiazolopiperazine scaffold to kinase inhibitor, the author designed and synthesized an oxadiazolopiperazine derivative **16**, which was screened against a panel of 30 kinases. At 10 μM, derivative **16** exhibited moderate inhibition of p38α mitogen activated protein kinase (MAP kinase). Thus, the author designed an oxadiazolopiperazine derivative **17c** that was a ring-constrained analogue of the p38α MAP kinase inhibitor **18**.[17] It was expected that the 1,2,4-oxadiazole substructure of **17c** could restrict the spatial disposition of the key phenyl group in **18**. The author assessed the inhibitory activities against p38 kinase-mediated phosphorylation of a modified erktide by mobility shift assay (Table 4). Compound **17c** showed more potent inhibition against p38α kinase compared with the simple piperazine derivative **18**

Table 4. Inhibitory activities of oxadiazolopiperazine derivatives against p38 MAP kinases.


17c

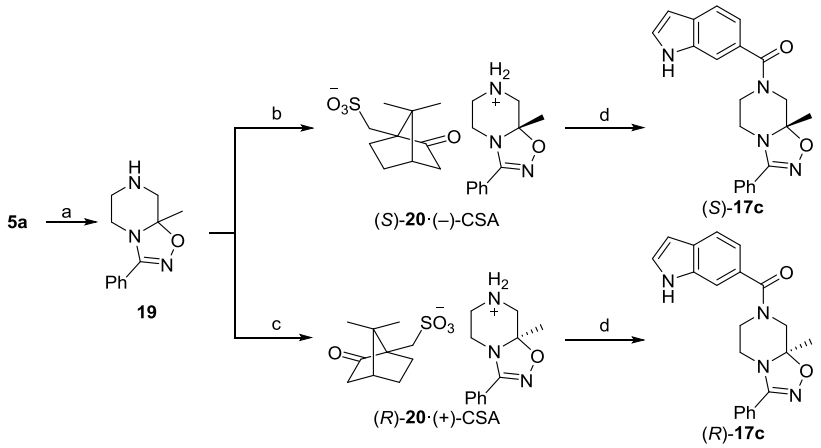

(*S*)-**17c**


(*R*)-**17c**


18

compound	IC_{50} (μM)[a]			
	p38α	p38β	p38γ	p38δ
17c	2.1	5.8	>30	>30
18	8.0	>30	>30	>30
(*S*)-**17c**	1.7	3.5	-[b]	-[b]
(*R*)-**17c**	29	163	-[b]	-[b]

[a]IC_{50} values were determined by p38 MAP kinase assay. [b]Not evaluated.

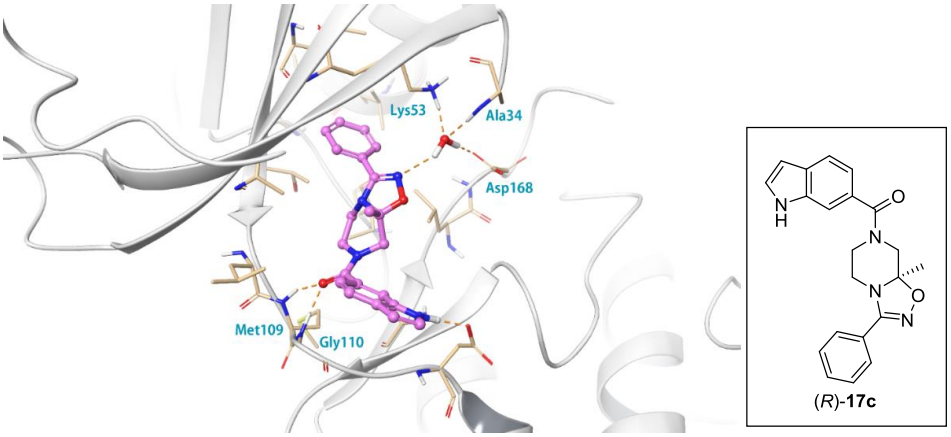


Scheme 6. Synthesis of oxadiazolopiperazine isomers (*R*)-**17c** and (*S*)-**17c**. *Reagents and conditions*: (a) SilliaMetS Thiol, Cs_2CO_3, MeCN, 65 °C; (b) (−)-CSA, recrystalization; (c) (+)-CSA, recrystalization; (d) indole-6-carboxylic acid, WSC·HCl, HOBt, CH_2Cl_2, rt.

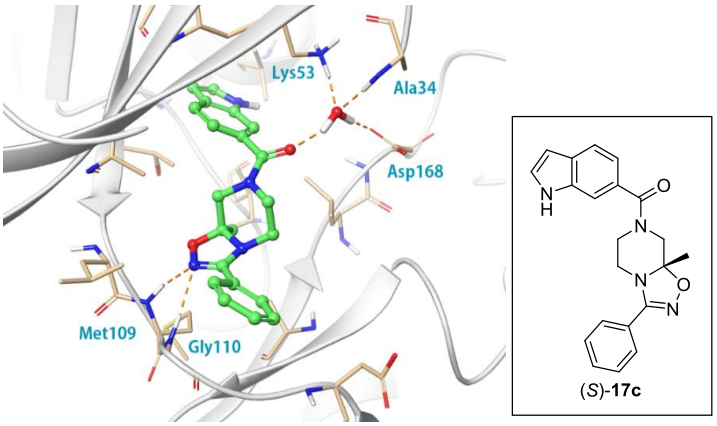
[IC_{50} (**17c**): 2.1 μM, IC_{50} (**18**): 8.0 μM]. Interestingly, oxadiazolopiperazine **17c** also exhibited potent p38β inhibition [IC_{50}(**17c**): 5.8 μM], although no inhibition of piperazine **18** was observed at 30 μM. No inhibitory activities of **17c** and **18** were observed against p38γ and p38δ kinases. Of note, compound **17c** was stable in aqueous buffer under various pH conditions (pH 4.0–10.0).

To identify the isomer of oxadiazolopiperazine **17c** that contributed to the bioactivity,

(A)



(B)



(C)

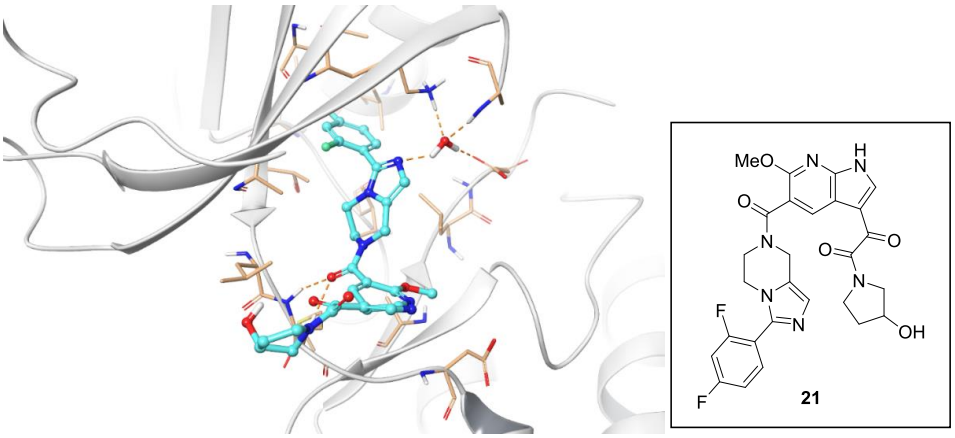


Figure 2 Binding modes of p38α kinase inhibitors. (A) Compound (*R*)-**17c**; (B) Compound (*S*)-**17c**; (C) Compound **21** (PDB ID: 2QD9).

two enantiomers (*R*)-**17c** and (*S*)-**17c** possessing a quaternary carbon stereocenter were synthesized from **5a** in three steps via Ns deprotection, chiral separation using camphorsulfonic acid, and coupling of indole-6-carboxylic acid using WSC·HCl and

HOBt in CH_2Cl_2 (Scheme 6). When these two isomers were independently evaluated for their inhibitory activity against p38α kinase, the *S*-isomer (*S*)-**17c** exhibited 18-fold more potent bioactivity compared with the *R*-isomer [IC_{50} ((*S*)-**17c**): 1.7 μM, IC_{50}((*R*)-**17c**): 29 μM] (Table 5). Similarly, (*S*)-**17c** showed more potent inhibition against p38β kinase [IC_{50}((*S*)-**17c**): 3.5 μM, IC_{50}((*R*)-**17c**): 163 μM].

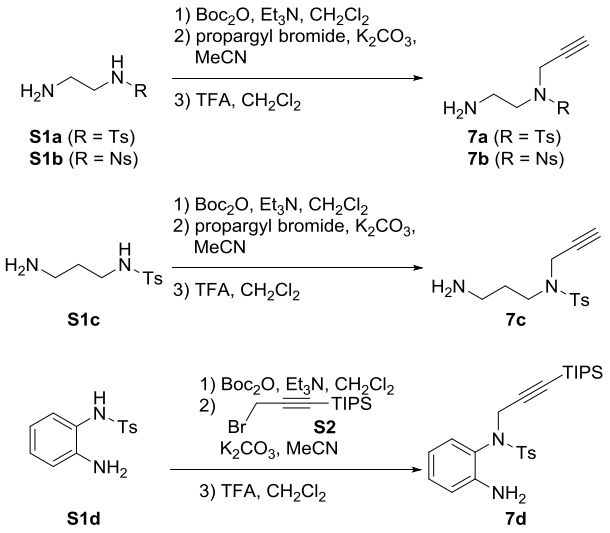
In order to rationalize the higher potency of the *S*-isomer (*S*)-**17c**, the possible binding modes of (*R*)-**17c** and (*S*)-**17c** with p38α kinase were estimated by docking simulation using Glide in Schrödinger Suite 2018-1 (Figure 2).[18] The docking models were obtained by energy minimization of the complex of each inhibitor and p38α kinase using MacroModel[19] with an OPLS3 force field.[20] For simulation, the crystal structure of the p38α kinase complexed with an imidazo[1,5-*a*]piperazine-type inhibitor **21** was employed (PDB ID: 2QD9; for the structure of **21**), because the 3-phenyl-5,6,7,8-tetrahydroimidazo[1,5-*a*]pyrazine substructure in **21** was similar to the 3-phenyl-5,6,8,8a-tetrahydro-7*H*-[1,2,4]oxadiazolo[4,5-*a*]pyrazine part in **17c**. In the crystal structure, a nitrogen atom at the ring junction (position 4 of imidazo[1,5-*a*]piperazine) in **21** interacts with the backbone NH group of Ala34, and the sidechains of Lys53 and Asp168 of p38α kinase through a water molecule (Figure 2C). The binding mode of the less potent (*R*)-**17c** was similar to that of **21**, in which an oxadiazole nitrogen atom interacts with p38α kinase through a water-mediated interaction (Figure 2A). The carbonyl oxygen makes hydrogen bonds with the NH groups of Met109 and Gly110 in the hinge region. Interestingly, the more potent (*S*)-**17c** binds with p38α with an alternative binding mode. The indolylcarbonyl group of (*S*)-**17c** occupied the pocket where the phenyloxadiazole moiety of (*R*)-**17c** was bound while the phenyloxadiazole of (*S*)-**17c** formed an interaction with the hinge region of the p38α kinase (Figure 2B). In this model, the oxadiazole nitrogen works as a hydrogen bond acceptor for the backbone NH groups of Met109 and Gly110. These results suggested that the more potent inhibition of (*S*)-**17c** is likely attributable to the different binding mode to allow the favorable interaction between the [1,2,4]oxadiazolo[4,5-*a*]piperazine scaffold and p38α kinase.

In summary, the author developed a facile synthetic process for triazolopiperazines via a gold-catalyzed cascade cyclization of amidrazones having a terminal alkyne moiety. The author demonstrated that a variety of fused piperazine derivatives can be constructed by this reaction. Additionally, the resulting oxadiazolopiperazine derivative exhibited potent p38α and p38β kinase inhibition. These types of fused piperazines may be applicable as a promising scaffold to design novel kinase inhibitors.

Experimental Section

General methods. ^{1}H NMR spectra were recorded using a JEOL AL-400 or a JEOL ECA-500. Chemical shifts are reported in δ (ppm) relative to Me_4Si as an internal standard. ^{13}C NMR spectra were recorded using a JEOL AL-400 or a JEOL ECA-500 and referenced to the residual solvent signal. Exact mass (HRMS) spectra were recorded on Shimadzu LC-ESI-IT-TOF-MS equipment. IR spectra were obtained on a JASCO FT/IR-4100 spectrometer. Melting points were measured by a hot stage melting points apparatus (uncorrected). For flash chromatography, Wakogel C-300E (Wako Pure Chemical Industries, Ltd) and Chromatorex NH-DM1020 (Fuji Silysia Chemical) were employed. Compounds **S1a**,[21] **S1b**,[22] **S1c**,[23] **S1d**,[24] **S2**,[25] **9**,[22] **10a**,[26] **10b**,[27] **10c**,[28] **12**,[29] and **18**[17] were synthesized according to the procedures in the literatures.

1. Preparation of *N*-propargyl sulfonamides and the derivatives



***N*-(2-Aminoethyl)-4-methyl-*N*-propargylbenzenesulfonamide (7a).**[30] To a solution of *N*-(2-aminoethyl)-4-methylbenzenesulfonamide (**S1a**) (1.07 g, 5.00 mmol) in CH_2Cl_2 (25.0 mL) were added Et_3N (746 μL, 5.50 mmol) and Boc_2O (1.20 g, 5.50 mmol). After being stirred for 45 min at room temperature, this solution was washed with H_2O and dried over $MgSO_4$. The filtrate was concentrated and the residue was dissolved in MeCN (25.0 mL). To the solution were added K_2CO_3 (2.07 g, 15.0 mmol) and propargyl bromide (565 μL, 7.50 mmol), and the mixture was stirred for 1.5 h at 60 °C. The reaction mixture was filtered through a pad of Celite, and the filtrate was concentrated. The residue was dissolved in CH_2Cl_2 (25.0 mL), and TFA (3.83 mL, 50.0 mmol) was added at 0 °C. After being stirred for 1 h at room temperature, this solution was basified by addition of 2 M NaOH aqueous solution. The organic layer was washed with H_2O and dried over Na_2SO_4.

The filtrate was concentrated and the residue was purified by column chromatography ($CHCl_3$/MeOH = 9/1) to give the title compound **7a** (777 mg, 62%): colorless amorphous solid. The ^{1}H NMR spectrum was in good agreement with that reported in literature.[30] ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.30 (br s, 2H), 2.07 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 2.89 (t, *J* = 6.1 Hz, 2H), 3.24 (t, *J* = 6.1 Hz, 2H), 4.15 (d, *J* = 2.3 Hz, 2H), 7.30 (d, *J* = 8.1 Hz, 2H), 7.73 (d, *J* = 8.1 Hz, 2H).

***N*-(2-Aminoethyl)-2-nitro-*N*-propargylbenzenesulfonamide (7b).** By use of a procedure similar to that described for the preparation of the compound **7a** from **S1a**, the compound **S1b** (1.73 g, 7.07 mmol) was converted to the title compound **7b** (818 mg, 41%): yellow oil; IR (neat) 2119 (C≡C), 1539 (NO_2), 1351 (NSO_2), 1161 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.45 (br s, 2H), 2.21 (t, *J* = 2.6 Hz, 1H), 2.94 (t, *J* = 6.3 Hz, 2H), 3.47 (t, *J* = 6.3 Hz, 2H), 4.24 (d, *J* = 2.6 Hz, 2H), 7.64-7.65 (m, 1H), 7.69-7.75 (m, 2H), 8.06-8.08 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 36.9, 39.5, 49.9, 73.9, 76.8, 124.1, 130.9, 131.7, 132.4, 133.8, 148.2; HRMS (ESI) calcd for $C_{11}H_{14}N_3O_4S$ $[M + H]^+$: 284.0700, found: 284.0690.

***N*-(3-Aminopropyl)-4-methyl-*N*-propargylbenzenesulfonamide (7c).** By use of a procedure similar to that described for the preparation of the compound **7a** from **S1a**, the compound **S1c** (1.14 g, 5.00 mmol) was converted to the title compound **7c** (514 mg, 39%): colorless amorphous solid; IR (neat) 2119 (C≡C), 1330 (NSO_2), 1157 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.72-1.77 (m, 2H), 2.04 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 2.80-2.83 (m, 4H), 3.28 (t, *J* = 6.9 Hz, 2H), 4.13 (d, *J* = 2.3 Hz, 2H), 7.29 (d, *J* = 8.6 Hz, 2H), 7.73 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 30.1, 36.1, 38.5, 43.5, 73.8, 76.3, 127.6 (2C), 129.4 (2C), 135.5, 143.5; HRMS (ESI) calcd for $C_{13}H_{19}N_2O_2S$ $[M + H]^+$: 267.1162, found: 267.1169.

***N*-(2-Aminophenyl)-4-methyl-*N*-[3-(triisopropylsilyl)prop-2-yn-1-yl]benzenesulfonamide (7d).** By use of a procedure similar to that described for the preparation of the compound **7a** from **S1a**, the compound **S1d** (952 mg, 3.63 mmol) was converted to the title compound **7d** (812 mg, 49%): colorless amorphous solid; IR (neat) 2174 (C≡C), 1348 (NSO_2), 1161 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 0.93 (s, 21H), 2.43 (s, 3H), 4.18 (br s, 2H), 4.29 (d, *J* = 17.8 Hz, 1H), 4.63 (d, *J* = 17.8 Hz, 1H), 6.48-6.51 (m, 1H), 6.66 (dd, *J* = 8.0, 1.1 Hz, 1H), 6.76 (dd, *J* = 8.0, 1.1 Hz, 1H), 7.07-7.10 (m, 1H), 7.26 (d, *J* = 8.0 Hz, 2H), 7.67 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 11.0 (3C), 18.4 (6C), 21.6, 41.4, 86.7, 101.0, 116.5, 117.6, 124.2, 128.2 (2C), 129.4 (2C), 129.5, 129.7, 135.7, 143.7, 146.3; HRMS (ESI) calcd for $C_{25}H_{37}N_2O_2SSi$ $[M + H]^+$: 457.2340, found: 457.2332.

2. Preparation of benzamides

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]benzamide (8a).** To the solution of benzoic acid (61.0 mg, 0.500 mmol), WSC·HCl (144 mg, 0.750 mmol) and HOBt (101 mg, 0.750 mmol) in CH_2Cl_2 (2.00 mL) was added the compound **7a** (126 mg, 0.500 mmol). After being stirred for 30 min at room temperature, this solution was washed with 1 M HCl, H_2O and dried over $MgSO_4$. The filtrate was concentrated and the residue was purified by column chromatography (*n*-hexane/EtOAc = 2/1) to give the title compound **8a** (90.2 mg, 51%): colorless solid; mp 91-93 °C; IR (neat) 1629 (C=O), 1332 (NSO_2), 1155 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.11 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 3.43-3.45 (m, 2H), 3.66 (td, *J* = 5.3, 5.3 Hz, 2H), 4.20 (d, *J* = 2.3 Hz, 2H), 7.04 (br s, 1H), 7.29 (d, *J* = 8.6 Hz, 2H), 7.42-7.45 (m, 2H), 7.48-7.51 (m, 1H), 7.74 (d, *J* = 8.6 Hz, 2H), 7.85-7.86 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 37.3, 37.8, 45.8, 74.2, 76.6, 127.0 (2C), 127.6 (2C), 128.5 (2C), 129.6 (2C), 131.4, 134.0, 135.2, 144.0, 167.6; *Anal.* calcd for $C_{19}H_{20}N_2O_3S$: C, 64.02; H, 5.66; N, 7.86. Found: C, 63.78; H, 5.61; N, 7.85.

2-Methyl-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8b). By use of a procedure similar to that described for the preparation of the compound **8a**, *o*-toluic acid (68.1 mg, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8b** (161 mg, 87%): colorless solid; mp 129-131 °C; IR (neat) 1657 (C=O), 1342 (NSO_2), 1158 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.08 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 2.48 (s, 3H), 3.42 (t, *J* = 5.4 Hz, 2H), 3.64 (td, *J* = 5.4, 5.4 Hz, 2H), 4.20 (d, *J* = 2.3 Hz, 2H), 6.41 (t, *J* = 5.4 Hz, 1H), 7.20-7.21 (m, 2H), 7.29-7.31 (m, 3H), 7.45-7.47 (m, 1H), 7.72 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.9, 21.5, 37.1, 37.3, 74.3, 76.4, 125.8, 126.9, 127.6 (3C), 129.6 (2C), 129.9, 131.0, 135.2, 135.8, 136.3, 143.9, 170.4; *Anal.* calcd for $C_{20}H_{22}N_2O_3S$: C, 64.84; H, 5.99; N, 7.56. Found: C, 64.73; H, 5.94; N, 7.56.

2-Methoxy-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8c). By use of a procedure similar to that described for the preparation of the compound **8a**, *o*-anisic acid (45.7 mg, 0.300 mmol) and **7a** (90.8 mg, 0.360 mmol) were converted to the title compound **8c** (97.1 mg, 84%): colorless solid; mp 140-142 °C; IR (neat) 1644 (C=O), 1329 (NSO_2), 1165 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.03 (t, *J* = 2.3 Hz, 1H), 2.41 (s, 3H), 3.48 (t, *J* = 5.7 Hz, 2H), 3.67-3.71 (m, 2H), 4.02 (s, 3H), 4.19-4.19 (m, 2H), 6.99 (d, *J* = 8.6 Hz, 1H), 7.05-7.08 (m, 1H), 7.28-7.29 (m, 2H), 7.43-7.47 (m, 1H), 7.73-7.74 (m, 2H), 8.19-8.21 (m, 1H), 8.34-8.36 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 36.7, 37.5, 45.5, 55.8, 74.2, 76.2, 111.2, 121.0, 127.6 (3C), 129.6 (2C), 132.1, 132.9, 135.4, 147.8, 157.8, 165.7; HRMS (ESI) calcd for $C_{20}H_{23}N_2O_4S$ $[M + H]^+$: 387.1373,

found: 387.1382.

2-Nitro-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8d). By use of a procedure similar to that described for the preparation of the compound **8a**, 2-nitrobenzoic acid (50.1 mg, 0.300 mmol) and **7a** (90.8 mg, 0.360 mmol) were converted to the title compound **8d** (87.3 mg, 71%): yellow solid; mp 127-129 °C; IR (neat) 1668 (C=O), 1348 (NSO_2), 1155 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.10 (t, *J* = 2.3 Hz, 1H), 2.43 (s, 3H), 3.42-3.43 (m, 2H), 3.69 (td, *J* = 5.4, 5.4 Hz, 2H), 4.21 (d, *J* = 2.3 Hz, 2H), 6.61 (t, *J* = 5.4 Hz, 1H), 7.31 (d, *J* = 8.0 Hz, 2H), 7.56-7.59 (m, 1H), 7.65-7.70 (m, 2H), 7.72-7.73 (m, 2H), 8.05 (d, *J* = 8.0 Hz, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 37.4, 37.7, 45.9, 74.3, 76.5, 124.5, 127.7 (2C), 128.8, 129.7 (2C), 130.5, 132.6, 133.8, 135.0, 144.1, 146.6, 166.8; HRMS (ESI) calcd for $C_{19}H_{20}N_3O_5S$ $[M + H]^+$: 402.1118; found: 402.1116.

2-Fluoro-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8e). By use of a procedure similar to that described for the preparation of the compound **8a**, 2-fluorobenzoic acid (70.1 mg, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8e** (167 mg, 89%): pale yellow solid; mp 118-119 °C; IR (neat) 1651 (C=O), 1326 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.09 (t, *J* = 2.3 Hz, 1H), 2.40 (s, 3H), 3.46 (t, *J* = 5.5 Hz, 2H), 3.71 (td, *J* = 5.5, 5.5 Hz, 2H), 4.20 (d, *J* = 2.3 Hz, 2H), 7.12-7.15 (m, 2H), 7.24-7.26 (m, 1H), 7.27-7.28 (m, 2H), 7.47-7.48 (m, 1H), 7.73 (d, *J* = 8.0 Hz, 2H), 8.05-8.08 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 37.3, 38.0, 45.9, 74.1, 76.6, 116.1 (d, $J_{C\text{-}F}$ = 24.0 Hz), 124.7 (d, $J_{C\text{-}F}$ = 2.4 Hz), 127.7 (2C), 129.6 (2C), 131.8, 133.3, 133.4, 135.4, 135.5, 143.8, 163.8 (d, $J_{C\text{-}F}$ = 2.4 Hz); *Anal.* calcd for $C_{19}H_{19}FN_2O_3S$: C, 60.95; H, 5.11; N, 7.48. Found: C, 60.75; H, 4.99; N, 7.36.

2-Chloro-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8f). By use of a procedure similar to that described for the preparation of the compound **8a**, 2-chlorobenzoic acid (78.3 mg, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8f** (184 mg, 94%): colorless solid; mp 101-103 °C; IR (neat) 1642 (C=O), 1346 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.08-2.09 (m, 1H), 2.42 (s, 3H), 3.43-3.45 (m, 2H), 3.68 (td, *J* = 5.5, 5.5 Hz, 2H), 4.21 (d, *J* = 2.3 Hz, 2H), 6.71 (br s, 1H), 7.28-7.30 (m, 2H), 7.31-7.33 (m, 1H), 7.34-7.37 (m, 1H), 7.39-7.41 (m, 1H), 7.65-7.66 (m, 1H), 7.72 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 37.2, 37.7, 45.8, 74.3, 76.4, 127.0, 127.6 (2C), 129.6 (2C), 129.8, 130.2, 130.8, 131.2, 134.9, 135.2, 143.9, 167.1; *Anal.* calcd for $C_{19}H_{19}ClN_2O_3S$: C, 58.38; H, 4.90; N, 7.17. Found: C, 58.17; H, 4.99; N, 7.19.

2-Bromo-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8g). By use of a

procedure similar to that described for the preparation of the compound **8a**, 2-bromobenzoic acid (101 mg, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8g** (97.2 mg, 45%): colorless solid; mp 112-114 °C; IR (neat) 1665 (C=O), 1327 (NSO_2), 1153 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.08 (t, *J* = 2.3 Hz, 1H), 2.41 (s, 3H), 3.43 (t, *J* = 5.4 Hz, 2H), 3.66 (td, *J* = 5.4, 5.4 Hz, 2H), 4.21 (d, *J* = 2.3 Hz, 2H), 6.61 (t, *J* = 5.4 Hz, 1H), 7.24-7.30 (m, 3H), 7.32-7.35 (m, 1H), 7.54-7.58 (m, 2H), 7.71 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 37.1, 37.6, 45.7, 74.3, 76.4, 119.4, 127.5, 127.6 (2C), 129.3, 129.6 (2C), 131.1, 132.2, 135.2, 137.5, 143.9, 168.1; *Anal.* calcd for $C_{19}H_{19}BrN_2O_3S$: C, 52.42; H, 4.40; N, 6.44. Found: C, 52.27; H, 4.42; N, 6.44.

2-Iodo-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8h). By use of a procedure similar to that described for the preparation of the compound **8a**, 2-iodobenzoic acid (124 mg, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8h** (227 mg, 94%): colorless solid; mp 131-132 °C; IR (neat) 2118 (C≡C), 1648 (C=O), 1330 (NSO_2), 1157 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.09 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 3.42-3.45 (m, 2H), 3.67 (td, *J* = 5.5, 5.5 Hz, 2H), 4.22 (d, *J* = 2.3 Hz, 2H), 6.40 (br s, 1H), 7.08-7.12 (m, 1H), 7.30 (d, *J* = 8.0 Hz, 2H), 7.38 (dd, *J* = 8.0, 8.0 Hz, 1H), 7.47-7.49 (m, 1H), 7.72 (d, *J* = 8.0 Hz, 2H), 7.86-7.87 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 37.2, 37.6, 45.8, 74.3, 76.4, 92.6, 127.6 (2C), 128.2, 128.3, 129.6 (2C), 131.1, 135.2, 139.8, 141.6, 143.9, 169.7; *Anal.* calcd for $C_{19}H_{19}IN_2O_3S$: C, 47.31; H, 3.97; N, 5.81. Found: C, 47.02; H, 3.99; N, 5.77.

3-Methyl-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8i). By use of a procedure similar to that described for the preparation of the compound **8a**, *m*-toluic acid (68.1 mg, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8i** (204 mg, quant.): colorless solid; mp 132-133 °C; IR (neat) 1637 (C=O), 1327 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.10-2.11 (m, 1H), 2.39 (s, 3H), 2.42 (s, 3H), 3.42-3.45 (m, 2H), 3.66 (td, *J* = 5.3, 5.3 Hz, 2H), 4.20 (d, *J* = 2.3 Hz, 2H), 6.96 (br s, 1H), 7.29-7.33 (m, 4H), 7.61-7.63 (m, 1H), 7.68 (s, 1H), 7.73-7.76 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.3, 21.5, 37.3, 37.8, 45.9, 74.2, 76.6, 123.9, 127.6 (2C), 127.8, 128.4, 129.6 (2C), 132.2, 134.0, 135.2, 138.3, 144.0, 167.9; *Anal.* calcd for $C_{20}H_{22}N_2O_3S$: C, 64.84; H, 5.99; N, 7.56. Found: C, 64.60; H, 6.00; N, 7.52.

4-Methyl-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]benzamide (8j). By use of a procedure similar to that described for the preparation of the compound **8a**, *p*-toluic acid (50.1 mg, 0.300 mmol) and **7a** (90.8 mg, 0.360 mmol) were converted to the title compound **8j** (172 mg, 93%): colorless solid; mp 80-82 °C; IR (neat) 1631 (C=O), 1319

(NSO_2), 1163 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.10 (t, *J* = 2.3 Hz, 1H), 2.39 (s, 3H), 2.42 (s, 3H), 3.43 (t, *J* = 5.4 Hz, 2H), 3.65 (td, *J* = 5.4, 5.4 Hz, 2H), 4.19 (d, *J* = 2.3 Hz, 2H), 6.96 (br s, 1H), 7.23 (d, *J* = 8.0 Hz, 2H), 7.29 (d, *J* = 8.0 Hz, 2H), 7.73-7.75 (m, 4H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.4, 21.5, 37.3, 37.8, 45.8, 74.2, 76.6, 127.0 (2C), 127.6 (2C), 129.2 (2C), 129.6 (2C), 131.2, 135.2, 141.8, 144.0, 167.6; HRMS (ESI) calcd for $C_{20}H_{23}N_2O_3S$ $[M + H]^+$: 371.1424, found: 371.1425.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]cyclohexanecarboxamide (8k).** By use of a procedure similar to that described for the preparation of the compound **8a**, cyclohexanecarboxylic acid (62.0 μL, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8k** (174 mg, 96%): colorless solid; mp 128-129 °C; IR (neat) 1640 (C=O), 1337 (NSO_2), 1156 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.19-1.31 (m, 3H), 1.40-1.48 (m, 2H), 1.66-1.68 (m, 1H), 1.78-1.81 (m, 2H), 1.88-1.91 (m, 2H), 2.08-2.09 (m, 1H), 2.10-2.14 (m, 1H), 2.43 (s, 3H), 3.32 (t, *J* = 5.4 Hz, 2H), 3.45 (td, *J* = 5.4, 5.4 Hz, 2H), 4.16 (d, *J* = 2.3 Hz, 2H), 6.11 (br s, 1H), 7.31 (d, *J* = 8.0 Hz, 2H), 7.72 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 25.7 (3C), 29.5 (2C), 36.8, 37.0, 45.4, 45.9, 74.1, 76.5, 127.6 (2C), 129.6 (2C), 135.3, 143.9, 176.6; *Anal.* calcd for $C_{19}H_{26}N_2O_3S$: C, 62.96; H, 7.23; N, 7.73. Found: C, 62.80; H, 7.23; N, 7.54.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]butyramide (8l).** By use of a procedure similar to that described for the preparation of the compound **8a**, butyric acid (82.6 μL, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8l** (202 mg, quant.): colorless oil; IR (neat) 2120 (C≡C), 1648 (C=O), 1332 (NSO_2), 1158 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 0.96 (t, *J* = 7.4 Hz, 3H), 1.64-1.71 (m, 2H), 2.09 (t, *J* = 2.3 Hz, 1H), 2.19 (t, *J* = 7.4 Hz, 2H), 2.43 (s, 3H), 3.32 (t, *J* = 5.5 Hz, 2H), 3.47 (td, *J* = 5.5, 5.5 Hz, 2H), 4.16 (d, *J* = 2.3 Hz, 2H), 6.17 (br s, 1H), 7.31 (d, *J* = 8.6 Hz, 2H), 7.72 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 13.7, 18.9, 21.5, 37.0, 37.1, 38.5, 45.9, 74.2, 76.4, 127.6 (2C), 129.6 (2C), 135.2, 143.9, 173.6; HRMS (ESI) calcd for $C_{16}H_{23}N_2O_3S$ $[M + H]^+$: 323.1424, found: 323.1416.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]thiophene-2-carboxamide (8m).** By use of a procedure similar to that described for the preparation of the compound **8a**, thiophene-2-carboxylic acid (64.1 mg, 0.500 mmol) and **7a** (151 mg, 0.600 mmol) were converted to the title compound **8m** (202 mg, quant.): colorless solid; mp 122-123 °C; IR (neat) 1624 (C=O), 1327 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.12 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 3.42 (t, *J* = 5.4 Hz, 2H), 3.64 (td, *J* = 5.4, 5.4 Hz, 2H), 4.20 (d, *J* = 2.3 Hz, 2H), 6.96 (br s, 1H), 7.06-7.08 (m, 1H), 7.30 (d, *J* = 8.6 Hz, 2H), 7.47-7.48 (m, 1H), 7.57-7.58 (m, 1H), 7.74 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5,

37.4, 37.8, 45.8, 74.2, 76.6, 127.6 (2C), 127.7, 128.1, 129.7 (2C), 130.1, 135.1, 138.8, 144.0, 162.2; *Anal.* calcd for $C_{17}H_{18}N_2O_3S_2$: C, 56.33; H, 5.01; N, 7.73. Found: C, 56.12; H, 4.96; N, 7.55.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]picolinamide (8n).** By use of a procedure similar to that described for the preparation of the compound **8a**, pyridine-2-carboxylic acid (123 mg, 1.00 mmol) and **7a** (282 mg, 1.00 mmol) were converted to the title compound **8n** (279 mg, 78%): pale yellow solid; mp 118-120 °C; IR (neat) 2117 (C≡C), 1669 (C=O), 1334 (NSO_2), 1158 (NSO_2) cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 2.08-2.09 (m, 1H), 2.38 (s, 3H), 3.46 (t, *J* = 6.0 Hz, 2H), 3.72 (td, *J* = 6.0, 6.0 Hz, 2H), 4.23 (d, *J* = 2.3 Hz, 2H), 7.25 (d, *J* = 8.1 Hz, 2H), 7.41-7.45 (m, 1H), 7.72-7.74 (m, 2H), 7.82-7.86 (m, 1H), 8.15-8.16 (m, 1H), 8.35-8.38 (m, 1H), 8.56-8.57 (m, 1H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 24.5, 37.2, 37.4, 45.9, 74.1, 76.5, 122.0, 126.2, 127.6 (2C), 129.5 (2C), 135.4, 137.2, 143.6, 148.1, 149.5, 164.7; HRMS (ESI) calcd for $C_{18}H_{20}N_3O_3S$ $[M + H]^+$: 358.1220, found: 358.1216.

***N*-{2-[*N*-(2-Nitrobenzenesulfonyl)-*N*-propargylamino]ethyl}benzamide (8o).** By use of a procedure similar to that described for the preparation of the compound **8a**, benzoic acid (73.3 mg, 0.600 mmol) and **7b** (142 mg, 0.500 mmol) were converted to the title compound **8o** (65.9 mg, 34%): pale yellow solid; mp 128-130 °C; IR (neat) 1637 (C=O), 1536 (NO_2), 1351 (NSO_2), 1163 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.25 (t, *J* = 2.3 Hz, 1H), 3.69-3.73 (m, 4H), 4.30 (d, *J* = 2.3 Hz, 2H), 6.69 (br s, 1H), 7.39-7.42 (m, 2H), 7.47-7.50 (m, 1H), 7.53-7.55 (m, 1H), 7.59-7.66 (m, 2H), 7.74 (d, *J* = 7.4 Hz, 2H), 8.04-8.05 (m, 1H); ^{13}C-NMR (125 MHz, CD_3OD) δ 36.2, 36.8, 45.9, 76.7, 77.1, 124.2, 127.1 (2C), 128.2 (2C), 130.2, 131.1, 132.2, 132.4, 134.2, 134.7, 147.6, 166.4; HRMS (ESI) calcd for $C_{18}H_{18}N_3O_5S$ $[M + H]^+$: 388.0962, found: 388.0966.

***N*-{2-[*N*-(3-Bromoprop-2-yn-1-yl)-*N*-tosylamino]ethyl}benzamide (8p).** To a solution of the compound **8a** (1.07 g, 3.00 mmol) in acetone (60.0 mL) was added $AgNO_3$ (255 mg, 1.50 mmol) under Ar and the mixture was stirred for 5 min at room temperature. NBS (561 mg, 3.15 mmol) was added to the mixture at 0 °C. After being stirred for 5 min at 0 °C, this solution was filtrated through a pad of Celite. The filtrate was concentrated and the residue was purified by column chromatography (*n*-hexane/EtOAc = 2/1) to give the bromide **8p** (1.07 mg, 82%): colorless solid; mp 151-153 °C; IR (neat) 1598 (C=O), 1335 (NSO_2), 1160 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.44 (s, 3H), 3.40-3.42 (m, 2H), 3.63-3.66 (m, 2H), 4.21 (s, 2H), 6.92 (br s, 1H), 7.33 (d, *J* = 8.0 Hz, 2H), 7.43-7.46 (m, 2H), 7.49-7.52 (m, 1H), 7.74-7.76 (m, 2H), 7.84-7.86 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.6, 37.8, 38.5, 45.8, 46.0, 72.9, 127.0 (2C), 127.7 (2C), 128.6 (2C), 129.7

(2C), 131.5, 134.0, 135.0, 144.2, 167.7; *Anal.* calcd for $C_{19}H_{19}BrN_2O_3S$: C, 52.42; H, 4.40; N, 6.44. Found: C, 52.18; H, 4.39; N, 6.59.

***N*-{2-[*N*-(But-2-yn-1-yl)-*N*-tosylamino]ethyl}benzamide (8q).** To a solution of the *N*-{2-[(4-methylphenyl)sulfonamide]ethyl}benzamide (**9**) (1.59 g, 5.00 mmol) in MeCN (25.0 mL) were added K_2CO_3 (2.07 g, 15.0 mmol) and the 1-bromo-2-butyne (667 μL, 7.50 mmol). After being stirred for 5 h at 60°C, the reaction mixture was filtered through a pad of Celite. The filtrate was concentrated and the residue was recrystallized from *n*-hexane/$CHCl_3$ to give the compound **8q** (1.78 g, 96%): colorless needles; mp 143-145 °C; IR (neat) 1645 (C=O), 1343 (NSO_2), 1162 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.59 (t, *J* = 2.3 Hz, 3H), 2.43 (s, 3H), 3.40-3.42 (m, 2H), 3.63-3.66 (m, 2H), 4.11-4.13 (m, 2H), 7.00 (br s, 1H), 7.31 (d, *J* = 8.0 Hz, 2H), 7.43-7.46 (m, 2H), 7.48-7.52 (m, 1H), 7.75-7.77 (m, 2H), 7.86-7.87 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 3.3, 21.5, 38.0, 38.1, 45.8, 71.8, 82.2, 127.0 (2C), 127.8 (2C), 128.6 (2C), 129.5 (2C), 131.5, 134.1, 135.4, 143.8, 167.6; HRMS (ESI) calcd for $C_{20}H_{23}N_2O_3S$ $[M + H]^+$: 371.1424, found: 371.1423.

***N*-{2-[*N*-(3-Phenylprop-2-yn-1-yl)-*N*-tosylamino]ethyl}benzamide (8r).** By use of a procedure similar to that described for the preparation of the compound **8q**, the sulfonamide **9** (1.15 g, 3.60 mmol) and the 3-bromo-1-phenylpropyne[31] (1.05 g, 5.40 mmol) were converted to the title compound **8r** (1.44 g, 92%): colorless needles: mp 108-110 °C; IR (neat) 2244 (C≡C), 1644 (C=O), 1344 (NSO_2), 1157 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.31 (s, 3H), 3.50 (t, *J* = 5.4 Hz, 2H), 3.68 (td, *J* = 5.4, 5.4 Hz, 2H), 4.41 (s, 2H), 7.08-7.09 (m, 2H), 7.21-7.29 (m, 6H), 7.37-7.40 (m, 2H), 7.44-7.47 (m, 1H), 7.78 (d, *J* = 8.0 Hz, 2H), 7.85-7.87 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.3, 37.8, 38.0, 45.8, 81.6, 85.7, 121.7, 126.9 (2C), 127.6 (2C), 128.0 (2C), 128.4 (3C), 129.5 (2C), 131.3, 131.4 (2C), 134.0, 135.2, 143.7, 167.6; HRMS (ESI) calcd for $C_{25}H_{25}N_2O_3S$ $[M + H]^+$: 433.1580, found: 433.1579.

3. Preparation of amidrazones

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]-*N'*-tosylbenzohydrazonamide (1a).** To a stirred solution of the compound **8a** (178 mg, 0.500 mmol) and 2-methoxypyridine (67.8 μL, 0.650 mmol) in CH_2Cl_2 (2.00 mL) was added Tf_2O (90.3 μL, 0.550 mmol) at −20 °C under Ar and the mixture was stirred for 10 min at −20 °C. Tosyl hydrazine (140 mg, 0.750 mmol) was added to the mixture. After being stirred for 1.5 h at room temperature, the reaction mixture was quenched with aqueous saturated $NaHCO_3$ and extracted with CH_2Cl_2. The combined organic layer was washed with brine, dried over $MgSO_4$, and concentrated. The residue was purified by column chromatography over NH silica gel (*n*-

hexane/EtOAc = 1/3) to give the title compounds **1Xa** and **1Ya** (157 mg, 60%; 43:57 mixture): colorless amorphous solid; IR (neat) 2119 (C≡C), 1598 (C=N), 1329 (NSO_2), 1157 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xa**, 500 MHz, $CDCl_3$) δ 2.06 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 2.43 (s, 3H), 3.36-3.39 (m, 2H), 3.44-3.46 (m, 2H), 4.05 (d, *J* = 2.3 Hz, 2H), 4.57 (br s, 1H), 7.27-7.31 (m, 5H), 7.37-7.44 (m, 4H), 7.69 (d, *J* = 8.6 Hz, 2H), 7.78 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xa**, 125 MHz, $CDCl_3$) δ 21.5 (2C), 36.9, 39.2, 45.3, 74.1, 76.4, 127.6 (2C), 127.9, 128.2 (2C), 128.5 (2C), 129.2, 129.3 (2C), 129.9, 130.4, 132.0, 135.2, 135.4, 143.5, 143.8, 157.2, 163.4; ^{1}H-NMR (**1Ya**, 500 MHz, $CDCl_3$) δ 1.98 (t, *J* = 2.3 Hz, 1H), 2.41 (s, 3H), 2.41 (s, 3H), 3.17 (t, *J* = 6.0 Hz, 2H), 3.33 (t, *J* = 6.0 Hz, 2H), 3.83 (d, *J* = 2.3 Hz, 2H), 6.39 (br s, 1H), 7.27-7.31 (m, 5H), 7.37-7.44 (m, 4H), 7.66 (d, *J* = 8.0 Hz, 2H), 7.86 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Ya**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 37.3, 42.1, 47.6, 74.2, 76.6, 127.7 (2C), 127.9, 128.3 (2C), 128.6 (2C), 129.3, 129.6 (2C), 130.2, 130.8, 132.0, 135.2, 135.4, 143.5, 143.9, 157.2, 163.4; HRMS (ESI) calcd for $C_{26}H_{29}N_4O_4S_2$ $[M + H]^+$: 525.1625, found: 525.1624.

2-Methyl-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N′*-tosylbenzohydrazonamide (1b). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8b** (55.5 mg, 0.150 mmol) was converted to the title compounds **1Xb** and **1Yb** (70.7 mg, 88%; 61:39 mixture): colorless amorphous solid; IR (neat) 1597 (C=N), 1332 (NSO_2), 1158 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xb**, 500 MHz, $CDCl_3$) δ 2.05-2.06 (m, 1H), 2.12 (s, 3H), 2.42 (s, 3H), 2.44 (s, 3H), 3.13 (br s, 4H), 4.05 (d, *J* = 2.3 Hz, 2H), 4.36 (br s, 1H), 6.91 (d, *J* = 6.9 Hz, 1H), 7.18-7.25 (m, 2H), 7.27-7.34 (m, 5H), 7.68 (d, *J* = 8.6 Hz, 2H), 7.77 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xb**, 125 MHz, $CDCl_3$) δ 18.8, 21.5 (2C), 36.8, 39.1, 45.4, 74.2, 76.4, 124.8, 126.2, 127.6 (2C), 128.1 (2C), 129.3 (2C), 129.6 (2C), 129.9, 130.6, 134.5, 135.2, 136.0, 139.4, 143.6, 144.0, 151.0; ^{1}H-NMR (**1Yb**, 500 MHz, $CDCl_3$) δ 2.02-2.03 (m, 1H), 2.27 (s, 3H), 2.37 (s, 3H), 2.42 (s, 3H), 3.13-3.43 (m, 2H), 3.49 (br s, 2H), 3.92 (d, *J* = 2.3 Hz, 2H), 6.20 (br s, 1H), 7.18-7.25 (m, 3H), 7.27-7.34 (m, 5H), 7.66 (d, *J* = 8.6 Hz, 2H), 7.81 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Yb**, 125 MHz, $CDCl_3$) δ 18.8, 21.6 (2C), 38.1, 42.5, 47.0, 74.4, 76.5, 124.8, 126.5, 127.7 (2C), 128.4 (2C), 129.5 (2C), 129.7 (2C), 130.4, 130.8, 134.9, 135.3, 136.4, 139.4, 143.9, 144.0, 165.5; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_4S_2$ $[M + H]^+$: 539.1781, found: 539.1778.

2-Methoxy-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N′*-tosylbenzohydrazonamide (1c). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8c** (57.9 mg, 0.150 mmol) was converted to the title compounds **1Xc** and **1Yc** (63.8 mg, 77%; 70:30 mixture): colorless amorphous solid; IR

(neat) 1599 (C=N), 1331 (NSO_2), 1160 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xc**, 500 MHz, $CDCl_3$) δ 2.04-2.06 (m, 1H), 2.42 (s, 3H), 2.43 (s, 3H), 3.36 (t, *J* = 5.4 Hz, 2H), 3.45-3.48 (m, 2H), 3.75 (s, 3H), 4.07 (d, *J* = 2.3 Hz, 2H), 4.41 (br s, 1H), 6.91-6.94 (m, 1H), 6.98-7.02 (m, 1H), 7.13-7.15 (m, 1H), 7.28-7.31 (m, 4H), 7.39-7.43 (m, 1H), 7.70 (d, *J* = 8.6 Hz, 2H), 7.80-7.81 (m, 2H); ^{13}C-NMR (**1Xc**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 37.0, 39.6, 45.3, 55.7, 74.0, 76.6, 111.0, 120.9, 121.6, 127.7 (2C), 128.2 (3C), 129.2 (3C), 129.6 (2C), 130.6, 131.9, 135.4, 143.8, 154.7, 155.4; ^{1}H-NMR (**1Yc**, 500 MHz, $CDCl_3$) δ 2.04-2.06 (m, 1H), 2.38 (s, 3H), 2.42 (s, 3H), 3.16-3.24 (m, 4H), 3.84 (s, 3H), 3.99 (d, *J* = 2.3 Hz, 2H), 6.66 (br s, 1H), 6.91-6.94 (m, 1H), 6.98-7.02 (m, 1H), 7.22-7.24 (m, 1H), 7.28-7.31 (m, 4H), 7.39-7.43 (m, 1H), 7.66 (d, *J* = 8.0 Hz, 2H), 7.82 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Yc**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 37.9, 42.3, 47.2, 55.6, 74.1, 76.9, 111.4, 120.5, 121.6, 127.7 (3C), 128.2 (3C), 129.4 (4C), 130.6, 131.6, 135.6, 143.2, 155.4, 156.7; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_5S_2$ $[M + H]^+$: 555.1730, found: 555.1738.

2-Nitro-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N'*-tosylbenzohydrazonamide (1d). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8d** (60.2 mg, 0.150 mmol) was converted to the title compounds **1Xd** and **1Yd** (68.8 mg, 81%; 39:61 mixture): pale yellow amorphous solid; IR (neat) 1530 (C=N), 1346 (NSO_2), 1161 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xd**, 400 MHz, $CDCl_3$) δ 2.07-2.08 (m, 1H), 2.43 (s, 6H), 3.25-3.55 (m, 4H), 4.08-4.13 (m, 2H), 4.83 (br s, 1H), 7.29-7.31 (m, 4H), 7.50-7.52 (m, 1H), 7.57-7.63 (m, 1H), 7.68-7.73 (m, 3H), 7.77 (d, *J* = 8.1 Hz, 2H), 8.02 (d, *J* = 8.1 Hz, 1H); ^{13}C-NMR (**1Xd**, 100 MHz, $CDCl_3$) δ 21.6 (2C), 37.1, 39.8, 45.2, 74.1, 77.2, 124.6 (2C), 127.7 (2C), 128.2 (2C), 129.3 (2C), 129.6 (2C), 130.7, 130.9, 133.5, 134.5, 135.1, 143.7, 144.0, 146.7, 160.9; ^{1}H-NMR (**1Yd**, 400 MHz, $CDCl_3$) δ 2.07-2.08 (m, 1H), 2.40 (s, 3H), 2.43 (s, 3H), 3.25-3.55 (m, 4H), 4.08-4.13 (m, 2H), 6.03 (br s, 1H), 7.29-7.31 (m, 4H), 7.50-7.52 (m, 1H), 7.57-7.63 (m, 1H), 7.68-7.73 (m, 3H), 7.77 (d, *J* = 8.1 Hz, 2H), 8.13 (d, *J* = 8.1 Hz, 1H); ^{13}C-NMR (**1Yd**, 100 MHz, $CDCl_3$) δ 21.6 (2C), 38.2, 43.1, 47.1, 74.3, 77.2, 124.6 (2C), 127.7 (2C), 128.3 (2C), 129.5 (2C), 129.7 (2C), 130.8, 131.1, 134.3, 135.0, 135.2, 143.9, 144.0, 148.2, 161.8; HRMS (ESI) calcd for $C_{26}H_{28}N_5O_6S_2$ $[M + H]^+$: 570.1476, found: 570.1468.

2-Fluoro-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N'*-tosylbenzohydrazonamide (1e). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8e** (56.1 mg, 0.150 mmol) was converted to the title compounds **1Xe** and **1Ye** (12.7 mg, 16%; 47:53 mixture): colorless amorphous solid; IR (neat) 1340 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xe**, 500 MHz, $CDCl_3$) δ 2.06 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 6H), 3.37 (t, *J* = 5.4 Hz, 2H), 3.47-3.50 (m, 2H), 3.92 (d, *J* = 2.3 Hz,

2H), 4.73 (br s, 1H), 7.07-7.13 (m, 1H), 7.16-7.21 (m, 1H), 7.28-7.34 (m, 5H), 7.39-7.46 (m, 1H), 7.70 (d, J = 8.0 Hz, 2H), 7.78 (d, J = 8.0 Hz, 2H); ^{13}C-NMR (**1Xe**, 125 MHz, $CDCl_3$) δ 21.5, 21.6, 37.1, 39.6, 45.2, 74.1, 76.6, 115.8 (d, $J_{C\text{-}F}$ = 21.2 Hz), 124.5 (d, $J_{C\text{-}F}$ = 3.9 Hz), 127.6 (2C), 128.2 (2C), 129.3 (2C), 129.6 (3C), 131.2, 132.0 (d, $J_{C\text{-}F}$ = 7.7 Hz), 134.8, 135.2, 143.5, 143.9, 155.6, 158.1 (d, $J_{C\text{-}F}$ = 132.9 Hz); ^{1}H-NMR (**1Ye**, 500 MHz, $CDCl_3$) δ 2.02 (t, J = 2.3 Hz, 1H), 2.39 (s, 3H), 2.42 (s, 3H), 3.17 (t, J = 6.0 Hz, 2H), 3.26 (t, J = 6.0 Hz, 2H), 3.92 (d, J = 2.3 Hz, 2H), 6.33 (br s, 1H), 7.07-7.13 (m, 1H), 7.16-7.21 (m, 1H), 7.28-7.34 (m, 5H), 7.39-7.46 (m, 1H), 7.67 (d, J = 8.0 Hz, 2H), 7.84 (d, J = 8.0 Hz, 2H); ^{13}C-NMR (**1Ye**, 125 MHz, $CDCl_3$) δ 21.5, 21.6, 37.7, 42.2, 47.3, 74.1, 76.6, 116.1 (d, $J_{C\text{-}F}$ = 21.2 Hz), 125.0 (d, $J_{C\text{-}F}$ = 3.9 Hz), 127.6 (2C), 128.3 (2C), 129.4 (2C), 129.6 (2C), 129.7, 131.2, 132.3 (d, $J_{C\text{-}F}$ = 7.7 Hz), 135.2 (2C), 143.7, 143.9, 160.0, 160.6 (d, $J_{C\text{-}F}$ = 134.9 Hz); HRMS (ESI) calcd for $C_{26}H_{28}FN_4O_4S_2$ $[M + H]^+$: 543.1531, found: 543.1524.

2-Chloro-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N′*-tosylbenzohydrazonamide (1f). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8f** (58.5 mg, 0.150 mmol) was converted to the title compounds **1Xf** and **1Yf** (37.3 mg, 45%; 45:55 mixture): colorless amorphous solid; IR (neat) 2075 (C≡C), 1598 (C=N), 1346 (NSO_2), 1161 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xf**, 500 MHz, $CDCl_3$) δ 2.07 (t, J = 2.0 Hz, 1H), 2.43-2.44 (m, 6H), 3.32-3.55 (m, 4H), 4.00-4.12 (m, 2H), 4.53 (br s, 1H), 7.23-7.24 (m, 1H), 7.27-7.41 (m, 7H), 7.67-7.71 (m, 2H), 7.79-7.80 (m, 2H); ^{13}C-NMR (**1Xf**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 37.0, 39.5, 45.3, 74.2, 76.5, 127.2, 127.7, 128.3 (2C), 129.3 (2C), 129.6 (2C), 129.8 (2C), 130.8, 131.2, 131.5, 132.8, 134.6, 135.2, 143.5, 156.2, 163.6; ^{1}H-NMR (**1Yf**, 500 MHz, $CDCl_3$) δ 2.07 (t, J = 2.0 Hz, 1H), 2.38-2.39 (m, 3H), 2.43-2.44 (m, 3H), 3.20-3.55 (m, 4H), 4.00-4.12 (m, 2H), 6.20 (br s, 1H), 7.27-7.41 (m, 8H), 7.67-7.71 (m, 2H), 7.83-7.84 (m, 2H); ^{13}C-NMR (**1Yf**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 37.9, 42.2, 47.2, 74.2, 76.7, 127.2, 127.7 (2C), 128.3 (2C), 129.4 (2C), 129.7 (2C), 129.8, 129.9, 130.8, 131.2, 131.5, 132.8, 135.3, 143.5, 156.2, 163.6; HRMS (ESI) calcd for $C_{26}H_{28}ClN_4O_4S_2$ $[M + H]^+$: 559.1235, found: 559.1231.

2-Bromo-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N′*-tosylbenzohydrazonamide (1g). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8g** (65.1 mg, 0.150 mmol) was converted to the title compounds **1Xg** and **1Yg** (36.2 mg, 40%; 72:28 mixture): colorless amorphous solid; IR (neat) 1599 (C=N), 1330 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xg**, 500 MHz, $CDCl_3$) δ 2.07 (t, J = 2.6 Hz, 1H), 2.43 (s, 6H), 3.21-3.57 (m, 4H), 4.01-4.05 (m, 2H), 4.46 (br s, 1H), 7.19-7.21 (m, 1H), 7.29-7.32 (m, 5H), 7.36-7.40 (m, 2H), 7.68-7.72 (m, 2H), 7.81

(d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xg**, 125 MHz, $CDCl_3$) δ 21.5, 21.6, 37.0, 39.5, 45.3, 74.2, 76.6, 120.7, 127.7 (2C), 128.2, 128.3 (2C), 129.2 (2C), 129.6 (2C), 130.0, 131.2, 132.9, 133.2, 135.2, 143.5, 143.9, 157.0, 163.2; ^{1}H-NMR (**1Yg**, 500 MHz, $CDCl_3$) δ 2.07 (t, *J* = 2.6 Hz, 1H), 2.38 (s, 3H), 2.43 (s, 3H), 3.21-3.57 (m, 4H), 4.13-4.17 (m, 2H), 6.17 (br s, 1H), 7.29-7.32 (m, 6H), 7.58-7.60 (m, 2H), 7.68-7.72 (m, 2H), 7.85 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Yg**, 125 MHz, $CDCl_3$) δ 21.5, 21.6, 37.9, 42.2, 47.2, 74.2, 76.7, 122.0, 127.7 (2C), 128.2, 128.4 (2C), 129.4 (2C), 129.6 (2C), 130.9, 131.6, 133.0, 133.3, 135.3, 143.8, 143.9, 157.0, 163.2; HRMS (ESI) calcd for $C_{26}H_{28}BrN_4O_4S_2$ $[M + H]^+$: 603.0730, found: 603.0728.

2-Iodo-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N'*-tosylbenzohydrazonamide (1h). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8h** (72.3 mg, 0.150 mmol) was converted to the title compounds **1Xh** and **1Yh** (29.7 mg, 31%; 38:62 mixture): colorless amorphous solid; IR (neat) 1600 (C=N), 1330 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xh**, 500 MHz, $CDCl_3$) δ 2.07-2.08 (m, 1H), 2.38 (s, 3H), 2.43 (s, 3H), 2.97-3.54 (m, 4H), 4.15-4.19 (m, 2H), 4.41 (br s, 1H), 7.09-7.16 (m, 2H), 7.29-7.31 (m, 4H), 7.69-7.72 (m, 2H), 7.81-7.85 (m, 2H), 7.88 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Xh**, 125 MHz, $CDCl_3$) δ 21.5, 21.6, 37.0, 39.4, 45.2, 74.2, 76.5, 94.8, 127.7 (2C), 128.3, 128.4 (2C), 129.2 (2C), 129.5, 129.6 (2C), 131.0, 135.1, 137.2, 139.3, 143.5, 143.9, 158.4, 164.7; ^{1}H-NMR (**1Yh**, 500 MHz, $CDCl_3$) δ 2.07-2.08 (m, 1H), 2.43 (s, 6H), 2.97-3.54 (m, 4H), 4.03-4.07 (m, 2H), 6.13 (br s, 1H), 7.09-7.16 (m, 2H), 7.29-7.31 (m, 4H), 7.39-7.43 (m, 2H), 7.69-7.72 (m, 2H), 7.1-7.85 (m, 2H); ^{13}C-NMR (**1Yh**, 125 MHz, $CDCl_3$) δ 21.5, 21.6, 37.8, 42.2, 47.1, 74.2, 76.6, 96.3, 127.7 (2C), 128.4 (2C), 128.9, 129.2 (2C), 129.6 (2C), 130.4, 131.5, 135.3, 137.4, 139.4, 143.7, 143.9, 158.4, 164.7; HRMS (ESI) calcd for $C_{26}H_{28}IN_4O_4S_2$ $[M + H]^+$: 651.0591, found: 651.0583.

3-Methyl-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N'*-tosylbenzohydrazonamide (1i). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8i** (55.5 mg, 0.150 mmol) was converted to the title compounds **1Xi** and **1Yi** (49.7 mg, 62%; 51:49 mixture): colorless amorphous solid; IR (neat) 1599 (C=N), 1331 (NSO_2), 1160 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xi**, 500 MHz, $CDCl_3$) δ 2.05-2.06 (m, 1H), 2.36 (s, 3H), 2.42 (s, 3H), 2.44 (s, 3H), 3.34-3.39 (m, 2H), 3.44-3.46 (m, 2H), 4.05 (d, *J* = 2.9 Hz, 2H), 4.47 (br s, 1H), 7.01-7.03 (m, 1H), 7.18-7.24 (m, 2H), 7.27-7.32 (m, 5H), 7.69-7.70 (m, 2H), 7.77 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xi**, 125 MHz, $CDCl_3$) δ 21.4, 21.5, 21.6, 36.9, 39.2, 45.3, 74.1, 76.4, 124.8, 127.6 (2C), 128.2 (2C), 128.3, 129.1, 129.3 (2C), 129.6 (2C), 131.1, 135.2 (2C), 138.3, 143.5, 143.8, 156.9, 164.3; ^{1}H-

NMR (**1Yi**, 500 MHz, $CDCl_3$) δ 2.01 (t, *J* = 2.3 Hz, 1H), 2.35 (s, 3H), 2.41 (s, 3H), 2.43 (s, 3H), 3.18 (t, *J* = 6.0 Hz, 2H), 3.34-3.39 (m, 2H), 3.88 (d, *J* = 2.3 Hz, 2H), 6.41 (br s, 1H), 7.01-7.03 (m, 1H), 7.18-7.24 (m, 2H), 7.27-7.32 (m, 5H), 7.67-7.68 (m, 2H), 7.85 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Yi**, 125 MHz, $CDCl_3$) δ 21.4, 21.5, 21.6, 37.4, 42.2, 47.5, 74.2, 76.5, 125.6, 127.7 (2C), 128.3 (2C), 128.4, 129.2, 129.3 (2C), 129.6 (2C), 131.2, 135.2, 135.3, 139.1, 143.6, 143.9, 156.9, 164.3; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_4S_2$ [M + H]$^+$: 539.1781, found: 539.1780.

4-Methyl-*N*-[2-(*N*-propargyl-*N*-tosylamino)ethyl]-*N′*-tosylbenzohydrazonamide (1j). By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8j** (55.5 mg, 0.150 mmol) was converted to the title compounds **1Xj** and **1Yj** (52.7 mg, 65%; 49:51 mixture): colorless amorphous solid; IR (neat) 1599 (C=N), 1331 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xj**, 500 MHz, $CDCl_3$) δ 2.05-2.06 (m, 1H), 2.38 (s, 3H), 2.42 (s, 3H), 2.43 (s, 3H), 3.18 (t, *J* = 5.7 Hz, 2H), 3.34-3.38 (m, 2H), 4.05 (d, *J* = 2.3 Hz, 2H), 4.50 (br s, 1H), 7.15-7.23 (m, 3H), 7.28-7.30 (m, 5H), 7.69 (d, *J* = 8.6 Hz, 2H), 7.77 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xj**, 125 MHz, $CDCl_3$) δ 21.4, 21.6 (2C), 37.3, 42.2, 47.5, 74.2, 76.6, 127.6 (2C), 127.8 (2C), 128.3 (2C), 129.2 (2C), 129.3 (2C), 129.6 (2C), 135.2, 135.4, 140.7, 143.5, 143.9, 157.0, 163.7; ^{1}H-NMR (**1Yj**, 500 MHz, $CDCl_3$) δ 2.00-2.01 (m, 1H), 2.36 (s, 3H), 2.41 (s, 3H), 2.43 (s, 3H), 3.34-3.38 (m, 2H), 3.42-3.43 (m, 2H), 3.86 (d, *J* = 2.3 Hz, 2H), 6.39 (br s, 1H), 7.15-7.23 (m, 3H), 7.28-7.30 (m, 5H), 7.67 (d, *J* = 8.6 Hz, 2H), 7.85 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Yj**, 125 MHz, $CDCl_3$) δ 21.4, 21.5 (2C), 36.9, 39.2, 45.3, 74.1, 76.4, 127.7 (2C), 128.2 (2C), 128.5 (2C), 129.3 (2C), 129.6 (2C), 129.8 (2C), 135.2, 135.3, 140.5, 143.5, 143.8, 157.0, 163.7; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_4S_2$ [M + H]$^+$: 539.1781, found: 539.1778.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]-*N′*-tosylcyclohexanecarbohydrazonamide (1k).** By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8k** (54.3 mg, 0.150 mmol) was converted to the title compounds **1Xk** and **1Yk** (53.9 mg, 68%; 54:46 mixture): colorless amorphous solid; IR (neat) 1635 (C=N), 1331 (NSO_2), 1158 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xk**, 500 MHz, $CDCl_3$) δ 1.19-1.40 (m, 7H), 1.75-1.79 (m, 4H), 2.16 (t, *J* = 2.3 Hz, 1H), 2.42 (s, 3H), 2.43-2.44 (m, 3H), 3.25-3.30 (m, 2H), 3.51 (t, *J* = 6.3 Hz, 2H), 4.12 (d, *J* = 2.3 Hz, 2H), 7.27-7.33 (m, 4H), 7.71-7.74 (m, 2H), 7.77 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xk**, 125 MHz, $CDCl_3$) δ 21.6, 25.6, 25.9 (2C), 29.9, 37.0, 38.1, 38.9, 45.1, 74.0, 76.7, 124.5, 127.6 (2C), 128.2 (2C), 128.7, 129.2 (2C), 129.6 (2C), 134.9, 135.4, 143.4, 143.9, 171.4; ^{1}H-NMR (**1Yk**, 500 MHz, $CDCl_3$) δ 1.19-1.40 (m, 7H), 1.75-1.79 (m, 4H), 2.07-2.08 (m, 1H), 2.41 (s, 3H), 2.43-2.44 (m, 3H), 3.25-3.30 (m, 4H), 4.05 (d, *J* = 2.3 Hz, 2H), 7.27-7.33 (m, 4H),

7.71-7.74 (m, 2H), 7.79 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Yk**, 125 MHz, $CDCl_3$) δ 21.6, 25.8, 25.9 (2C), 30.2, 37.0, 38.6, 41.7, 47.4, 74.4, 76.7, 124.5, 127.7 (2C), 128.4 (2C), 128.7, 129.2 (2C), 129.7 (2C), 135.2, 135.4, 143.4, 144.1, 171.4; HRMS (ESI) calcd for $C_{26}H_{35}N_4O_4S_2$ $[M + H]^+$: 531.2094, found: 531.2084.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]-*N'*-tosylbutyrohydrazonamide (1l).** By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8l** (48.3 mg, 0.150 mmol) was converted to the title compounds **1Xl** and **1Yl** (59.7 mg, 81%; 47:53 mixture): colorless amorphous solid; IR (neat) 1647 (C=N), 1344 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xl**, 500 MHz, $CDCl_3$) δ 0.97 (t, *J* = 7.4 Hz, 3H), 1.55-1.63 (m, 2H), 2.17 (t, *J* = 2.3 Hz, 1H), 2.41 (s, 3H), 2.44 (s, 3H), 2.46-2.47 (m, 2H), 3.25-3.29 (m, 2H), 3.48 (t, *J* = 6.3 Hz, 2H), 4.13-4.13 (m, 2H), 4.85 (br s, 1H), 7.27-7.33 (m, 4H), 7.71-7.74 (m, 2H), 7.78 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xl**, 125 MHz, $CDCl_3$) δ 13.8, 19.8, 21.6 (2C), 32.0, 37.0, 38.9, 45.2, 74.0, 77.2, 127.7 (2C), 128.1 (2C), 129.2 (2C), 129.6 (3C), 135.3, 143.4, 143.9, 168.5; ^{1}H-NMR (**1Yl**, 500 MHz, $CDCl_3$) δ 0.97 (t, *J* = 7.4 Hz, 3H), 1.55-1.63 (m, 2H), 2.07 (t, *J* = 2.3 Hz, 1H), 2.23-2.26 (m, 2H), 2.41 (s, 3H), 2.44 (s, 3H), 3.25-3.29 (m, 4H), 4.06 (d, *J* = 2.3 Hz, 2H), 5.99 (br s, 1H), 7.27-7.33 (m, 4H), 7.71-7.74 (m, 2H), 7.78 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Yl**, 125 MHz, $CDCl_3$) δ 14.0, 20.1, 21.6 (2C), 32.1, 38.4, 41.8, 47.3, 74.4, 77.2, 127.7 (2C), 128.3 (2C), 129.3 (2C), 129.7 (2C), 129.9, 135.4, 143.4, 144.0, 168.5; HRMS (ESI) calcd for $C_{23}H_{31}N_4O_4S_2$ $[M + H]^+$: 491.1781, found: 491.1777.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]-*N'*-tosylthiophene-2-carbohydrazonamide (1m).** By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8m** (54.3 mg, 0.150 mmol) was converted to the title compounds **1Xm** and **1Ym** (23.9 mg, 30%; 52:48 mixture): colorless amorphous solid; IR (neat) 1597 (C=N), 1331 (NSO_2), 1162 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xm**, 500 MHz, $CDCl_3$) δ 2.08 (t, *J* = 2.3 Hz, 1H), 2.43-2.43 (m, 6H), 3.39-3.40 (m, 2H), 3.64 (br s, 2H), 4.08-4.08 (m, 2H), 4.88 (br s, 1H), 7.10-7.12 (m, 1H), 7.29-7.34 (m, 4H), 7.38-7.39 (m, 1H), 7.49-7.50 (m, 1H), 7.70-7.72 (m, 2H), 7.81 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Xm**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 37.1, 39.7, 45.2, 74.1, 76.4, 127.3 (2C), 127.7 (2C), 128.3 (2C), 128.6, 129.3 (2C), 129.5, 129.6 (2C), 134.7, 135.1, 143.7, 143.9, 151.4; ^{1}H-NMR (**1Ym**, 500 MHz, $CDCl_3$) δ 2.08 (t, *J* = 2.3 Hz, 1H), 2.43-2.43 (m, 6H), 3.30 (t, *J* = 6.0 Hz, 2H), 3.39-3.40 (m, 2H), 4.01 (d, *J* = 2.3 Hz, 2H), 6.59 (br s, 1H), 7.05-7.07 (m, 1H), 7.29-7.34 (m, 5H), 7.42-7.43 (m, 1H), 7.70-7.72 (m, 2H), 7.86 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (**1Ym**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 37.4, 42.7, 47.4, 74.4, 76.4, 127.4 (2C), 127.7 (2C), 128.5 (2C), 128.8, 129.3 (2C), 129.7 (2C), 130.0, 135.0, 135.3, 143.9, 144.1,

157.7; HRMS (ESI) calcd for $C_{24}H_{27}N_4O_4S_3$ $[M + H]^+$: 531.1189, found: 531.1187.

***N*-[2-(*N*-Propargyl-*N*-tosylamino)ethyl]-*N'*-tosylpicolinohydrazonamide (1n).** To a stirred solution of the compound **8n** (714 mg, 2.00 mmol) in toluene (20.0 mL) was added PCl_5 (2.08 g, 10.0 mmol) under Ar. After refluxing for 2.5 h, the reaction mixture was cooled to room temperature. Tosyl hydrazine (559 mg, 3.00 mmol) was added and the mixture was stirred for 15 h. After concentration, the residue was purified by column chromatography over NH silica gel (*n*-hexane/EtOAc = 1/2) to give the title compounds **1Xn** and **1Yn** (100 mg, 10%; 47:53 mixture): colorless amorphous solid; IR (neat) 1330 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xn**, 400 MHz, $CDCl_3$) δ 2.07 (t, *J* = 2.3 Hz, 1H), 2.41 (s, 3H), 2.43 (s, 3H), 3.38-3.40 (m, 2H), 3.73 (br s, 2H), 4.08-4.10 (m, 2H), 4.88 (br s, 1H), 7.28-7.33 (m, 4H), 7.36-7.39 (m, 1H), 7.65-7.74 (m, 3H), 7.78-7.90 (m, 3H), 8.63-8.64 (m, 1H), 10.72 (s, 1H); ^{13}C-NMR (**1Xn**, 100 MHz, $CDCl_3$) δ 21.6 (2C), 37.1, 39.5, 45.2, 74.1, 76.5, 122.5, 127.6 (2C), 128.0 (2C), 129.2 (2C), 129.6 (2C), 134.9, 135.3, 136.8, 143.1, 143.9, 147.7, 148.3, 150.7, 155.2; ^{1}H-NMR (**1Yn**, 400 MHz, $CDCl_3$) δ 2.05 (t, *J* = 2.3 Hz, 1H), 2.40 (s, 3H), 2.43 (s, 3H), 3.38-3.40 (m, 4H), 4.08-4.10 (m, 2H), 6.56 (br s, 1H), 7.28-7.33 (m, 4H), 7.36-7.39 (m, 1H), 7.65-7.74 (m, 3H), 7.78-7.90 (m, 3H), 8.53-8.54 (m, 1H), 10.72 (s, 1H); ^{13}C-NMR (**1Yn**, 100 MHz, $CDCl_3$) δ 21.6 (2C), 37.3, 41.8, 47.3, 74.2, 76.6, 124.8, 127.8 (2C), 128.4 (2C), 129.4 (2C), 129.6 (2C), 135.3, 135.9, 138.1, 143.8, 143.9, 148.0, 148.3, 150.4, 155.2; HRMS (ESI) calcd for $C_{25}H_{28}N_5O_4S_2$ $[M + H]^+$: 526.1577, found: 526.1582.

***N*-(2-[*N*-(2-Nitrobenzenesulfonyl)-*N*-propargylamino]ethyl)-*N'*-tosylbenzo-hydrazonamide (1o).** By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8o** (774 mg, 2.00 mmol) was converted to the title compounds **1Xo** and **1Yo** (564 mg, 51%; 48:52 mixture): colorless amorphous solid; IR (neat) 2121 (C≡C), 1598 (C=N), 1542 (NO_2), 1357 (NSO_2), 1159 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xo**, 500 MHz, $CDCl_3$) δ 2.19 (t, *J* = 2.3 Hz, 1H), 2.44 (s, 3H), 3.51 (t, *J* = 5.4 Hz, 2H), 3.62 (t, *J* = 5.7 Hz, 2H), 4.10 (d, *J* = 2.3 Hz, 2H), 4.49 (br s, 1H), 7.28-7.30 (m, 1H), 7.31-7.32 (m, 2H), 7.39-7.46 (m, 4H), 7.60-7.72 (m, 3H), 7.78 (d, *J* = 8.6 Hz, 2H), 8.00 (dd, *J* = 8.0, 1.7 Hz, 1H); ^{13}C-NMR (**1Xo**, 125 MHz, $CDCl_3$) δ 21.6, 36.7, 38.9, 45.9, 74.1, 76.6, 124.2, 127.9 (2C), 128.3 (2C), 128.7 (2C), 129.4 (2C), 130.5, 131.1, 131.9, 132.1, 133.9, 134.9, 143.7, 148.1, 157.3, 164.1; ^{1}H-NMR (**1Yo**, 500 MHz, $CDCl_3$) δ 2.13 (t, *J* = 2.3 Hz, 1H), 2.40 (s, 3H), 3.42-3.47 (m, 4H), 3.99 (d, *J* = 2.3 Hz, 2H), 6.42 (br s, 1H), 7.24-7.26 (m, 2H), 7.28-7.30 (m, 1H), 7.39-7.46 (m, 4H), 7.60-7.72 (m, 3H), 7.83 (d, *J* = 8.0 Hz, 2H), 8.05-8.06 (m, 1H); ^{13}C-NMR (**1Yo**, 125 MHz, $CDCl_3$) δ 21.6, 37.4, 42.1, 47.7, 74.4, 76.9, 124.4, 128.2 (2C), 128.5 (2C), 129.2 (2C), 129.5 (2C), 130.6,

131.2, 131.9, 132.4, 134.0, 135.2, 143.9, 148.2, 157.3, 164.1; HRMS (ESI) calcd for $C_{25}H_{26}N_5O_6S_2$ $[M + H]^+$: 556.1319, found: 556.1312.

***N*-{2-[*N*-(3-Bromoprop-2-yn-1-yl)-*N*-tosylamino]ethyl}-*N'*-tosylbenzohydrazonamide (1p).** By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8p** (434 mg, 1.00 mmol) was converted to the title compounds **1Xp** and **1Yp** (328 mg, 55%; 47:53 mixture): colorless amorphous solid; IR (neat) 1598 (C=N), 1335 (NSO_2), 1160 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xp**, 500 MHz, $CDCl_3$) δ 2.43 (s, 3H), 2.44 (s, 3H), 3.31-3.34 (m, 2H), 3.41-3.44 (m, 2H), 4.04 (s, 2H), 4.48 (br s, 1H), 7.28-7.33 (m, 5H), 7.37-7.45 (m, 4H), 7.68-7.70 (m, 2H), 7.78 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Xp**, 125 MHz, $CDCl_3$) δ 21.5 (2C), 38.0, 39.1, 45.4, 45.5, 72.7, 127.6 (2C), 127.9, 128.2 (2C), 128.5 (2C), 129.2 (2C), 129.3 (3C), 130.3, 134.9, 135.2, 143.6, 144.0, 157.3, 163.3; ^{1}H-NMR (**1Yp**, 500 MHz, $CDCl_3$) δ 2.42 (s, 3H), 2.44 (s, 3H), 3.14 (t, *J* = 5.7 Hz, 2H), 3.31-3.34 (m, 2H), 3.82 (s, 2H), 6.36 (br s, 1H), 7.28-7.33 (m, 5H), 7.37-7.45 (m, 4H), 7.65-7.67 (m, 2H), 7.86 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Yp**, 125 MHz, $CDCl_3$) δ 21.6 (2C), 38.3, 42.0, 45.5, 47.7, 72.9, 127.7 (2C), 127.9, 128.3 (2C), 128.6 (2C), 129.3 (2C), 129.6 (3C), 130.5, 135.1, 135.2, 143.6, 144.0, 157.3, 163.3; HRMS (ESI) calcd for $C_{26}H_{28}BrN_4O_4S_2$ $[M + H]^+$: 603.0730, found: 603.0720.

***N*-{2-[*N*-(But-2-yn-1-yl)-*N*-tosylamino]ethyl}-*N'*-tosylbenzohydrazonamide (1q).** By use of a procedure similar to that described for the preparation of the compound **1a** from **8a**, the compound **8q** (740 mg, 2.00 mmol) was converted to the title compounds **1Xq** and **1Yq** (285 mg, 27%; 41:59 mixture): colorless amorphous solid; IR (neat) 1599 (C=N), 1329 (NSO_2), 1158 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xq**, 500 MHz, $CDCl_3$) δ 1.54-1.55 (m, 3H), 2.41 (s, 3H), 2.43 (s, 3H), 3.30-3.36 (m, 2H), 3.42-3.44 (m, 2H), 3.98 (d, *J* = 2.3 Hz, 2H), 4.63 (br s, 1H), 7.28-7.30 (m, 4H), 7.37-7.45 (m, 5H), 7.70 (d, *J* = 8.0 Hz, 2H), 7.77-7.79 (m, 2H); ^{13}C-NMR (**1Xq**, 125 MHz, $CDCl_3$) δ 3.2, 21.5 (2C), 37.5, 39.3, 45.3, 71.5, 82.0, 127.7 (2C), 127.8 (2C), 128.2 (2C), 128.4 (2C), 129.1 (2C), 129.4 (2C), 130.2, 130.8, 135.1, 135.4, 143.5, 143.6, 157.4; ^{1}H-NMR (**1Yq**, 500 MHz, $CDCl_3$) δ 1.50 (t, *J* = 2.3 Hz, 3H), 2.41 (s, 3H), 2.42 (s, 3H), 3.15 (t, *J* = 5.7 Hz, 2H), 3.30-3.36 (m, 2H), 3.75 (d, *J* = 2.3 Hz, 2H), 6.40 (br s, 1H), 7.28-7.30 (m, 4H), 7.37-7.45 (m, 5H), 7.67 (d, *J* = 8.0 Hz, 2H), 7.86 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Yq**, 125 MHz, $CDCl_3$) δ 3.2, 21.6 (2C), 37.7, 42.1, 47.5, 71.8, 82.0, 127.8 (2C), 127.9 (2C), 128.3 (2C), 128.6 (2C), 129.3 (2C), 129.4 (2C), 130.4, 131.9, 135.2, 135.5, 143.5, 143.6, 163.0; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_4S_2$ $[M + H]^+$: 539.1781, found: 539.1784.

***N*-{2-[*N*-(3-Phenylprop-2-yn-1-yl)-*N*-tosylamino]ethyl}-*N'*-tosylbenzohydrazonamide (1r).** By use of a procedure similar to that described for the preparation of the

compound **1a** from **8a**, the compound **8r** (864 mg, 2.00 mmol) was converted to the title compounds **1Xr** and **1Yr** (837 mg, 70%; 36:64 mixture): colorless amorphous solid; IR (neat) 1599 (C=N), 1328 (NSO_2), 1157 (NSO_2) cm^{-1}; ^{1}H-NMR (**1Xr**, 500 MHz, $CDCl_3$) δ 2.31 (s, 3H), 2.37 (s, 3H), 3.43-3.48 (m, 4H), 4.26 (s, 2H), 4.75 (br s, 1H), 7.06 (d, *J* = 6.9 Hz, 2H), 7.18-7.37 (m, 12H), 7.73 (d, *J* = 8.0 Hz, 2H), 7.78 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Xr**, 125 MHz, $CDCl_3$) δ 21.2 (2C), 37.6, 39.1, 45.3, 81.4, 85.6, 121.6, 127.5 (2C), 127.8 (2C), 128.0 (2C), 128.3 (2C), 128.4, 128.9 (2C), 129.1 (2C), 129.2, 129.4 (2C), 130.1, 131.3 (2C), 135.0, 135.3, 143.1, 143.6, 157.9; ^{1}H-NMR (**1Yr**, 500 MHz, $CDCl_3$) δ 2.28 (s, 3H), 2.35 (s, 3H), 3.23 (t, *J* = 5.4 Hz, 2H), 3.31 (t, *J* = 5.4 Hz, 2H), 3.96 (s, 2H), 6.41 (br s, 1H), 6.97 (d, *J* = 7.4 Hz, 2H), 7.18-7.37 (m, 12H), 7.68-7.70 (m, 2H), 7.88 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (**1Yr**, 125 MHz, $CDCl_3$) δ 21.4 (2C), 37.7, 41.8, 47.4, 81.6, 85.6, 121.7, 127.6 (2C), 127.9 (2C), 128.0 (2C), 128.1 (2C), 128.3 (2C), 128.6 (2C), 129.1 (2C), 129.4 (2C), 130.1, 131.3 (2C), 135.2, 135.5, 143.4, 143.6, 161.8; HRMS (ESI) calcd for $C_{32}H_{33}N_4O_4S_2$ $[M + H]^+$: 601.1938, found: 601.1939.

4. Gold-catalyzed cyclization of amidrazones

8a-Methyl-3-phenyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]-piperazine (2a). To a stirred solution of Ph_3PAuCl (49.5 mg, 0.100 mmol) and $AgNTf_2$ (38.8 mg, 0.100 mmol) in toluene (50.0 mL) was added the compound **1a** (524 mg, 1.00 mmol). After being stirred for 2.5 h at 80 °C, the reaction mixture was concentrated. The residue was purified by column chromatography (*n*-hexane/EtOAc = 2/1) to give the triazolopiperazine **2a** (389 mg, 74%): colorless amorphous solid; IR (neat) 1596 (C=N), 1351 (NSO_2), 1163 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.86 (s, 3H), 2.05-2.10 (m, 1H), 2.42 (s, 3H), 2.45 (s, 3H), 2.61 (d, *J* = 11.5 Hz, 1H), 3.39-3.57 (m, 3H), 4.07-4.09 (m, 1H), 7.27-7.28 (m, 2H), 7.32 (d, *J* = 8.0 Hz, 2H), 7.35-7.46 (m, 5H), 7.57 (d, *J* = 8.0 Hz, 2H), 7.88 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 20.2, 21.6 (2C), 41.1, 45.7, 52.9, 82.4, 125.9, 127.5 (2C), 128.0 (2C), 128.3 (2C), 128.7 (2C), 129.4 (2C), 129.9 (2C), 130.8, 132.5, 136.7, 143.7, 144.2, 153.7; HRMS (ESI) calcd for $C_{26}H_{29}N_4O_4S_2$ $[M + H]^+$: 525.1625, found: 525.1624.

8a-Methyl-3-(*o*-tolyl)-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]-piperazine (2b). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1b** (32.3 mg, 0.0600 mmol) was converted to the title compound **2b** (22.3 mg, 69%): colorless amorphous solid; IR (neat) 1597 (C=N), 1352 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.90 (s, 3H), 1.98-2.04 (m, 1H), 2.12 (s, 3H), 2.44 (s, 3H), 2.47 (s, 3H), 2.57 (d, *J* = 11.5 Hz, 1H), 3.00-3.04 (m, 1H), 3.24-3.30 (m, 1H), 3.50-3.53 (m, 1H), 4.03-4.06 (m, 1H), 7.109-7.11 (m, 1H), 7.16-

7.20 (m, 2H), 7.28-7.33 (m, 5H), 7.55 (d, *J* = 8.6 Hz, 2H), 7.90 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.2, 20.4, 21.6 (2C), 40.4, 45.9, 52.3, 81.9, 125.4, 126.0, 127.5 (2C), 128.1 (2C), 129.3 (3C), 129.9 (2C), 130.5, 130.7, 132.5, 136.8, 137.2, 143.7, 144.1, 153.7; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_4S_2$ $[M + H]^+$: 539.1781, found: 539.1779.

3-(2-Methoxyphenyl)-8a-methyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo-[4,3-*a*]piperazine (2c). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1c** (33.3 mg, 0.0600 mmol) was converted to the title compound **2c** (20.6 mg, 62%): colorless amorphous solid; IR (neat) 1355 (NSO_2), 1167 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.80 (s, 3H), 2.13-2.19 (m, 1H), 2.44 (s, 3H), 2.46 (s, 3H), 2.62 (d, *J* = 10.9 Hz, 1H), 3.01-3.04 (m, 1H), 3.28-3.34 (m, 1H), 3.52-3.54 (m, 1H), 3.63 (s, 3H), 4.09 (d, *J* = 10.9 Hz, 1H), 6.86-6.87 (m, 1H), 6.93-6.96 (m, 1H), 7.25-7.25 (m, 1H), 7.28 (d, *J* = 8.0 Hz, 2H), 7.33 (d, *J* = 8.0 Hz, 2H), 7.38-7.42 (m, 1H), 7.57 (d, *J* = 8.0 Hz, 2H), 7.91 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.2, 21.5, 21.6, 40.6, 45.6, 53.0, 55.4, 82.3, 111.0, 115.0, 120.9, 127.6 (2C), 128.1 (2C), 129.3 (2C), 129.8 (2C), 131.3, 132.3, 132.6, 137.0, 143.4, 144.0, 152.2, 157.4; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_5S_2$ $[M + H]^+$: 555.1730, found: 555.1729.

8a-Methyl-3-(2-nitrophenyl)-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]piperazine (2d). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1d** (34.1 mg, 0.0600 mmol) was converted to the title compound **2d** (24.5 mg, 69%): yellow amorphous solid; IR (neat) 1598 (C=N), 1591 (NO_2), 1351 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.93 (s, 3H), 2.31-2.36 (m, 1H), 2.46 (s, 3H), 2.47 (s, 3H), 2.57 (d, *J* = 11.5 Hz, 1H), 3.04-3.07 (m, 1H), 3.33-3.39 (m, 1H), 3.47-3.51 (m, 1H), 3.97-4.00 (m, 1H), 7.28-7.31 (m, 4H), 7.43-7.45 (m, 1H), 7.49 (d, *J* = 8.6 Hz, 2H), 7.62-7.70 (m, 2H), 7.81-7.83 (m, 2H), 7.98-8.00 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 20.8, 21.6, 21.7, 40.8, 45.0, 52.0, 82.9, 121.5, 124.8, 127.4 (2C), 127.9 (2C), 129.5 (2C), 129.8 (2C), 131.5, 131.7, 132.6, 133.5, 136.9, 143.7, 144.1, 149.0, 150.5; HRMS (ESI) calcd for $C_{26}H_{28}N_5O_6S_2$ $[M + H]^+$: 570.1476, found: 570.1482.

3-(2-Fluoro4phenyl)-8a-methyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo-[4,3-*a*]piperazine (2e). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1e** (16.3 mg, 0.0300 mmol) was converted to the title compound **2e** (10.2 mg, 63%): colorless amorphous solid; IR (neat) 1597 (C=N), 1354 (NSO_2), 1167 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.81 (s, 3H), 2.13-2.19 (m, 1H), 2.43 (s, 3H), 2.46 (s, 3H), 2.64 (d, *J* = 11.5 Hz, 1H), 3.15-3.18 (m, 1H), 3.38-3.44 (m, 1H), 3.59-3.61 (m, 1H), 4.12-4.14 (m, 1H), 7.07-7.11 (m, 1H), 7.16-

7.19 (m, 1H), 7.29 (d, *J* = 8.6 Hz, 2H), 7.33 (d, *J* = 8.6 Hz, 2H), 7.40-7.47 (m, 2H), 7.58 (d, *J* = 8.6 Hz, 2H), 7.89 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.6, 21.6 (2C), 40.9, 45.7, 53.3, 82.6, 116.1 (d, $J_{C\text{-}F}$ = 20.4 Hz), 124.8 (d, $J_{C\text{-}F}$= 2.4 Hz), 127.5 (2C), 128.0 (2C), 129.4 (2C), 129.9 (2C), 131.4, 132.7, 132.9 (d, $J_{C\text{-}F}$ = 8.4 Hz), 136.7 (2C), 143.7, 144.1, 149.6, 159.9 (d, $J_{C\text{-}F}$ = 251.9 Hz); HRMS (ESI) calcd for $C_{26}H_{28}FN_4O_4S_2$ $[M + H]^+$: 543.1531, found: 543.1531.

3-(2-Chlorophenyl)-8a-methyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo-[4,3-*a*]piperazine (2f). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1f** (22.3 mg, 0.0400 mmol) was converted to the title compound **2f** (13.0 mg, 58%): colorless amorphous solid; IR (neat) 1354 (NSO_2), 1167 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.83 (s, 3H), 2.13-2.19 (m, 1H), 2.44 (s, 3H), 2.46 (s, 3H), 2.61 (d, *J* = 10.9 Hz, 1H), 2.97-2.99 (m, 1H), 3.34-3.39 (m, 1H), 3.54-3.56 (m, 1H), 4.06-4.09 (m, 1H), 7.28-7.32 (m, 6H), 7.39-7.40 (m, 2H), 7.54-7.55 (m, 2H), 7.91 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.6 (3C), 40.5, 45.7 (2C), 82.4, 125.4, 127.2, 127.5 (2C), 128.1 (2C), 129.4 (2C), 129.9 (2C), 130.0, 131.5, 132.0, 132.5, 133.7, 136.9, 143.7, 144.1, 151.5; HRMS (ESI) calcd for $C_{26}H_{28}ClN_4O_4S_2$ $[M + H]^+$: 559.1235, found: 559.1231.

3-(2-Bromophenyl)-8a-methyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo-[4,3-*a*]piperazine (2g). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1g** (36.1 mg, 0.0600 mmol) was converted to the title compound **2g** (21.1 mg, 58%): colorless amorphous solid; IR (neat) 1354 (NSO_2), 1166 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.86 (s, 3H), 2.16-2.21 (m, 1H), 2.46 (s, 6H), 2.60-2.62 (m, 1H), 2.94-2.97 (m, 1H), 3.33-3.37 (br m, 1H), 3.52-3.54 (m, 1H), 4.04 (br s, 1H), 7.29-7.36 (m, 7H), 7.52-7.57 (m, 3H), 7.93 (d, *J* = 8.6 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.6 (2C), 21.7, 40.5, 45.7 (2C), 82.5, 122.9, 127.4, 127.6 (2C), 127.7 (2C), 128.2 (2C), 129.4 (2C), 129.8 (2C), 131.5, 132.1, 133.3, 137.0, 143.7, 144.1, 152.6; HRMS (ESI) calcd for $C_{26}H_{28}BrN_4O_4S_2$ $[M + H]^+$: 603.0730, found: 603.0725.

3-(2-Iodophenyl)-8a-methyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]piperazine (2h). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1h** (39.0 mg, 0.0600 mmol) was converted to the title compound **2h** (20.1 mg, 52%): colorless amorphous solid; IR (neat) 1597 (C=N), 1353 (NSO_2), 1165 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.91 (s, 3H), 2.25 (br s, 1H), 2.44 (s, 3H), 2.46 (s, 3H), 2.66 (d, *J* = 11.5 Hz, 1H), 2.91-2.94 (m, 1H), 3.31-3.34 (m, 1H), 3.50 (br s, 1H), 4.00 (br s, 1H), 7.11-7.14 (m, 1H), 7.20-7.22 (m, 1H), 7.28-7.32

(m, 4H), 7.35-7.38 (m, 1H), 7.51 (br s, 2H), 7.80-7.82 (m, 1H), 7.95 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 20.3, 21.5, 21.6, 40.7, 45.8 (2C), 82.6, 97.0, 127.8 (2C), 128.4 (2C), 129.5 (3C), 129.9 (2C), 130.9 (2C), 131.7, 131.9, 137.5, 139.9, 143.6, 144.0, 154.5; HRMS (ESI) calcd for $C_{26}H_{28}IN_4O_4S_2$ $[M + H]^+$: 651.0591, found: 651.0589.

8a-Methyl-3-(*m*-tolyl)-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]-piperazine (2i). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1i** (32.3 mg, 0.0600 mmol) was converted to the title compound **2i** (25.0 mg, 78%): colorless amorphous solid; IR (neat) 1352 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.84 (s, 3H), 2.06-2.11 (m, 1H), 2.33 (s, 3H), 2.41 (s, 3H), 2.45 (s, 3H), 2.63 (d, *J* = 11.5 Hz, 1H), 3.38-3.44 (m, 1H), 3.49-3.57 (m, 2H), 4.07-4.10 (m, 1H), 7.12-7.13 (m, 1H), 7.20 (s, 1H), 7.24-7.28 (m, 4H), 7.32 (d, *J* = 8.0 Hz, 2H), 7.57 (d, *J* = 8.0 Hz, 2H), 7.88 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 20.1, 21.2, 21.6 (2C), 41.1, 45.7, 53.0, 82.2, 125.3, 125.8, 127.5 (2C), 128.0 (2C), 128.5, 128.9, 129.4 (2C), 129.9 (2C), 131.5, 132.6, 136.8, 138.7, 143.6, 144.1, 153.9; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_4S_2$ $[M + H]^+$: 539.1781, found: 539.1784.

8a-Methyl-3-(*p*-tolyl)-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]-piperazine (2j). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1j** (32.3 mg, 0.0600 mmol) was converted to the title compound **2j** (26.7 mg, 83%): colorless amorphous solid; IR (neat) 1595 (C=N), 1351 (NSO_2), 1163 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.83 (s, 3H), 2.04-2.09 (m, 1H), 2.35 (s, 3H), 2.41 (s, 3H), 2.44 (s, 3H), 2.60 (d, *J* = 11.5 Hz, 1H), 3.37-3.43 (m, 1H), 3.48-3.55 (m, 2H), 4.07 (d, *J* = 11.5 Hz, 1H), 7.18 (d, *J* = 8.0 Hz, 2H), 7.24-7.27 (m, 4H), 7.31 (d, *J* = 8.0 Hz, 2H), 7.56 (d, *J* = 8.0 Hz, 2H), 7.88 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 20.1, 21.4, 21.6 (2C), 41.0, 45.6, 53.9, 82.2, 123.0, 127.5 (2C), 128.0 (2C), 128.2 (2C), 129.4 (4C), 129.9 (2C), 132.5, 136.8, 141.1, 143.6, 144.1, 153.8; HRMS (ESI) calcd for $C_{27}H_{31}N_4O_4S_2$ $[M + H]^+$: 539.1781, found: 539.1787.

3-Cyclohexyl-8a-methyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]-piperazine (2k). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1k** (31.8 mg, 0.0600 mmol) was converted to the title compound **2k** (15.2 mg, 48%): colorless amorphous solid; IR (neat) 1598 (C=N), 1351 (NSO_2), 1161 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.16-1.20 (m, 3H), 1.32-1.36 (m, 1H), 1.49-1.52 (m, 1H), 1.68-1.78 (m, 8H), 1.92-1.98 (m, 1H), 2.09-2.14 (m, 1H), 2.35 (d, *J* = 10.9 Hz, 1H), 2.43 (s, 3H), 2.45 (s, 3H), 3.29-3.34 (m, 2H), 3.63-3.66 (m, 1H), 3.94-3.95 (m, 1H), 7.27-7.28 (m, 2H), 7.32 (d, *J* = 8.0 Hz, 2H), 7.56 (d, *J* = 8.0 Hz, 2H), 7.86 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.3, 21.6 (2C), 25.6,

15.7, 15.8, 19.2, 30.5, 34.3, 39.7, 45.7, 52.5, 81.3, 127.5 (2C), 128.0 (2C), 129.2 (2C), 129.9 (2C), 132.6, 136.9, 143.3, 144.1, 157.4; HRMS (ESI) calcd for $C_{26}H_{35}N_4O_4S_2$ [M + H]$^+$: 531.2094, found: 531.2092.

8a-Methyl-3-propyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]piperazine (2l). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1l** (19.6 mg, 0.0400 mmol) was converted to the title compound **2l** (13.2 mg, 68%): colorless amorphous solid; IR (neat) 1598 (C=N), 1350 (NSO_2), 1161 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 0.87 (t, *J* = 7.4 Hz, 3H), 1.45-1.54 (m, 2H), 1.76 (s, 3H), 1.98-2.15 (m, 3H), 2.41 (d, *J* = 11.5 Hz, 1H), 2.43 (s, 3H), 2.45 (s, 3H), 3.32-3.34 (m, 2H), 3.63-3.66 (m, 1H), 3.93-3.96 (m, 1H), 7.28 (d, *J* = 8.0 Hz, 2H), 7.32 (d, *J* = 8.0 Hz, 2H), 7.55 (d, *J* = 8.0 Hz, 2H), 7.85-7.87 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 13.6, 19.4, 19.7, 21.6 (2C), 26.8, 39.9, 45.6, 52.1, 81.6, 127.5 (2C), 128.0 (2C), 129.3 (2C), 129.9 (2C), 132.5, 136.9, 143.5, 144.2, 153.9; HRMS (ESI) calcd for $C_{23}H_{31}N_4O_4S_2$ [M + H]$^+$: 491.1781, found: 491.1778.

8a-Methyl-3-(thiophen-2-yl)-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]piperazine (2m). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1m** (10.6 mg, 0.0200 mmol) was converted to the title compound **2m** (8.54 mg, 81%): colorless amorphous solid; IR (neat) 1597 (C=N), 1351 (NSO_2), 1163 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.81 (s, 3H), 2.16-2.21 (m, 1H), 2.42 (s, 3H), 2.45 (s, 3H), 2.64 (d, *J* = 11.5 Hz, 1H), 3.46-3.48 (m, 1H), 3.61-3.65 (m, 1H), 3.77-3.80 (m, 1H), 4.10-4.12 (m, 1H), 7.05-7.06 (m, 1H), 7.21-7.22 (m, 1H), 7.28 (d, *J* = 8.0 Hz, 2H), 7.32 (d, *J* = 8.0 Hz, 2H), 7.41-7.41 (m, 1H), 7.58 (d, *J* = 8.0 Hz, 2H), 7.88-7.89 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.8, 21.6 (2C), 41.0, 45.6, 53.2, 82.8, 126.7, 127.4, 127.5 (2C), 128.1 (2C), 128.7, 129.1, 129.4 (2C), 130.0 (2C), 132.5, 136.6, 143.8, 144.2, 148.0; HRMS (ESI) calcd for $C_{24}H_{27}N_4O_4S_3$ [M + H]$^+$: 531.1189, found: 531.1187.

8a-Methyl-3-(pyridin-2-yl)-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]piperazine (2n). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1n** (31.5 mg, 0.0600 mmol) was converted to the title compound **2n** (23.7 mg, 75%): colorless amorphous solid; IR (neat) 1352 (NSO_2), 1166 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.87 (s, 3H), 2.22-2.28 (m, 1H), 2.42 (s, 3H), 2.43 (s, 3H), 2.60 (d, *J* = 11.5 Hz, 1H), 3.43-3.50 (m, 1H), 3.62-3.65 (m, 1H), 4.06-4.09 (m, 1H), 4.81-4.85 (m, 1H), 7.27-7.32 (m, 5H), 7.57 (d, *J* = 8.6 Hz, 2H), 7.68-7.72 (m, 1H), 7.87-7.90 (m, 3H), 8.50-8.52 (m, 1H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 19.8, 21.6 (2C), 41.0, 46.1, 52.8, 83.5, 124.5, 124.8, 127.6 (2C), 128.0 (2C), 129.4 (2C), 129.9

(2C), 132.5, 136.7 (2C), 143.8, 144.0, 147.0, 148.4, 150.1; HRMS (ESI) calcd for $C_{25}H_{28}N_5O_4S_2$ $[M + H]^+$: 526.1577, found: 526.1571.

8a-Methyl-7-(2-nitrobenzenesulfonyl)-3-phenyl-1-tosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]piperazine (2o). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1o** (33.3 mg, 0.0600 mmol) was converted to the title compound **2o** (21.6 mg, 65%): pale yellow amorphous solid; IR (neat) 1542 (NO_2), 1356 (NSO_2), 1160 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.76 (s, 3H), 2.41 (s, 3H), 2.72-2.78 (m, 1H), 3.27 (d, *J* = 12.0 Hz, 1H), 3.43-3.48 (m, 1H), 3.53-3.57 (m, 1H), 3.71-3.74 (m, 1H), 4.13-4.16 (m, 1H), 7.27-7.29 (m, 2H), 7.41-7.42 (m, 4H), 7.45-7.48 (m, 1H), 7.65-7.66 (m, 1H), 7.74-7.76 (m, 2H), 7.89 (d, *J* = 8.0 Hz, 2H), 8.03-8.04 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.8, 21.6, 41.4, 45.4, 52.3, 82.2, 124.3, 125.9, 128.1 (2C), 128.4 (3C), 128.8 (2C), 129.4 (2C), 130.9, 131.3, 132.0, 134.1, 136.5, 143.9, 148.1, 153.9; HRMS (ESI) calcd for $C_{25}H_{26}N_5O_6S_2$ $[M + H]^+$: 556.1319, found: 556.1314.

8a-(Bromomethyl)-3-phenyl-1,7-ditosyl-1,5,6,7,8,8a-hexahydro-[1,2,4]triazolo[4,3-*a*]piperazine (2p). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **1p** (241 mg, 0.400 mmol) was converted to the title compound **2p** (72.5 mg, 30%): colorless amorphous solid; IR (neat) 1596 (C=N), 1354 (NSO_2), 1163 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.14-2.20 (m, 1H), 2.40 (s, 3H), 2.46 (s, 3H), 2.97 (d, *J* = 12.0 Hz, 1H), 3.38-3.44 (m, 1H), 3.58-3.63 (m, 2H), 4.19-4.21 (m, 1H), 4.37 (d, *J* = 12.0 Hz, 2H), 7.28 (d, *J* = 8.6 Hz, 2H), 7.34-7.45 (m, 7H), 7.61 (d, *J* = 8.6 Hz, 2H), 7.91-7.93 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.6 (2C), 34.6, 41.0, 45.2, 50.4, 82.7, 125.5, 127.5 (2C), 128.1 (2C), 128.5 (2C), 128.8 (2C), 129.5 (2C), 130.1 (2C), 130.8, 132.4, 136.2, 144.1, 144.5, 153.1; HRMS (ESI) calcd for $C_{26}H_{28}BrN_4O_4S_2$ $[M + H]^+$: 603.0730, found: 603.0724.

5. Preparation of amidoximes

***N'*-Hydroxy-*N*-{2-[*N*-(2-nitrobenzenesulfonyl)-*N*-propargylamino]ethyl}benz-imidamide (4a).** To a stirred solution of the compound **10a** (726 mg, 6.00 mmol) in DMF (30.0 mL) was added *N*-chlorosuccinimide (1.20 g, 9.00 mmol). After being stirred for 3 h at 40 °C, the reaction mixture was extracted with EtOAc. The combined organic layer was washed with H_2O, dried over $MgSO_4$, and concentrated. The residue was dissolved in CH_2Cl_2. To this solution were added the compound **7b** (1.70 g, 6.00 mmol) and Et_3N (3.26 mL, 24.0 mmol). After being stirred for 15 h at 40 °C, the reaction mixture was washed with brine and dried over Na_2SO_4. After concentration, the residue was purified by column chromatography (*n*-hexane/EtOAc = 1/2) to give the amidoxime **4a** (1.62 g,

67%): pale yellow amorphous solid; IR (neat) 1542 (NO_2), 1359 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.12 (t, *J* = 2.3 Hz, 1H), 3.27-3.29 (m, 2H), 3.38 (t, *J* = 6.3 Hz, 2H), 4.07 (d, *J* = 2.3 Hz, 2H), 5.50 (br s, 1H), 7.41-7.44 (m, 5H), 7.55-7.58 (m, 1H), 7.61-7.64 (m, 2H), 7.94-7.96 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 37.3, 41.3, 47.7, 74.1, 76.7, 124.3, 128.3, 128.5, 128.6, 128.9, 129.8, 130.7, 130.9, 131.8, 132.2, 134.0, 148.0, 155.5; HRMS (ESI) calcd for $C_{18}H_{19}N_4O_5S$ $[M + H]^+$: 403.1071, found: 403.1066.

***N'*-Hydroxy-*N*-{2-[*N*-(2-nitrobenzenesulfonyl)-*N*-propargylamino]ethyl}picolin-imidamide (4b).** By use of a procedure similar to that described for the preparation of the compound **4a** from **10a** and **7b**, the compound **10b** (61.0 mg, 0.500 mmol) and **7b** (142 mg, 0.500 mmol) were converted to the title compound **4b** (120 mg, 59%): pale yellow amorphous solid; IR (neat) 2122 (C≡C), 1631 (C=N), 1542 (NO_2), 1357 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.17 (t, *J* = 2.3 Hz, 1H), 3.67-3.74 (m, 4H), 4.26 (d, *J* = 2.3 Hz, 2H), 5.97 (t, *J* = 6.3 Hz, 1H), 7.30-7.33 (m, 1H), 7.55-7.62 (m, 3H), 7.69-7.74 (m, 2H), 8.03-8.05 (m, 1H), 8.55-8.56 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 37.0, 41.5, 48.1, 73.8, 77.1, 122.3, 124.2, 124.3, 130.9, 131.8, 132.6, 133.7, 136.9, 147.9, 148.3, 149.9, 151.0; HRMS (ESI) calcd for $C_{17}H_{18}N_5O_5S$ $[M + H]^+$: 404.1023, found: 404.1017.

***N'*-Hydroxy-*N*-{2-[*N*-(2-nitrobenzenesulfonyl)-*N*-propargylamino]ethyl}quinoline-2-carboximidamide (4c).** By use of a procedure similar to that described for the preparation of the compound **4a** from **10a** and **7b**, the compounds **10c** (172 mg, 1.00 mmol) and **7b** (283 mg, 1.00 mmol) were converted to the title compound **4c** (217 mg, 48%): pale yellow amorphous solid; IR (neat) 1621 (C=N), 1542 (NO_2), 1360 (NSO_2), 1165 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 3.18 (t, *J* = 2.3 Hz, 1H), 3.57 (t, *J* = 6.3 Hz, 2H), 3.72-3.75 (m, 2H), 4.25 (d, *J* = 2.3 Hz, 2H), 6.40-6.43 (m, 1H), 7.62-7.65 (m, 1H), 7.73-7.81 (m, 3H), 7.85-7.88 (m, 2H), 7.98-8.03 (m, 3H), 8.34 (d, *J* = 8.6 Hz, 1H), 10.20 (s, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 36.5, 41.1, 48.2, 76.3, 77.5, 119.3, 124.1, 127.0, 127.6, 127.8, 128.8, 129.9, 130.1, 131.1, 132.3, 134.6, 136.4, 146.1, 147.6, 149.3, 151.1; HRMS (ESI) calcd for $C_{21}H_{20}N_5O_5S$ $[M + H]^+$: 454.1180, found: 454.1181.

***N'*-Hydroxy-*N*-[3-(*N*-tosyl-*N*-propargylamino)propyl]benzimidamide (4d).** By use of a procedure similar to that described for the preparation of the compound **4a** from **10a** and **7b**, the compounds **10a** (90.8 mg, 0.750 mmol) and **7c** (133 mg, 0.500 mmol) were converted to the title compound **4d** (68.5 mg, 36%): colorless amorphous solid; IR (neat) 1626 (C=N), 1342 (NSO_2), 1157 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.54-1.60 (m, 2H), 2.00 (t, *J* = 2.3 Hz, 1H), 2.39 (s, 3H), 3.11-3.12 (m, 2H), 3.16 (t, *J* = 6.6 Hz, 2H), 3.98 (d, *J* = 2.3 Hz, 2H), 5.62 (br s, 1H), 7.23 (d, *J* = 8.0 Hz, 2H), 7.37-7.44 (m, 3H),

7.46-7.48 (m, 2H), 7.68-7.69 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.4, 28.9, 36.3, 40.2, 43.3, 73.9, 76.3, 127.6 (2C), 128.4 (4C), 129.4 (2C), 129.5, 131.3, 135.2, 143.5, 156.2; HRMS (ESI) calcd for $C_{20}H_{24}N_3O_3S$ $[M + H]^+$: 386.1533, found: 386.1534.

***N'*-Hydroxy-*N*-(2-{*N*-tosyl-*N*-[3-(triisopropylsilyl)prop-2-yn-1-yl]amino}phenyl)-benzimidamide (11).** By use of a procedure similar to that described for the preparation of the compound **4a** from **10a** and **7b**, the compounds **10a** (18.2 mg, 0.150 mmol) and **7d** (45.6 mg, 0.100 mmol) were converted to the title compound **11** (22.4 mg, 39%): colorless amorphous solid; IR (neat) 2357 (C≡C), 1624 (C=N), 1353 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 0.95 (s, 21H), 2.41 (s, 3H), 4.37 (br s, 1H), 4.82 (br s, 1H), 6.34-6.36 (m, 1H), 6.68-6.71 (m, 1H), 6.88-6.93 (m, 2H), 7.25-7.27 (m, 3H), 7.31-7.34 (m, 2H), 7.37-7.40 (m, 1H), 7.44-7.45 (m, 2H), 7.70 (d, *J* = 8.0 Hz, 2H), 8.00 (s, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 11.1 (3C), 18.4 (6C), 21.6, 41.6, 87.4, 100.7, 121.6, 122.1, 128.2 (2C), 128.3 (2C), 128.5 (2C), 128.7 (2C), 129.5 (2C), 129.7, 130.0, 131.3, 135.4, 139.5, 144.0, 150.8; HRMS (ESI) calcd for $C_{32}H_{42}N_3O_3SSi$ $[M + H]^+$: 576.2711; found: 576.2717.

***N'*-Hydroxy-*N*-[2-(*N*-tosyl-*N*-propargylamino)phenyl]benzimidamide (4e).** To a stirred solution of the compound **11** (13.5 mg, 0.0235 mmol) in THF (235 μL) was added TBAF in THF (1 M, 35.3 μL, 0.0353 mmol) at 0 °C. After being stirred for 1 h at room temperature, the reaction mixture was quenched by H_2O and extracted with EtOAc. The combined organic layer was dried over $MgSO_4$, and concentrated. The residue was purified by column chromatography (*n*-hexane/EtOAc = 2/1) to give the alkyne **4e** (4.32 mg, 44%): pale yellow amorphous solid; IR (neat) 1624 (C=N), 1348 (NSO_2), 1162 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.25-2.26 (m, 1H), 2.43 (s, 3H), 4.48 (d, *J* = 2.3 Hz, 2H), 6.37-6.39 (m, 1H), 6.74-6.75 (m, 2H), 6.90-6.94 (m, 1H), 7.28-7.32 (m, 4H), 7.37-7.38 (m, 1H), 7.46-7.47 (m, 2H), 7.67 (d, *J* = 8.6 Hz, 2H), 8.11 (s, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.6, 41.3, 74.0, 77.8, 121.9, 122.6, 128.2 (2C), 128.3 (2C), 128.5 (2C), 128.6, 128.8, 129.1, 129.5 (2C), 129.7, 131.2, 134.8, 139.9, 144.3, 151.1; HRMS (ESI) calcd for $C_{23}H_{22}N_3O_3S$ $[M + H]^+$: 420.1376, found: 420.1368.

***N*-(Prop-2-yn-1-yl)-*N*-tosylglycine (13).** To a stirred solution of the compound **12** (843 mg, 3.00 mmol) in THF (9.00 mL) was added a solution of $LiOH·H_2O$ (818 mg, 19.5 mmol) in H_2O (9.00 mL). After being stirred for 1 h at room temperature, the reaction mixture was extracted with EtOAc and washed with H_2O. The aqueous layer was acidified by 1 M HCl at 0 °C and extracted with EtOAc. The organic layer was washed with H_2O, dried over $MgSO_4$ and concentrated to give the title compound **13** (758 mg, 95%): colorless amorphous solid; mp 97-99 °C; IR (neat) 1728 (C=O), 1332 (NSO_2),

1156 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.17 (t, *J* = 2.3 Hz, 1H), 2.43 (s, 3H), 4.15 (s, 2H), 4.25 (d, *J* = 2.3 Hz, 2H), 7.32 (d, *J* = 8.6 Hz, 2H), 7.73 (d, *J* = 8.6 Hz, 2H), 10.68 (br s, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 37.4, 46.5, 74.7, 76.0, 127.5 (2C), 129.7 (2C), 135.6, 144.1, 174.4; HRMS (ESI) calcd for $C_{12}H_{14}NO_4S$ $[M + H]^+$: 268.0638, found: 268.0634.

***N*-[(Hydroxyimino)(phenyl)methyl]-2-[*N*-(prop-2-yn-1-yl)-*N*-tosylamino]acetamide (4f).** To a stirred solution of the compound **13** (53.4 mg, 0.20 mmol), HATU (76.1 mg, 0.200 mmol) and *i*-Pr_2NEt (69.7 μL, 0.400 mmol) in CH_2Cl_2 (1.00 mL) was added the *N*-hydroxybenzimidamide[31] (27.2 mg, 0.200 mmol). After being stirred for 2 h at room temperature, the reaction mixture was washed with 1 M HCl and H_2O. The combined organic layer was dried over $MgSO_4$, and concentrated. The residue was purified by column chromatography (*n*-hexane/EtOAc = 2/1) to give the title compound **4f** (49.3 mg, 64%): colorless amorphous solid; IR (neat) 1759 (C=O), 1635 (C=N), 1335 (NSO_2), 1158 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.20-2.21 (m, 1H), 2.44 (s, 3H), 4.25 (d, *J* = 2.3 Hz, 2H), 4.33 (s, 2H), 5.38 (br s, 1H), 7.33 (d, *J* = 8.0 Hz, 2H), 7.42-7.45 (m, 2H), 7.49-7.52 (m, 1H), 7.69-7.71 (m, 2H), 7.77-7.78 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.6, 37.7, 47.1, 74.5, 76.3, 126.8, 127.6 (3C), 128.8 (2C), 129.7, 129.8 (2C), 130.8, 131.2, 135.5, 144.2, 157.7; HRMS (ESI) calcd for $C_{19}H_{20}N_3O_4S$ $[M + H]^+$: 386.1169, found: 386.1175.

6. Gold-catalyzed cyclization of amidoximes

8a-Methyl-7-[(2-nitrophenyl)sulfonyl]-3-phenyl-6,7,8,8a-tetrahydro-5*H*-[1,2,4]oxadiazolo[4,5-*a*]piperazine (5a). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **4a** (402 mg, 1.00 mmol) was converted to the title compound **5a** (116 mg, 29%): colorless amorphous solid; IR (neat) 1542 (NO_2), 1372 (NSO_2), 1167 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.78 (s, 3H), 2.68-2.74 (m, 1H), 3.20 (d, *J* = 12.6 Hz, 1H), 3.39-3.45 (m, 1H), 3.54-3.58 (m, 2H), 3.61-3.64 (m, 1H), 7.44-7.51 (m, 5H), 7.63-7.65 (m, 1H), 7.70-7.76 (m, 2H), 7.95-7.97 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.0, 41.6, 45.2, 49.2, 95.4, 124.2, 124.4, 127.9 (2C), 129.0 (2C), 130.8, 130.9, 131.0, 131.8, 134.1, 148.2, 157.9; HRMS (ESI) calcd for $C_{18}H_{19}N_4O_5S$ $[M+ H]^+$: 403.1071, found: 403.1064.

8a-Methyl-7-[(2-nitrophenyl)sulfonyl]-3-(pyridin-2-yl)-6,7,8,8a-tetrahydro-5*H*-[1,2,4]oxadiazolo[4,5-*a*]piperazine (5b). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **4b** (24.2 mg, 0.0600 mmol) was converted to the title compound **5b** (10.7 mg, 44%): colorless amorphous solid; IR (neat) 1544 (NO_2), 1357 (NSO_2), 1200 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz,

$CDCl_3$) δ 1.82 (s, 3H), 2.90-2.95 (m, 1H), 3.18 (d, *J* = 12.0 Hz, 1H), 3.43-3.49 (m, 1H), 3.56-3.58 (m, 1H), 3.73-3.76 (m, 1H), 4.78-4.82 (m, 1H), 7.35-7.38 (m, 1H), 7.61-7.63 (m, 1H), 7.68-7.76 (m, 3H), 7.81-7.84 (m, 1H), 7.94-7.96 (m, 1H), 8.62-8.63 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 20.5, 41.8, 45.9, 49.3, 96.4, 124.1, 124.2 (2C), 125.1, 131.1, 131.7, 134.0 (2C), 136.9, 145.6, 149.1, 154.8; HRMS (ESI) calcd for $C_{17}H_{18}N_5O_5S$ $[M + H]^+$: 404.1023, found: 404.1016.

8a-Methyl-7-[(2-nitrophenyl)sulfonyl]-3-(quinolin-2-yl)-6,7,8,8a-tetrahydro-5*H*-[1,2,4]oxadiazolo[4,5-*a*]piperazine (5c). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **4c** (27.2 mg, 0.0600 mmol) was converted to the title compound **5c** (22.2 mg, 82%): colorless amorphous solid; IR (neat) 1545 (NO_2), 1362 (NSO_2), 1169 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.86 (s, 3H), 2.92-2.98 (m, 1H), 3.20 (d, *J* = 13.2 Hz, 1H), 3.53-3.62 (m, 2H), 3.77-3.80 (m, 1H), 5.29-5.33 (m, 1H), 7.60-7.63 (m, 2H), 7.67-7.73 (m, 2H), 7.75-7.78 (m, 1H), 7.83-7.85 (m, 1H), 7.92 (d, *J* = 8.6 Hz, 1H), 7.94-7.96 (m, 1H), 8.08 (d, *J* = 8.6 Hz, 1H), 8.18 (d, *J* = 8.6 Hz, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 20.6, 42.0, 46.0, 49.4, 96.7, 120.7 (2C), 124.2, 127.7, 127.9, 128.2, 129.6, 130.2, 130.9, 131.1, 131.7, 134.0, 136.7 (2C), 145.8, 154.6; HRMS (ESI) calcd for $C_{21}H_{20}N_5O_5S$ $[M + H]^+$: 454.1180, found: 454.1173.

9a-Methyl-3-phenyl-7-tosyl-5,6,7,8,9,9a-hexahydro-[1,2,4]oxadiazolo[4,5-*d*][1,4]diazepine (5d). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **4d** (28.1 mg, 0.0729 mmol) was converted to the title compound **5d** (5.49 mg, 20%): colorless amorphous solid; IR (neat) 1338 (NSO_2), 1162 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.49-1.50 (m, 1H), 1.62 (s, 3H), 1.64-1.67 (m, 1H), 2.45 (s, 3H), 3.05-3.10 (m, 1H), 3.20-3.28 (m, 2H), 3.35-3.38 (m, 1H), 3.48-3.53 (m, 1H), 3.75-3.78 (m, 1H), 7.34 (d, *J* = 8.0 Hz, 2H), 7.39-7.47 (m, 5H), 7.74-7.77 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.5, 23.6, 29.4, 42.0, 49.5, 55.2, 99.9, 127.0, 127.5 (2C), 127.6, 128.2, 128.6, 128.8 (2C), 129.6, 129.7 (2C), 130.5, 143.6; HRMS (ESI) calcd for $C_{20}H_{24}N_3O_3S$ $[M + H]^+$: 386.1533, found: 386.1532.

3a-Methyl-1-phenyl-5-tosyl-3a,4-dihydro-5*H*-[1,2,4]oxadiazolo[4,5-*a*]quinoxaline (5e). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **4e** (4.32 mg, 0.0103 mmol) was converted to the title compound **5e** (2.41 mg, 56%): colorless amorphous solid; IR (neat) 1492 (C=N), 1354 (NSO_2), 1166 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.56 (s, 3H), 2.42 (s, 3H), 4.15 (br s, 2H), 6.60-6.62 (m, 1H), 6.80-6.84 (m, 1H), 6.97-7.01 (m, 1H), 7.20-7.22 (m, 2H), 7.26-7.28 (m, 2H), 7.34-7.38 (m, 2H), 7.44-7.47 (m, 1H), 7.68-7.72 (m, 3H); ^{13}C-NMR

(125 MHz, $CDCl_3$) δ 21.5, 23.0, 51.9, 98.1, 122.3, 123.4, 124.2, 125.4, 125.8, 127.5 (2C), 128.5 (2C), 129.1 (2C), 129.4, 129.6 (2C), 130.9, 131.5, 136.7, 143.9, 155.6; HRMS (ESI) calcd for $C_{23}H_{22}N_3O_3S$ $[M + H]^+$: 420.1376, found: 420.1378.

5-[*N*-(Prop-2-yn-1-yl)-*N*-tosylaminomethyl]-3-phenyl-1,2,4-oxadiazole (5f'). By use of a procedure similar to that described for the preparation of the compound **2a** from **1a**, the compound **4f** (23.1 mg, 0.0600 mmol) was converted to the title compound **5f'** (10.0 mg, 46%): colorless solid; mp 120-122 °C; IR (neat) 1352 (NSO_2), 1164 (NSO_2) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.20-2.21 (m, 1H), 2.31 (s, 3H), 4.40 (d, *J* = 2.3 Hz, 2H), 4.84 (s, 2H), 7.25-7.26 (m, 2H), 7.45-7.54 (m, 3H), 7.72 (d, *J* = 8.0 Hz, 2H), 7.95-7.97 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 21.4, 37.6, 41.5, 74.9, 75.9, 126.2, 127.4 (2C), 127.5 (2C), 128.8, 129.7 (2C), 131.4, 135.3, 144.3, 168.3, 174.1 (2C); HRMS (ESI) calcd for $C_{19}H_{18}N_3O_3S$ $[M + H]^+$: 368.1063, found: 368.1061.

7. Preparation of potential bioactive substances

(4-Fluorophenyl)(8a-methyl-3-phenyl-1-tosyl-5,6,8,8a-tetrahydro-[1,2,4]triazolo-[4,3-*a*]pyrazin-7(1*H*)-yl)methanone (14). To a stirred solution of the compound **2o** (335 mg, 0.603 mmol) in MeCN (6.03 mL) were added K_2CO_3 (250 mg, 1.81 mmol) and PhSH (154 μL, 1.51 mmol). After being stirred for 2 h at room temperature, the reaction mixture was filtered through a short pad of silica gel. After the silica gel was washed with *n*-hexane/EtOAc (5/1), the amine was eluted with $CHCl_3$/MeOH (9/1). After the concentration, the residue was dissolved in CH_2Cl_2 (12.0 mL). To the solution was added 4-fluorobenzoyl chloride (108 μL, 0.905 mmol) and Et_3N (164 μL, 1.21 mmol) at 0 ºC. After being stirred for 30 min at room temperature, the reaction mixture was concentrated and purified by column chromatography (*n*-hexane/EtOAc = 1/1) to give the title compound **14** (258 mg, 87%): colorless amorphous solid; IR (neat) 1638 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.59 (s, 3H), 2.38 (s, 3H), 2.99 (s, 2H), 3.00-3.06 (m, 1H), 3.60 (d, *J* = 13.2 Hz, 1H), 3.65-3.67 (m, 1H), 4.24 (br s, 1H), 7.24-7.27 (m, 2H), 7.38 (d, *J* = 8.0 Hz, 2H), 7.45-7.52 (m, 7H), 7.76 (d, *J* = 8.0 Hz, 2H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 20.5 (2C), 40.3, 43.5, 49.8, 82.6, 114.9 (d, $J_{C\text{-}F}$ = 21.6 Hz) (2C), 125.9, 127.1 (3C), 127.7 (2C), 128.4 (2C), 129.0 (d, $J_{C\text{-}F}$ = 9.6 Hz) (2C), 129.2, 130.3, 131.4 (d, $J_{C\text{-}F}$ = 3.6 Hz), 136.5, 143.3, 153.7, 162.4 (d, $J_{C\text{-}F}$ = 248.3 Hz), 168.8; HRMS (ESI) calcd for $C_{26}H_{26}FN_4O_3S$ $[M + H]^+$: 493.1704; found: 493.1699.

(8a-Methyl-3-phenyl-5,6,8,8a-tetrahydro-7*H*-[1,2,4]oxadiazolo[4,5-*a*]pyrazin-7-yl)(phenyl)methanone (16). To a stirred solution of the compound **5a** (200 mg, 0.500 mmol) in MeCN (5.00 mL) were added SilliaMetS Thiol (1.49 g, 2.00 mmol) and Cs_2CO_3 (489 mg, 1.50 mmol). After being stirred for 1.5 h at 65 °C, the reaction mixture was

filtrated by celite and concentrated. To a stirred solution of the crude material (10.9 mg, 0.0500 mmol) were added benzoyl chloride (6.97 μL, 0.0600 mmol) and Et_3N (13.9 μL, 0.100 mmol). After being stirred for 30 min at room temperature, the reaction mixture was washed with 1 M HCl and H_2O, and dried over $MgSO_4$. After concentration of the filtrate, the residue was purified by column chromatography (*n*-hexane/EtOAc = 1/1) to give the title compound **16** (6.57 mg, 41%): pale yellow amorphous solid; IR (neat) 1648 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.49 (s, 3H), 2.96 (br s, 1H), 3.11-3.19 (m, 1H), 3.36-3.38 (m, 1H), 3.51-3.54 (m, 1H), 3.60 (d, *J* = 13.3 Hz, 1H), 3.71-3.74 (m, 1H), 7.40-7.57 (m, 10H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 22.5, 40.7, 48.2, 79.1, 96.8, 125.2, 126.9 (2C), 127.8 (2C), 128.4 (2C), 128.9 (2C), 129.5, 130.7, 135.8, 157.2, 170.1; HRMS (ESI) calcd for $C_{19}H_{20}N_3O_2$ $[M + H]^+$: 322.1550; found: 322.1542.

(4-Fluorophenyl)[8a-methyl-3-(pyridine-2-yl)-5,6,8,8a-tetrahydro-7*H*-[1,2,4]oxadiazolo[4,5-*a*]pyrazin-7-yl]methanone (17a). By use of a procedure similar to that described for the preparation of the compound **14** from **2o**, the compound **5b** (42.3 mg, 0.105 mmol) was converted to the title compound **17a** (15.3 mg, 43%): colorless solid; mp 95-97 °C; IR (neat) 1623 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.54 (s, 3H), 3.19-3.23 (m, 1H), 3.33-3.38 (m, 1H), 3.51 (d, *J* = 13.2 Hz, 1H), 3.62-3.64 (m, 1H), 3.74 (br s, 1H), 4.28-4.30 (m, 1H), 7.21-7.25 (m, 2H), 7.46-7.49 (m, 3H), 7.80 (d, *J* = 8.0 Hz, 1H), 7.88-7.91 (m, 1H), 8.65 (br s, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 21.1, 40.2, 43.5, 48.0, 96.7, 114.7 (2C) (d, $J_{C\text{-}F}$ = 9.6 Hz), 123.0, 124.5, 128.9 (2C) (d, $J_{C\text{-}F}$ = 9.6 Hz), 131.6, 136.5, 145.1, 148.6, 154.5, 163.2 (d, $J_{C\text{-}F}$ = 6.0 Hz), 168.6; HRMS (ESI) calcd for $C_{18}H_{18}FN_4O_2$ $[M + H]^+$: 341.1408; found: 341.1411.

(4-Fluorophenyl)[8a-methyl-3-(quinolin-2-yl)-5,6,8,8a-tetrahydro-7*H*-[1,2,4]oxadiazolo[4,5-*a*]pyrazin-7-yl]methanone (17b). By use of a procedure similar to that described for the preparation of the compound **14** from **2o**, the compound **5c** (97.1 mg, 0.214 mmol) was converted to the title compound **17b** (59.5 mg, 71%): pale yellow solid; mp 107-109 °C; IR (neat) 1637 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.59 (s, 3H), 3.25-3.31 (m, 1H), 3.44-3.50 (m, 1H), 3.56 (d, *J* = 13.7 Hz, 1H), 3.65-3.67 (m, 1H), 3.79-3.81 (m, 1H), 4.72-4.76 (m, 1H), 7.21-7.26 (m, 2H), 7.47-7.51 (m, 2H), 7.65-7.69 (m, 1H), 7.80-7.83 (m, 1H), 7.91 (d, *J* = 8.6 Hz, 1H), 8.00-8.02 (m, 1H), 8.06-8.07 (m, 1H), 8.42 (d, *J* = 8.6 Hz, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 21.3, 40.5, 43.7, 47.9, 97.4, 114.8 (2C) (d, $J_{C\text{-}F}$ = 21.6 Hz), 119.9, 127.3, 127.4, 127.5, 128.7, 129.0 (2C) (d, $J_{C\text{-}F}$ = 7.2 Hz), 129.8, 131.6, 136.5, 145.4, 146.3, 154.4, 163.2, 168.7; HRMS (ESI) calcd for $C_{22}H_{20}FN_4O_2$ $[M + H]^+$: 391.1565; found: 391.1560.

(1*H*-Indol-6-yl)(8a-methyl-3-phenyl-5,6,8,8a-tetrahydro-7*H*-[1,2,4]oxadiazolo[4,5-

***a*]pyrazin-7-yl)methanone (17c).** To a stirred solution of the compound **5a** (200 mg, 0.500 mmol) in MeCN (5.00 mL) were added SilliaMetS Thiol (1.49 g, 2.00 mmol) and Cs_2CO_3 (489 mg, 1.50 mmol). After being stirred for 1.5 h at 65 °C, the reaction mixture was filtrated by celite and concentrated. The crude material (21.7 mg, 0.100 mmol) was added to a solution of indole-6-carboxylic acid (32.2 mg, 0.200 mmol), HATU (76.1 mg, 0.200 mmol) and *i*-Pr_2NEt (69.7 μL, 0.400 mmol) in DMF (500 μL). After being stirred for 30 min at room temperature, the reaction mixture was extracted with EtOAc, washed with H_2O and dried over $MgSO_4$. After concentration of the filtrate, the residue was purified by column chromatography (*n*-hexane/EtOAc = 1/1) to give the title compound **17c** (35.6 mg, 99%): colorless solid: mp 232-234 °C; IR (neat) 1623 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.57 (s, 3H), 3.17-3.22 (m, 1H), 3.30-3.36 (m, 1H), 3.47-3.50 (m, 1H), 3.78-3.84 (m, 3H), 6.57 (br s, 1H), 7.18 (d, *J* = 8.0 Hz, 1H), 7.28-7.29 (m, 1H), 7.43-7.48 (m, 3H), 7.57-7.58 (m, 3H), 7.64 (d, *J* = 8.0 Hz, 1H), 8.46 (s, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 22.0, 40.5, 43.0, 47.9, 96.3, 100.8 (2C), 110.3 (2C), 117.5, 119.2, 124.8, 126.6, 127.3 (2C), 127.7, 128.4 (2C), 130.2, 134.8, 156.8; HRMS (ESI) calcd for $C_{21}H_{21}N_4O_2$ $[M + H]^+$: 361.1659, found: 361.1659.

(*S*)-8a-Methyl-3-phenyl-6,7,8,8a-tetrahydro-5*H*-[1,2,4]oxadiazolo[4,5-*a*]pyrazin-7-ium (−)-10-camphorsulfonate [(*S*)-20·(−)-CSA] and (*R*)-8a-methyl-3-phenyl-6,7,8,8a-tetrahydro-5*H*-[1,2,4]oxadiazolo[4,5-*a*]pyrazin-7-ium (+)-10-camphorsulfonate [(*R*)-20·(+)-CSA]. To a stirred solution of the compound **5a** (35.3 mg, 0.0878 mmol) in MeCN (1.00 mL) were added SilliaMetS Thiol (266 mg, 0.351 mmol) and Cs_2CO_3 (85.7 mg, 0.263 mmol). After being stirred for 2.5 h at 65 °C, the reaction mixture was filtrated by celite and concentrated. To a solution of this amine in Et_2O–MeOH (9:1, 500 μL) was added (+)-camphorsulfonic acid ((+)-CSA; 9.18 mg, 0.0395 mmol). The resulting solid was collected by filtration to give an (*R*)-**20**·(+)-CSA. The combined filtrate was concentrated and dissolved in EtOAc and saturated $NaHCO_3$. The organic layer was dried over Na_2SO_4 and concentrated. This amine was subjected to the identical protocol using (−)-camphorsulfonic acid ((−)-CSA) to give (*S*)-**20**·(−)-CSA. This series of procedures were repeated several times using (+)-CSA or (−)-CSA. Enantiomeric excess was assessed using chiral HPLC analysis [COSMOSIL CHiRAL 5B (nacalai tesque), flow rate: 1 mL/min, mobile phase: *n*-hexane–EtOH (80:20), UV detection: 254 nm]. The absolute configuration of (*R*)-**20**·(+)-CSA was determined by X-ray crystal structure analysis.

(*S*)-**20**·(−)-CSA: colorless fibrous crystals (8.30 mg, 21%, >99% ee): mp 207-209 °C; IR (neat) 1742 (C=O), 1160 (SO_2), 1043 (SO_2); $[\alpha]^{26}_D$ –6.30 (*c* 0.80, MeOH); ^{1}H-NMR (500 MHz, CD_3OD) δ 0.85 (s, 3H), 1.10 (s, 3H), 1.39-1.44 (m, 1H), 1.62-1.68 (m, 1H), 1.75-

1.76 (m, 3H), 1.88-1.92 (m, 1H), 2.01-2.06 (m, 2H), 2.32-2.37 (m, 1H), 2.59-2.62 (m, 1H), 2.77 (d, *J* = 14.9 Hz, 1H), 2.92-2.98 (m, 1H), 3.21-3.29 (m, 2H), 3.39-3.49 (m, 2H), 3.56-3.66 (m, 2H), 7.52-7.63 (m, 5H); ^{13}C-NMR (125 MHz, CD_3OD) δ 20.3, 20.4 (2C), 22.8, 25.9, 27.9, 39.9, 42.4, 42.5, 43.8, 44.2, 45.4, 59.7, 95.7, 125.5, 129.4 (2C), 130.6 (2C), 132.8, 160.0, 218.7; HRMS (ESI) calcd for $C_{12}H_{16}N_3O$ ((*S*)-**S20**, $[M + H]^+$): 218.1288, found: 218.1289.

(*R*)-**20**·(+)-CSA: colorless fibrous crystals (5.65 mg, 14%, >99% ee): mp 209-211 °C; IR (neat) 1745 (C=O), 1192 (SO_2), 1053 (SO_2); $[\alpha]^{26}_D$ +7.12 (*c* 0.19, MeOH); ^{1}H-NMR (500 MHz, CD_3OD) δ 0.85 (s, 3H), 1.11 (s, 3H), 1.39-1.43 (m, 1H), 1.61-1.67 (m, 1H), 1.75-1.75 (m, 3H), 1.88-1.92 (m, 1H), 2.01-2.06 (m, 2H), 2.32-2.36 (m, 1H), 2.59-2.64 (m, 1H), 2.77 (d, *J* = 14.9 Hz, 1H), 2.91-2.97 (m, 1H), 3.21-3.26 (m, 2H), 3.37-3.51 (m, 2H), 3.57-3.66 (m, 2H), 7.52-7.63 (m, 5H); ^{13}C-NMR (125 MHz, CD_3OD) δ 20.3, 20.5 (2C), 22.8, 25.9, 27.9, 39.9, 42.4, 42.5, 43.8, 44.2, 45.4, 59.7, 95.8, 125.5, 129.4 (2C), 130.6 (2C), 132.8, 160.0, 218.7; HRMS (ESI) calcd for $C_{12}H_{16}N_3O$ ((*R*)-**S20**, $[M + H]^+$): 218.1288, found: 218.1292.

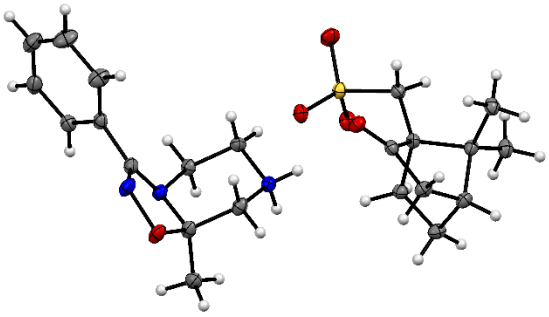
(*S*)-(1*H*-Indol-6-yl)(8a-methyl-3-phenyl-5,6,8,8a-tetrahydro-7*H*-[1,2,4]oxadiazolo-[4,5-*a*]pyrazin-7-yl)methanone [(*S*)-17c]. The compound (*S*)-**20**·(−)-CSA (2.38 mg, 0.00530 mmol) was dissolved in a mixture of EtOAc and saturated $NaHCO_3$ solution. The organic layer was dried over Na_2SO_4 and concentrated. The solution of this residue in CH_2Cl_2 (250 μL) was added to a solution of indole-6-carboxylic acid (32.2 mg, 0.20 mmol), WSC·HCl (1.52 mg, 0.00795 mmol) and HOBt (1.07 mg, 0.00795 mmol) in CH_2Cl_2 (250 μL). After being stirred for 3.5 h at room temperature, the reaction mixture was washed with 1 M HCl and brine and dried over $MgSO_4$. After concentration of the filtrate, the residue was purified by preparative TLC (*n*-hexane/EtOAc = 1/3) to give the title compound (*S*)-**17c** (1.70 mg, 89%): colorless solid: mp 228-230 °C; IR (neat) 1734 (C=O), 1461 (C=N); $[\alpha]^{26}_D$ –42.6 (*c* 0.14, $CHCl_3$); ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.57 (s, 3H), 3.19 (br s, 1H), 3.33 (br s, 1H), 3.47 (br s, 1H), 3.75-3.84 (m, 3H), 6.57 (br s, 1H), 7.18 (d, *J* = 8.6 Hz, 1H), 7.28-7.29 (m, 1H), 7.45-7.48 (m, 3H), 7.57-7.65 (m, 4H), 8.35 (s, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 22.0, 40.5, 43.0, 47.8, 96.3, 100.8 (2C), 110.3 (2C), 117.5, 119.2, 124.8, 126.6, 127.3 (2C), 127.7, 128.4 (2C), 130.1, 134.8, 156.7; HRMS (ESI) calcd for $C_{21}H_{21}N_4O_2$ $[M + H]^+$: 361.1659, found: 361.1656.

(*R*)-(1*H*-Indol-6-yl)(8a-methyl-3-phenyl-5,6,8,8a-tetrahydro-7*H*-[1,2,4]oxadiazolo-[4,5-*a*]pyrazin-7-yl)methanone [(*R*)-17c]. By use of a procedure similar to that described for the preparation of the compound (*S*)-**17c** from (*S*)-**20**·(−)-CSA, the compound (*R*)-**20**·(+)-CSA (2.78 mg, 0.00619 mmol) was converted to the title

compound (*R*)-**17c** (0.790 mg, 35%): colorless solid: mp 226-228 °C; IR (neat) 1730 (C=O), 1461 (C=N); $[\alpha]^{25}_{D}$ +48.4 (*c* 0.16, $CHCl_3$); ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.57 (s, 3H), 3.17-3.22 (m, 1H), 3.31-3.36 (m, 1H), 3.49 (d, *J* = 14.3 Hz, 1H), 3.78-3.84 (m, 3H), 6.58 (br s, 1H), 7.18 (d, *J* = 8.0 Hz, 1H), 7.29 (br s, 1H), 7.44-7.48 (m, 3H), 7.58-7.59 (m, 3H), 7.65 (d, *J* = 8.0 Hz, 1H), 8.35 (s, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 22.0, 40.5, 43.0, 47.9, 96.3, 100.8 (2C), 110.3 (2C), 117.5, 119.2, 124.8, 126.6, 127.3 (2C), 127.7, 128.4 (2C), 130.1, 134.8, 153.2; HRMS (ESI) calcd for $C_{21}H_{21}N_4O_2$ [M + H]$^+$: 361.1659, found: 361.1659.

8. Crystallography

The data of the compound (*R*)-**20**·(+)-CSA ($C_{22}H_{31}N_3O_5S$) was collected with a Rigaku XtaLAB P200 diffractometer using multi-layer mirror monochromated Cu-Kα radiation at 93 K. The substance was crystallized from Et_2O–MeOH as clear block crystals and solved in primitive orthorhombic space group $P2_12_12_1$ with Z = 4. The unit cell dimensions are a = 6.22470(10), b = 15.0053(2), c = 24.0893(2), V = 2300.83(5) Å^3, Dcalc = 1.327 g/cm^3, Mw: 449.56. R = 0.0282, GOF = 1.033, Flack parameter = −0.007(6). The CCDC deposition number: CCDC 1872940.



9. Biological evaluations and binding mode analysis

p38α and p38β kinase assay. p38α and p38β inhibitory activities were evaluated by the off-chip mobility shift assay by the QuickScout® service from Carna Bioscience (Kobe, Japan). Human GST-fusion p38α (9-352) and p38β (1-364) were expressed using *E. coli* expression system. GST-p38α and p38β were purified by using glutathione sepharose chromatography. Each chemical in DMSO at different concentrations was diluted fourfold with reaction buffer [20 mM HEPES (pH 7.5), 0.01% Triton X-100, 2 mM DTT]. For p38 reactions, a combination of the compound, 1 μM modified erketide, 5 mM $MgCl_2$, 150 μM ATP (150 μM for p38α; 75 μM for p38β) in reaction buffer (20 μL) were incubated with each p38 in PP 384-well plates at room temperature for 1 h (n = 2). The reaction was terminated by addition of 70 μL of termination buffer (Carna Biosciences). Substrate and product were separated by electrophoretic means using the LabChip3000 system. The kinase reaction was evaluated by the product ratio, which was calculated

from the peak heights of the substrate (S) and product (P): [P/(P+S)]. Inhibition data were calculated by comparing with no-enzyme controls for 100% inhibition and no-inhibitor reactions for 0% inhibition. IC_{50} values were calculated using GraphPad Prism 5 software (GraphPad Software, Incorporated, La Jolla, CA, USA).

Inhibitory activity of the compounds 17a and 17b against NK3R activation. NK3R antagonistic activity of the compound **17a** and **17b** was evaluated by $[Ca^{2+}]_i$ flux assay. NK3R expressing CHO cells (4.0×10^4 cells/50 μL/well) were inoculated in 10% FBS/Ham's F-12 onto a 96-well black clear-bottom plate (Greiner), followed by incubation at 37 °C for 24 h in 5% CO_2. After the medium was removed, 50 μL of the pigment mixture (Calcium 4 assay kit, Molecular Devices) and 50 μL of the compound solution at different concentrations in assay buffer (HANKS/HEPES containing 2.5 mM probenecid, 0.2 % BSA, and 0.1% CHAPS) was dispensed into each well of the plate, followed by incubation at 37 °C for 1 h. Separately, an NK3R agonist solution (0.05 nM senktide) in assay buffer was prepared on a 96-well sample plate (V-bottom plate, Coster). The cell and agonist solution plates were set in FDSS/μcell (Hamamatsu) and 25 μL of agonist solution was automatically transferred to the cell plate.

Docking simulation of p38 kinase inhibitors. The docking simulation of the compounds (*R*)-**17c** and (*S*)-**17c** for p38α protein kinase (PDB code: 2QD9) was performed. We chose the 2QD9 structure as a reference because of the similar structure of inhibitors. For the docking simulation, we generated the 3D structures of **17c** using LigPrep[33] from the SMILES format. Two mirror-image structures of 1**7c** were generated in this step [(*R*)-**17c** and (*S*)-**17c**]. The protein structure was preprocessed and optimized using Protein Preparation Wizard[34] with the default settings. After the preparation, the remaining water molecule which interacts with Lys53, Asp168, and the inhibitor molecule was removed. A grid box was generated using grid generation function in Glide[18] and a center of the grid was defined to the ligand structure. Subsequently, (*R*)-**17c** and (*S*)-**17c** were docked using Glide in SP mode. After the docking calculation, a water molecule, which interacts with the inhibitor in 2QD9, was put back in the complex structures. The complex structures of p38α–(*R*)-**17c** and p38α–(*S*)-**17c** were energy minimized using MacroModel.[19] The atoms of the residues, which are located within 4 Å from the inhibitor, were set to "freely moving atoms", while the atoms of the residues, which are located within 4 Å from "freely moving atoms", were constrained (force constant = 200 kJ/mol·Å^2). The atoms of the residues, which are located within 4 Å from the constrained atoms, were set to "frozen atoms". The atoms outside of the frozen atoms were not included in the calculations.

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Chapter 2. Development of Novel NK3R Antagonists with a Degradable Scaffold in the Natural Environment

Section 2. Identification of NK3R Antagonists with a Decomposable Core Structure by Scaffold Hopping

Summary

For development of novel NK3R antagonists with less environmental toxicity, a series of heterocyclic scaffolds for the triazolopiperazine substructure in fezolinetant were designed and synthesized. An isoxazolo[3,4-c]piperidine derivative exhibited moderate NK3R antagonistic activity with the favorable properties decomposable under environmental conditions.

In Chapter 2, Section 1, the author established a process for synthesis of novel triazolopiperazine and oxadiazolopiperazine scaffolds. However, the fezolinetant derivative with the 4,5-dihydro-1,2,4-oxadiazole moiety did not show inhibitory activity against NK3R activation, probably because of the loss of a planar heterocyclic scaffold. To identify novel core motif(s) degradable in the natural environment, the author investigated a series of aromatic heterocycles mimicking the 1,2,4-triazole moiety of fezolinetant with maintenance of NK3R antagonistic activity.

Initially, the author designed potential NK3R antagonists, in which the scaffolds could be decomposed via hydrolysis or photolysis under environmental conditions (Figure 1). The author selected the quinoline derivative **1a** as a lead compound for this structure-activity relationship study because of the synthetic feasibility of a series of derivatives. Considering the substitution of the 1,2,4-triazole moiety in **1a** with five-

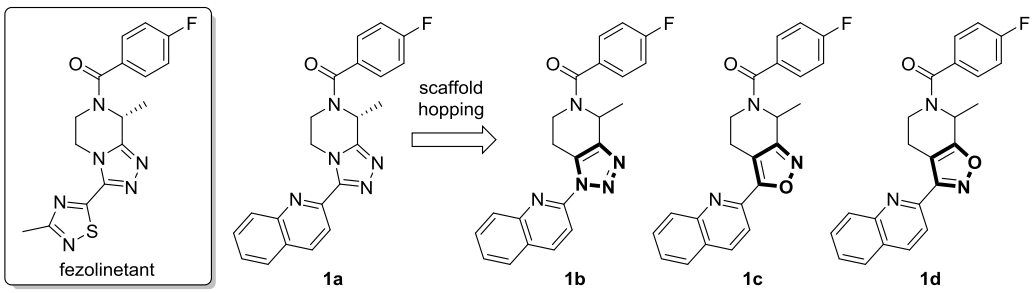
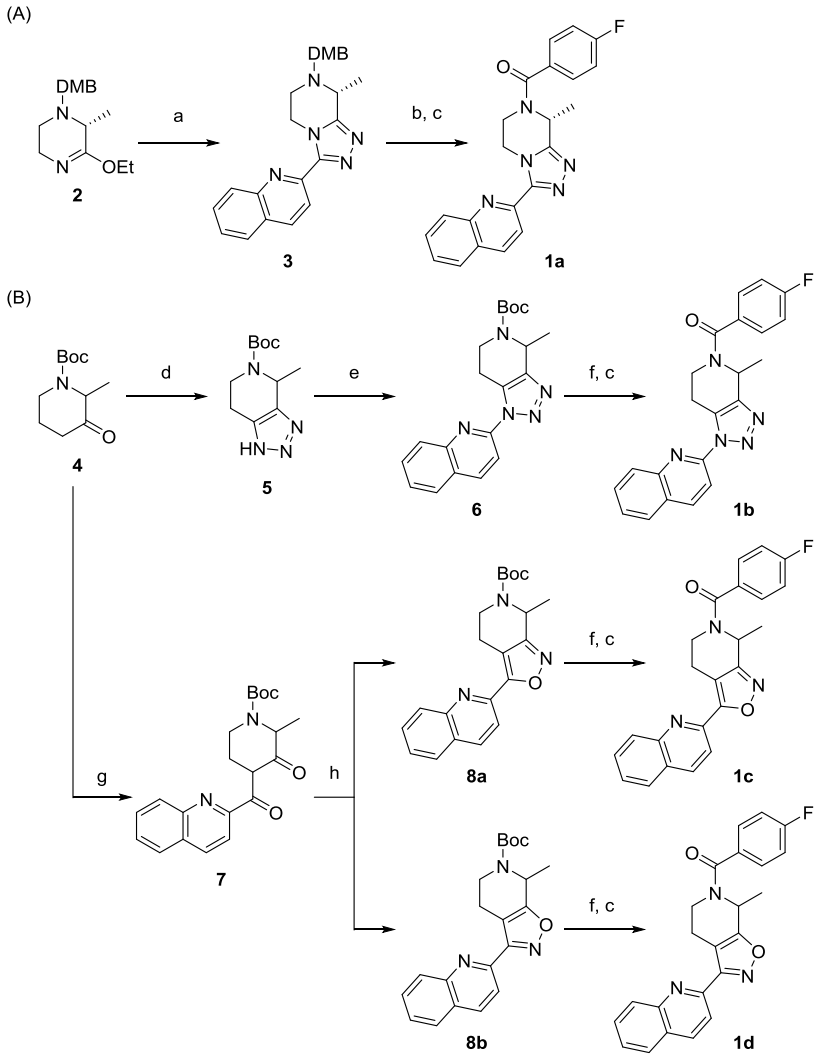


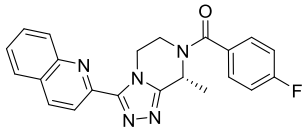
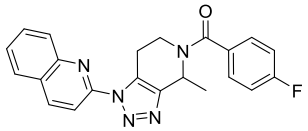
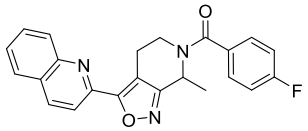
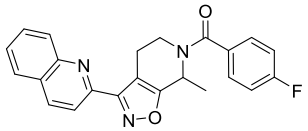
Figure 1. Scaffold hopping from fezolinetant for design of novel NK3R antagonists with reduced environmental toxicity.



Scheme 1. Synthesis of fezolinetant derivatives **1a**–**d**. *Reagent and conditions:* (a) quinoline-2-carbohydrazide, EtOH, 70 °C; (b) TFA, CH_2Cl_2, 0 °C to rt; (c) 4-fluorobenzoyl chloride, Et_3N, CH_2Cl_2, rt; (d) 1-azido-4-nitrobenzene, NH_4OAc, DMF, 80 °C; (e) 2-chloroquinoline, *i*-Pr_2NEt, 120 °C; (f) HCl/dioxane, CH_2Cl_2, rt; (g) LDA, quinaldoyl chloride, THF, −78 °C to rt; (h) $NH_2OH{\cdot}HCl$, NaOH, *i*-PrOH, reflux.

membered aromatic heterocycles consisting of a combination of nitrogens and/or oxygens, three fused piperidine derivatives **1b**–**d** were possible. The arrangements of hydrogen bond acceptors derived from two imino nitrogens of 1,2,4-triazole in **1a** were reproduced in 1,2,3-triazole **1b** and isoxazoles **1c** and **1d**. The [1,2,3]triazolo[4,5-*c*]piperidine scaffold in **1b** was expected to be degradable in response to sunlight exposure because 1,2,3-triazole is decomposed by UV irradiation.[1] Isoxazolo[3,4-*c*]piperidine **1c** and isoxazolo[5,4-*c*]piperidine **1d** were also designed on the basis of possible degradation

Table 1. NK3R antagonism of fused piperidine derivatives.

compound	structure	IC_{50} (μM)[a]
1a		0.26 ± 0.02
1b		4.1 ± 0.5
1c		9.5 ± 1.1
1d		8.2 ± 1.7

[a]IC_{50} values are the concentrations required for 50% inhibition of the NKB-mediated activation of NK3R (n = 6).

of the isoxazole moiety via hydrolysis or photodegradation.[2,3]

The author synthesized the fezolinetant derivatives **1a**–**d**. Triazolopiperazine **3** was obtained by treatment of piperazine **2** with quinoline-2-carbohydrazide. [1,2,4]Triazolo-[3,4-*a*]piperazine **1a** was prepared from **3** in two steps via deprotection of the dimethoxybenzyl (DMB) group followed by acylation with 4-fluorobenzoyl chloride (Scheme 1A).[4] The fused piperidine derivatives **1b**–**d** were synthesized from a common substrate 3-oxopiperidine derivative **4** (Scheme 1B). Reaction of ketone **4** with 1-azido-4-nitrobenzene provided 1,2,3-triazole **5**.[5] N^1-arylation on triazole **5** proceeded by treatment of triazole **5** with 2-chloroquinoline in the presence of *i*-Pr_2NEt to afford the desired 1-(quinolin-2-yl)triazole **6** after separation from a mixture of regioisomers. Boc deprotection of **6** followed by acylation with 4-fluorobenzoyl chloride gave the expected [1,2,3]triazolo[4,5-*c*]piperidine **1b**. The structure of **1b** was determined by X-ray crystal structure analysis. Synthesis of isoxazolopiperidines **1c** and **1d** began with treatment of ketone **4** with quinaldoyl chloride and LDA. The resulting diketone **7** was reacted with hydroxylamine under basic conditions to provide isoxazolo[3,4-*c*]piperidine **8a** and isoxazolo[5,4-*c*]piperidine **8b** after separation by column chromatography. Isoxazoles **1c** and **1d** were obtained by the same manipulations of **8a** and **8b**, respectively, as for the synthesis of **1b**.[6]

Next, the author investigated the biological activity of fused piperidine derivatives

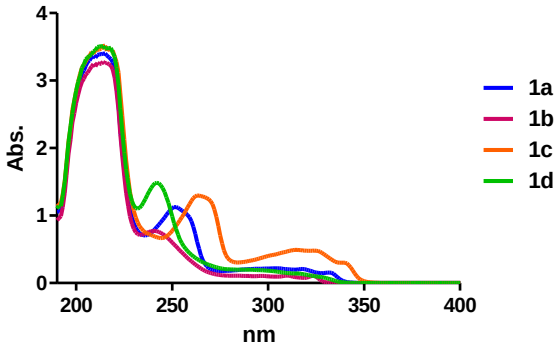


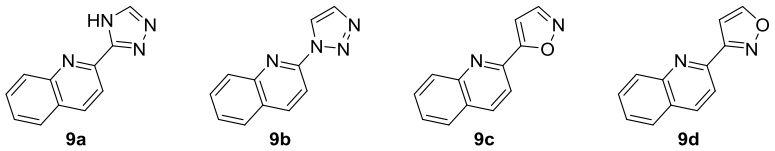
Figure 2. Absorption spectra of fused piperidine-type NK3R antagonists in 50 mM phosphine buffer (pH 7.4)–EtOH (7:3).

1b–**d** by evaluating the antagonism of NKB-induced activation for human NK3R (Table 1). 1,2,3-Triazole **1b** exhibited 15-fold less potent of NK3R inhibition than 1,2,4-triazole **1a** [IC_{50} (**1a**): 0.26 μM, IC_{50} (**1b**): 4.1 μM]. Similarly, isoxazoles **1c** and **1d** showed moderate NK3R inhibition [IC_{50} (**1c**): 9.5 μM, IC_{50} (**1d**): 8.2 μM]. These findings suggested that the more electron deficient heteroaromatic ring was favorable to NK3R inhibition.

The author investigated the UV-vis spectra and photodegradation profiles of **1a**–**d**. It has been reported that the absorption spectra of photodegradable compounds overlap with the actinic spectrum.[7] Initially, the absorption spectra of all derivatives were evaluated to identify those with the potential for photodegradation by sunlight exposure. The UV-vis spectrum of isoxazolo[3,4-*c*]piperidine **1c** showed strong absorption in the wavelength range of 300–350 nm (Figure 2). In contrast, [1,2,4]triazolo[3,4-*a*]piperazine **1a**, [1,2,3]triazolo[4,5-*c*]piperidine **1b** and isoxazolo[5,4-*c*]piperidine **1d** exhibited low UV-visible absorption at above 300 nm. In general, compounds with small HOMO-LUMO energy gap exhibit the red-shifted absorption spectrum.[8] To rationalize the observed red-shifted absorption of **1c**, HOMO-LUMO gaps of the partial structures **9a**–**d** in fezolinetant derivatives **1a**–**d** were calculated using Gaussian 16W program (Table 2).[9] Among the model structures **9a**–**d**, 5-(quinolin-2-yl)isoxazole **9c** had smaller HOMO-LUMO energy gap. The small HOMO-LUMO gap of **1c** would be derived from the extended π-conjugation of 5-(quinolin-2-yl)isoxazole part.

The photochemical stability was assessed by irradiation via a mercury xenon lamp through a UV cut-off filter (below 325 nm), in which the optical spectrum corresponds to that of UV-A (Figure 3A).[10] Among triazolopiperazine **1a** and fused piperidines **1b**–**d**, isoxazolo[3,4-*c*]piperidine **1c** was promptly decomposed, while other scaffolds (**1a**, **1b**

Table 2. HOMO-LUMO energy gaps of the partial structures in fezolinetant derivatives.



	9a	9b	9c	9d
HOMO-LUMO energy gap (eV)	−0.16652	−0.17202	−0.15977	−0.17012

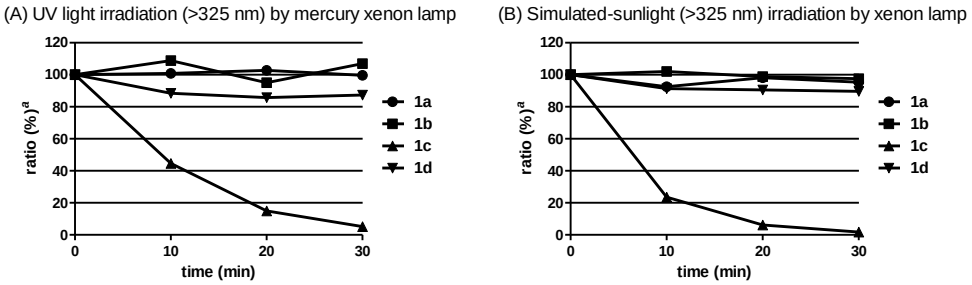


Figure 3. Photodegradation of fused piperidine-type NK3R antagonists. Degradation profile by irradiation of UV light using mercury xenon lamp (A) and by irradiation of simulated-sunlight using xenon lamp (B). Compounds were dissolved in 50 mM phosphine buffer (pH 7.4)–EtOH (7:3) at 30 μM. The light intensities of mercury xenon lamp and xenon lamp were approximately 1500 W/m^2 and 900 W/m^2, respectively. [a]The ratios of compounds were calculated by HPLC analysis using calibration curves

and **1d**) were stable under irradiation with a mercury xenon lamp. The simulated sunlight irradiation by the xenon lamp (>325 nm) also led to decomposition of **1c** (Figure 3B). The less stability of **1c** would be attributable to the strong absorption and/or red-shifted absorption possibly via homolytic cleavage of N-O bond by UV irradiation.[11] The sample after UV-irradiation of **1c** did not show NK3R inhibition (IC_{50} >100 μM). These findings indicated that decomposition would depend on the excitation characteristics of the scaffold by photoirradiation, and that isoxazole **1c** could be inactivated by sunlight exposure.

The author also assessed the stability of fused piperidine derivatives in aqueous buffer by monitoring the intact compounds by HPLC (Figure 4A–C). No hydrolysate was observed from any derivatives at any pH (pH 4.0, 7.4 and 10.0), suggesting that fused

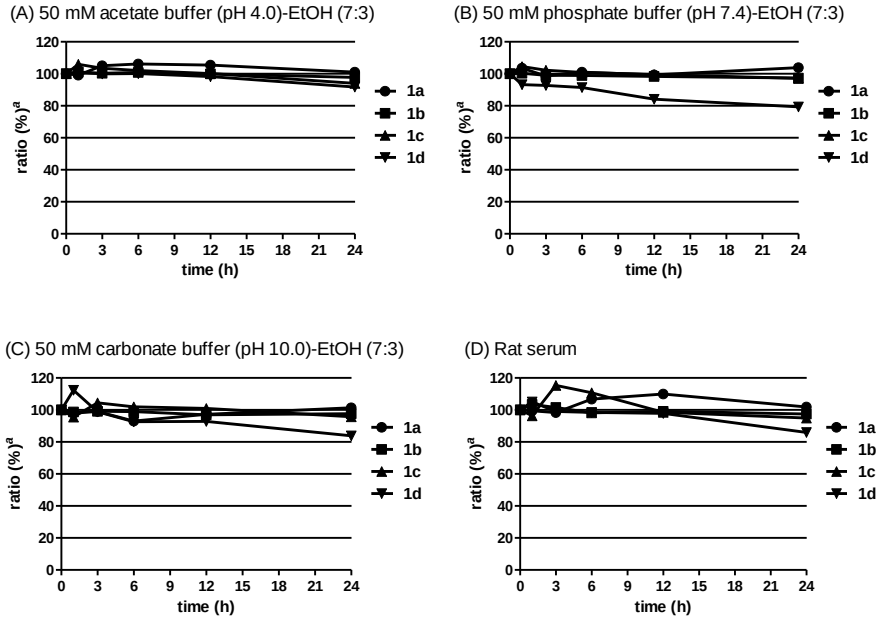


Figure 4. Investigation of the stability of fused piperidine derivatives in aqueous buffer and in rat serum at 37 °C. (A–C) Compounds were dissolved in aqueous buffer (pH 4.0–10.0)–EtOH (7:3) at 30 µM. (D) Compounds were dissolved in rat serum at 5 µM. [a]The ratios of intact compounds were calculated by HPLC analysis using calibration curves.

piperidine scaffolds were unlikely to be decomposed via hydrolysis under environmental conditions. Additionally, the author investigated the stability of fused piperidine derivatives in rat serum (Figure 4D) and found no degradation of any of the compounds. Thus, derivative **1c** could be a novel NK3R antagonist that is stable in the serum and inactivated in the natural environment via solar light irradiation after excretion.

In conclusion, the author identified isoxazolo[3,4-*c*]piperidine **1c** as an NK3R antagonist with a photodegradable scaffold by scaffold hopping of triazolopiperazine. Isoxazolo[3,4-*c*]piperidine **1c** was stable in the aqueous buffer and rat serum, and decomposed only by UV irradiation. Additionally, the result indicated isoxazolo[3,4-*c*]piperidine **1c** may be a candidate for the treatment of sex-hormone disorders with less environmental impact.

Experimental Section

General methods. ^{1}H NMR spectra were recorded using a JEOL AL-400 or a JEOL ECA-500. Chemical shifts are reported in δ (ppm) relative to Me_4Si as an internal standard. ^{13}C NMR spectra were recorded using a JEOL AL-400 or a JEOL ECA-500 and referenced to the residual solvent signal. Exact mass (HRMS) spectra were recorded on Shimadzu LC-ESI-IT-TOF-MS equipment. IR spectra were obtained on a JASCO FT/IR-4100 spectrometer. Optical rotations were measured with a JASCO P-1020 polarimeter. Melting points were measured by a hot stage melting points apparatus (uncorrected). For flash chromatography, Wakogel C-300E (Wako Pure Chemical Industries, Ltd) was employed.

(*R*)-2-[7-(2,4-Dimethoxylbenzyl)-8-methyl-5,6,7,8-tetrahydro-[1,2,4]triazolo[4,3-*a*]pyrazin-3-yl]quinoline (3). To a solution of (*R*)-1-(2,4-dimethoxylbenzyl)-5-ethoxy-6-methyl-piperazine **2**[4] (158 mg, 1.00 mmol) in EtOH (2.00 mL) was added quinoline-2-carbohydrazide[12] (187 mg, 1.00 mmol). After being stirred for 17 h at 70 °C, the reaction mixture was concentrated. The residue was dissolved with EtOAc, washed with 1 M NaOH aqueous solution and brine, and dried over Na_2SO_4. After the filtrate was concentrated, the residue was purified by column chromatography on silica gel (hexane/EtOAc = 1/3) to give the title compound **3** (107 mg, 26%): pale yellow amorphous solid; $[\alpha]^{24}_{D}$ +13.3 (*c* 1.06, $CHCl_3$); ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.76 (d, *J* = 6.3 Hz, 3H), 2.75-2.80 (m, 1H), 3.24-3.27 (m, 1H), 3.61-3.63 (m, 1H), 3.80-3.83 (m, 6H), 3.96 (d, *J* = 13.7 Hz, 1H), 4.11 (d, *J* = 6.3 Hz, 1H), 4.53-4.59 (m, 1H), 4.82-4.86 (m, 1H), 6.48-6.51 (m, 2H), 7.30-7.32 (m, 1H), 7.54-7.55 (m, 1H), 7.68-7.71 (m, 1H), 7.81 (d, *J* = 8.0 Hz, 1H), 8.01 (d, *J* = 8.0 Hz, 1H), 8.20-8.22 (m, 1H), 8.42-8.44 (m, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 17.7, 45.6, 46.0, 50.2, 54.1, 55.4 (2C), 98.6, 104.4, 118.4, 120.3, 127.0, 127.7 (2C), 129.4, 129.7, 131.0, 136.6, 147.3, 148.2, 151.0, 156.2, 158.9, 160.3; HRMS (ESI) calcd for $C_{24}H_{26}N_5O_2$ $[M + H]^+$: 416.2081, found: 416.2086.

(*R*)-(4-Fluorophenyl)[8-methyl-3-(quinolin-2-yl)-5,6-dihydro-[1,2,4]triazolo[4,3-*a*]pyrazin-7(8*H*)-yl]methanone (1a). To a solution of compound **3** (83.0 mg, 0.20 mmol) in CH_2Cl_2 (1.00 mL) was added TFA (115 μL, 1.50 mmol) at 0 °C. After being stirred for 4.5 h at room temperature, the reaction mixture was extracted with EtOAc, washed with H_2O. The aqueous layer was basified by 2 M NaOH and extracted with EtOAc. The organic layer was washed with brine and dried over Na_2SO_4. After the filtrate was concentrated, the residue was dissolved in CH_2Cl_2 (2.00 mL). To the solution were added Et_3N (55.8 μL, 0.0642 mmol) and 4-fluorobenzoyl chloride (3.88 μL, 0.0321 mmol). After being stirred for 15 min, the reaction mixture was diluted with CH_2Cl_2, washed with

1 M HCl aqueous solution and brine, and dried over $MgSO_4$. After the filtrate was concentrated, the residue was purified by column chromatography on silica gel (hexane/EtOAc = 1/3) to give the title compound **1a** (20.3 mg, 26%): colorless solid; mp 232-234 °C; $[\alpha]^{24}_{D}$ +57.3 (*c* 0.60, MeOH); IR (neat) 1641 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.67 (d, *J* = 6.9 Hz, 3H), 3.66-3.71 (m, 1H), 4.19-4.22 (m, 1H), 4.42-4.46 (m, 1H), 5.08-5.11 (m, 1H), 5.67 (br s, 1H), 7.28-7.31 (m, 2H), 7.59-7.67 (m, 3H), 7.80-7.83 (m, 1H), 8.01-8.10 (m, 2H), 8.29-8.30 (m, 1H), 8.49 (d, *J* = 8.0 Hz, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 18.6, 38.4, 45.3, 46.2, 115.0 (d, $J_{C\text{-}F}$ = 21.6 Hz) (2C), 119.2, 126.9, 127.0, 128.6, 128.9 (d, $J_{C\text{-}F}$ = 9.6 Hz) (2C), 129.7 (2C), 131.5 (d, $J_{C\text{-}F}$ = 3.6 Hz), 136.8, 146.3, 147.1, 150.1, 153.2, 163.4, 168.4; HRMS (ESI) calcd for $C_{22}H_{19}FN_5O$ $[M + H]^+$: 388.1568, found: 388.1566.

***tert*-Butyl 4-methyl-1,4,6,7-tetrahydro-5*H*-[1,2,3]triazolo[4,5-*c*]pyridine-5-carboxylate (5).** To a solution of *tert*-butyl 2-methyl-3-oxopiperidine-1-carboxylate **4** (213 mg, 1.00 mmol) in DMF (2.00 mL) were added 1-azido-4-nitrobenzene[13] (213 mg, 1.30 mmol) and NH_4OAc (385 mg, 5.00 mmol). After being stirred for 11 h at 80 °C, the reaction mixture was extracted with EtOAc, washed with 1 M HCl aqueous solution and brine, and dried over $MgSO_4$. After the filtrate was concentrated, the residue was purified by column chromatography on silica gel (hexane/EtOAc = 3/1) to give the title compound **5** (148 mg, 62%): colorless amorphous solid; IR (neat) 1693 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.47 (d, *J* = 6.3 Hz, 3H), 1.50 (s, 9H), 2.78-2.83 (m, 2H), 3.03-3.09 (m, 1H), 4.44 (br s, 1H), 5.40 (br s, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.4, 22.4, 28.5 (3C), 37.4, 47.0, 80.4, 140.7, 145.3, 154.7; HRMS (ESI) calcd for $C_{11}H_{18}N_4O_2$ $[M + H]^+$: 239.1503, found: 239.1501.

***tert*-Butyl 4-methyl-1-(quinolin-2-yl)-1,4,6,7-tetrahydro-5*H*-[1,2,3]triazolo[4,5-*c*]-pyridine-5-carboxylate (6).** A solution of triazole **5** (71.4 mg, 0.300 mmol) and 2-chloroquinoline (172 mg, 0.900 mmol) in *i*-Pr_2NEt (209 μL, 1.20 mmol) was stirred for 58 h at 120 °C. The reaction mixture was extracted with EtOAc, washed brine, and dried over $MgSO_4$. After the filtrate was concentrated, the residue was purified by column chromatography on silica gel (hexane/EtOAc = 6/1) to give the title compound **6** (23.0 mg, 21%): colorless amorphous solid; IR (neat) 1695 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.52 (s, 9H), 1.56 (d, *J* = 6.9 Hz, 3H), 3.08-3.13 (m, 1H), 3.27-3.34 (m, 1H), 3.55-3.59 (m, 1H), 4.51 (br s, 1H), 5.45 (br s, 1H), 7.57-7.60 (m, 1H), 7.74-7.77 (m, 1H), 7.87 (d, *J* = 8.6 Hz, 1H), 8.01 (d, *J* = 8.6 Hz, 1H), 8.29-8.35 (m, 2H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 19.0, 25.0, 28.5 (3C), 36.5, 47.0, 80.2, 113.9, 127.0, 127.4, 127.7, 128.9, 130.5, 131.4, 139.3, 146.3, 146.9, 149.6, 154.5; HRMS (ESI) calcd for $C_{20}H_{24}N_5O_2$ [M +

H]$^+$: 366.1925, found: 366.1921.

(4-Fluorophenyl)[4-methyl-1-(quinolin-2-yl)-1,4,6,7-tetrahydro-5*H*-[1,2,3]triazolo-[4,5-*c*]pyridin-5-yl]methanone (1b). To a solution of quinoline **6** (7.80 mg, 0.0214 mmol) in CH_2Cl_2 (500 μL) was added HCl (4 M in dioxane; 250 μL, 1.00 mmol). After being stirred for 1 h, the reaction mixture was basified by 2 M NaOH solution at 0 °C, and the whole was extracted with CH_2Cl_2. The organic layer was washed with brine, dried over Na_2SO_4, and concentrated. The residue was dissolved in CH_2Cl_2 (500 μL). To the solution were added Et_3N (8.95 μL, 0.0642 mmol) and 4-fluorobenzoyl chloride (3.88 μL, 0.0321 mmol). After being stirred for 15 min, the reaction mixture was diluted with CH_2Cl_2, washed with 1 M HCl aqueous solution and brine, and dried over $MgSO_4$. After the filtrate was concentrated, the residue was purified by column chromatography on silica gel (hexane/EtOAc = 3/1) to give the title compound **1b** (7.30 mg, 88%): colorless solid; mp 199-201 °C; IR (neat) 1637 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.58 (d, J = 6.9 Hz, 3H), 3.30-3.54 (m, 3H), 4.17 (br s, 1H), 5.50 (br s, 1H), 7.26-7.29 (m, 2H), 7.53-7.55 (m, 2H), 7.65-7.67 (m, 1H), 7.83-7.84 (m, 1H), 8.01-8.07 (m, 2H), 8.22 (d, J = 8.6 Hz, 1H), 8.62 (d, J = 8.6 Hz, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 18.5, 23.8, 38.0, 46.1, 113.2, 115.0 (d, $J_{C\text{-}F}$ = 21.6 Hz) (2C), 126.7 (2C), 127.5, 127.9, 128.6 (d, $J_{C\text{-}F}$ = 4.8 Hz) (2C), 130.3, 130.7, 132.4 (d, $J_{C\text{-}F}$ = 3.6 Hz), 139.6, 145.2, 145.3, 148.6, 162.3 (d, $J_{C\text{-}F}$ = 247.1 Hz), 168.7; HRMS (ESI) calcd for $C_{22}H_{19}FN_5O$ [M + H]$^+$: 388.1568, found: 388.1566.

***tert*-Butyl 2-methyl-3-oxo-4-(quinoline-2-carbonyl)piperidine-1-carboxylate (7).** To a solution of *i*-Pr_2NH (425 μL, 3.00 mmol) in THF (2.00 mL) was added *n*-BuLi (1.39 M in hexane; 2.45 mL, 3.40 mmol) at −78 °C under argon and the mixture was stirred for 20 min at 0 °C. A solution of ketone **4** (426 mg, 2.00 mmol) in THF (2.00 mL) was added to the mixture at −78 °C and the mixture was stirred for 30 min at this temperature. A solution of quinaldoyl chloride (497 mg, 2.60 mmol) in THF (4.00 mL) was added to the mixture. After being stirred for 6 h at room temperature, the reaction mixture was quenched with 1 M HCl solution and the whole was extracted with EtOAc. The organic layer was washed with aqueous saturated $NaHCO_3$ and brine, dried over by $MgSO_4$ and concentrated. The residue was purified by column chromatography on silica gel (hexane/EtOAc = 10/1) to give the title compound **7** (271 mg, 37%): yellow solid; mp 123-125 °C; IR (neat) 1687 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.49-1.50 (m, 12H), 2.78-2.81 (m, 1H), 2.97-2.99 (m, 2H), 4.10-4.12 (m, 1H), 4.75 (br s, 1H), 7.62-7.66 (m, 1H), 7.76-7.80 (m, 1H), 7.87 (d, J = 8.6 Hz, 1H), 8.04 (d, J = 8.6 Hz, 1H), 8.14 (d, J = 8.6 Hz, 1H), 8.31 (d, J = 8.6 Hz, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ 17.5, 25.9, 28.5

(3C), 37.6, 52.8, 80.0, 106.8, 120.6, 127.7, 128.4, 128.6, 129.4, 130.4, 137.3, 137.5, 145.6, 153.9, 154.1, 186.6; HRMS (ESI) calcd for $C_{21}H_{25}N_2O_4$ $[M + H]^+$: 369.1809, found: 369.1811.

***tert*-Butyl 7-methyl-3-(quinolin-2-yl)-4,5-dihydroisoxazolo[3,4-*c*]pyridine-6-(7*H*)-carboxylate (8a) and *tert*-butyl 7-methyl-3-(quinolin-2-yl)-4,7-dihydroisoxazolo[5,4-*c*]pyridine-6-(5*H*)-carboxylate (8b).** To a solution of diketone **7** (159 mg, 0.432 mmol) in *i*-PrOH (2.00 mL) were added $NH_2OH·HCl$ (60.0 mg, 0.864 mmol) and NaOH (52.0 mg, 1.30 mmol). After being stirred for 2 h under reflux conditions, the reaction mixture was extracted with EtOAc. The organic layer was washed with aqueous saturated $NaHCO_3$ and brine, dried over Na_2SO_4, and concentrated. The residue was purified by column chromatography on silica gel (hexane/EtOAc = 20/1) to give the title compounds **8a** (35.2 mg, 22%) and **8b** (43.5 mg, 28%).

Compound **8a**: colorless solid; mp 137-139 °C; IR (neat) 1698 (C=O) cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.52 (s, 9H), 1.56 (d, *J* = 7.0 Hz, 3H), 2.93-3.08 (m, 2H), 3.39-3.44 (m, 1H), 4.43 (br s, 1H), 5.55 (br s, 1H), 7.55-7.59 (m, 1H), 7.72-7.76 (m, 1H), 7.83 (d, *J* = 8.1 Hz, 1H), 7.97 (d, *J* = 8.7 Hz, 1H), 8.09 (d, *J* = 8.1 Hz, 1H), 8.24 (d, *J* = 8.7 Hz, 1H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 19.7, 22.1, 28.4 (3C), 36.3, 46.4, 80.3, 113.1, 118.5, 127.3, 127.5, 127.6, 129.7, 130.0, 136.9, 147.6, 147.9, 154.4, 161.9, 163.9; HRMS (ESI) calcd for $C_{21}H_{24}N_3O_3$ $[M + H]^+$: 366.1812, found: 366.1812.

Compound **8b**: colorless solid; mp 127-129 °C; IR (neat) 1697 (C=O) cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.51-1.53 (m, 12H), 2.93-3.05 (m, 2H), 3.24-3.28 (m, 1H), 4.35-4.48 (br m, 1H), 5.29-5.45 (br m, 1H), 7.56-7.60 (m, 1H), 7.71-7.76 (m, 1H), 7.83-7.85 (m, 1H), 8.10-8.14 (m, 2H), 8.23 (d, *J* = 8.2 Hz, 1H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 17.9, 22.5, 28.4 (3C), 37.0, 47.6, 80.4, 111.7, 119.6, 127.2, 127.6, 127.9, 129.7, 129.8, 136.7, 147.9, 149.7, 154.3, 159.6, 169.3; HRMS (ESI) calcd for $C_{21}H_{24}N_3O_3$ $[M + H]^+$: 366.1812, found: 366.1822.

(4-Fluorophenyl)[7-methyl-3-(quinolin-2-yl)-4,5-dihydroisoxazolo[3,4-*c*]pyridin-6(7*H*)-yl]methanone (1c). By use of a procedure similar to that described for the preparation of the compound **1b** from **6**, the compound **8a** (43.5 mg, 0.119 mmol) was converted to the title compound **1c** (36.8 mg, 80%): colorless solid; mp 172-174 °C; IR (neat) 1640 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.60 (d, *J* = 6.9 Hz, 3H), 2.99-3.06 (m, 1H), 3.30-3.42 (m, 2H), 4.04-4.06 (m, 1H), 5.63-5.65 (m, 1H), 7.26-7.30 (m, 2H), 7.54-7.56 (m, 2H), 7.63-7.67 (m, 1H), 7.80-7.83 (m, 1H), 7.97-8.02 (m, 2H), 8.06 (d, *J* = 8.6 Hz, 1H), 8.51 (d, *J* = 8.6 Hz, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 18.9, 21.3, 38.4, 45.4, 112.0, 115.0 (d, $J_{C\text{-}F}$ = 21.6 Hz) (2C), 117.9, 127.0 (d, $J_{C\text{-}F}$ = 13.2 Hz)

(2C), 127.5, 128.6, 128.7, 130.0, 132.1 (d, $J_{C\text{-}F}$ = 3.6 Hz), 137.1, 146.5, 147.0, 161.2, 161.3, 162.3 (d, $J_{C\text{-}F}$ = 248.3 Hz), 163.3, 168.7; HRMS (ESI) calcd for $C_{23}H_{19}FN_3O_2$ [M + H]$^+$: 388.1456, found: 388.1458.

(4-Fluorophenyl)[7-methyl-3-(quinolin-2-yl)-4,7-dihydroisoxazolo[5,4-*c*]pyridin-6(5*H*)-yl]methanone (1d). By use of a procedure similar to that described for the preparation of the compound **1b** from **6**, the compound **8b** (35.2 mg, 0.0961 mmol) was converted to the title compound **1d** (25.2 mg, 68%): colorless solid; mp 161-163 °C; IR (neat) 1603 (C=O) cm^{-1}; ^{1}H-NMR (500 MHz, DMSO-d_6) δ 1.60 (d, J = 6.9 Hz, 3H), 2.99-3.06 (m, 1H), 3.30-3.42 (m, 2H), 4.04-4.06 (m, 1H), 5.63-5.65 (m, 1H), 7.26-7.30 (m, 2H), 7.54-7.56 (m, 2H), 7.63-7.67 (m, 1H), 7.80-7.83 (m, 1H), 7.97-8.02 (m, 2H), 8.06 (d, J = 8.6 Hz, 1H), 8.51 (d, J = 8.6 Hz, 1H); ^{13}C-NMR (125 MHz, DMSO-d_6) δ 17.1, 21.8, 37.2, 46.3, 110.6, 115.0 (d, $J_{C\text{-}F}$ = 19.2 Hz) (2C), 118.7, 127.0, 127.4 (d, $J_{C\text{-}F}$ = 18.0 Hz) (2C), 128.7 (2C), 129.6, 132.1 (d, $J_{C\text{-}F}$ = 3.6 Hz) , 136.8, 146.9, 148.6, 158.8, 161.3, 162.3 (d, $J_{C\text{-}F}$ = 247.1 Hz), 168.4, 168.6; HRMS (ESI) calcd for $C_{23}H_{19}FN_3O_2$ [M + H]$^+$: 388.1456, found: 388.1464.

Inhibitory activity of the fused piperidine derivatives against NK3R. NK3R antagonistic activity of the fused piperidine derivatives was evaluated by $[Ca^{2+}]_i$ flux assay. NK3R expressing CHO cells (4.0×10^4 cells/50 μL/well) were inoculated in 10% FBS/Ham's F-12 onto a 96-well black clear-bottom plate (Greiner), followed by incubation at 37 °C for 24 h in 5% CO_2. After the medium was removed, 50 μL of the pigment mixture (Calcium 4 assay kit, Molecular Devices) and 50 μL of the compound solution at different concentrations in assay buffer (HANKS/HEPES containing 2.5 mM probenecid, 0.2 % BSA, and 0.1% CHAPS) was dispensed into each well of the plate, followed by incubation at 37 °C for 1 h. Separately, an NK3R agonist solution (0.1 nM senktide) in assay buffer was prepared on a 96-well sample plate (V-bottom plate, Coster). The cell and agonist solution plates were set in FDSS/μcell (Hamamatsu) and 25 μL of agonist solution was automatically transferred to the cell plate.

UV-vis spectra. UV-vis spectra were recorded on Shimadzu UV-2450 UV-vis spectrophotometer at 20 °C. Compounds (30 μM) were dissolved in a mixture of 50 mM phosphate buffer (pH 7.4) and EtOH [70:30 (v/v)] (containing 0.1% DMSO).

Investigation of photodegradation of fused piperidine derivatives. Compounds (30 μM) were dissolved in a mixture of 50 mM phosphate buffer (pH 7.4) and EtOH [70:30 (v/v)] (containing 0.1% DMSO) and the reaction mixture was exposed UV-light irradiation using a MAX-303 (Asahi-bunko, Japan) or an MUV-202U (Moritex Co.,

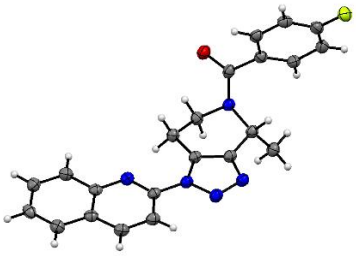
Japan) and special glass filters restricting the transmission of wavelength below 325 nm. A 50 μL aliquot was sampled at the indicated intervals, and distilled with MeCN (50 μL). An aliquot of the sample was analyzed by HPLC and the peak area was recorded by UV detection at 254 nm. The ratios of the intact compounds were calculated from the calibration curves.

Investigation of the stability of fused piperidine derivatives in aqueous buffer. Compounds (30 μM) were incubated in a mixture of 50 mM aqueous buffer [acetate buffer (pH 4.0), phosphate buffer (pH 7.4), or carbonate buffer (pH 10.0)] and EtOH [70:30 (v/v)] (containing 0.1% DMSO) at 37 °C. An aliquot of the sample was analyzed by HPLC at the indicated intervals and the peak area was recorded by UV detection at 254 nm. The ratios of the intact compounds were calculated from the calibration curves.

Investigation of the stability of fused piperidine derivatives in rat serum. Compounds (5.0 μM) were incubated in rat serum (containing 1% DMSO) at 37 °C. A 10 μL aliquot was sampled at the indicated intervals, and extracted with MeCN (50 μL). An aliquot of the sample was analyzed by HPLC and the peak area was recorded by UV detection at 254 nm. The ratios of the intact compounds were calculated from the calibration curves.

Crystallography

The data of the compound **1b** ($C_{22}H_{18}FN_5O$) was collected with a Rigaku XtaLAB P200 diffractometer using multi-layer mirror monochromated Cu-Kα radiation at 93 K. The substance was crystallized from $CHCl_3$–*n*-hexane as clear colorless block crystals and solved in primitive orthorhombic space group $P2_1/c$ with $Z = 4$. The unit cell dimensions are $a = 7.5117(3)$, $b = 35.6861(8)$, $c = 7.4465(4)$, $V = 1788.70(15)$ Å^3, Dcalc = 1.439 g/cm^3, Mw: 387.41. $R = 0.0746$, GOF = 1.036. The CCDC deposition number: CCDC 1893155.



Computational details

All DFT calculations were performed using the Gaussian 16W package.[9] HOMO and LUMO orbital energies were determined from structures optimized at the B3LYP 6-31G bias set.

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Chapter 3. Conclusions

1. Structure-activity relationship studies of a quinoline-base NK3R antagonist, talnetant, were conducted to develop NK3R antagonists with less environmental impact. Diol and thiol derivatives exhibited comparable NK3R binding inhibition as the parent talnetant. Among them, thiol derivative was spontaneously converted into the inactive form via air oxidation.

2. To explore the potentially decomposable scaffolds by scaffold hopping from a triazolopiperazine-based NK3R antagonist, fezolinetant, facile synthetic methods of [1,2,4]triazolo[4,3-*a*]piperazine and [1,2,4]oxadiazolo[4,5-*a*]piperazine derivatives via gold(I)-catalyzed domino cyclization were developed. While the oxadiazolopiperazine derivative did not exhibit NK3R antagonistic activity, the scaffold was applicable to the design of p38 MAPK inhibitors.

3. A structure-activity relationship study of the 1,2,4-triazole moiety in fezolinetant was conducted. Fezolinetant derivatives with fused piperidine scaffolds exhibited moderate NK3R antagonistic activity. Among them, an isoxazolo[3,4-*c*]piperidine scaffold had a favorable photodegradable property, being decomposed by UV irradiation.

In summary, through structure-activity relationship studies of NK3R antagonists, the author developed two types of novel NK3R antagonists that were converted into their inactive forms via air oxidation and sunlight exposure under environmental conditions. It is still challenging to design drug candidates, which satisfy two apparently inconsistent properties: high stability for prolonged storage before therapeutic application and readily decomposable structure to avoid the environmental toxicity after excretion from the body. The authors believe that the findings from the structure-activity relationship studies in this thesis would be useful for development of therapeutic agents with reduced environmental impact.

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List of Publications

This study was published in the following papers.

Chapter 1. Development of novel NK3 receptor antagonists with reduced environmental impact
Koki Yamamoto, Shiho Okazaki, Hiroaki Ohno, Fuko Matsuda, Satoshi Ohkura, Kei-ichiro Maeda, Nobutaka Fujii, Shinya Oishi
Bioorg. Med. Chem. **2016**, *24*, 3494–3500.

Chapter 2.

Section 1. Synthesis of triazolo- and oxadiazolopiperazines by gold(I)-catalyzed domino cyclization: application to the design of a mitogen activated protein (MAP) kinase inhibitor
Koki Yamamoto, Yasushi Yoshikawa, Masahito Ohue, Shinsuke Inuki, Hiroaki Ohno, Shinya Oishi
Org. Lett. **2019**, *21*, 373–377.

Section 2. Scaffold hopping of fused piperidine-type NK3 receptor antagonists for reducing the environmental impact
Koki Yamamoto, Shinsuke Inuki, Hiroaki Ohno, Shinya Oishi
Manuscript in preparation.