博士論文

TSPO リガンドの合成と構造活性相関

および薬理作用に関する研究

深谷　孝幸

本学位論文は、下記の原著論文を基に作成され、東京大学大学院薬学系研究科に提出されたものである。

1. Design, synthesis and structure-activity relationship of novel tricyclic benzimidazolone derivatives as potent 18 kDa translocator protein (TSPO) ligands
Takayuki Fukaya, Toru Kodo, Takeo Ishiyama, Hiroyuki Nishikawa, Satoko Baba, Shuji Masumoto
Bioorg. Med. Chem. **2013**, *21*, 1257-1267.

2. Design, synthesis and structure-activity relationship of novel benzoxazolone derivatives as 18 kDa translocator protein (TSPO) ligands
Takayuki Fukaya, Toru Kodo, Takeo Ishiyama, Hroyoshi Kakuyama, Hiroyuki Nishikawa, Satoko Baba, Shuji Masumoto
Bioorg. Med. Chem. **2012**, *20*, 5568-5582.

3. Convenient Synthesis of Benzoxazolone Derivatives by Cross-Coupling of Benzoxazolone Boronates with Aryl Halides
Takayuki Fukaya, Shuji Masumoto
Synthesis, **2013**, *45*, 3269-3275.

4. Identification of a Novel Benzoxazolone Derivative as a Selective, Orally active 18 kDa Translocator Protein (TSPO) Ligand
Takayuki Fukaya, Takeo Ishiyama, Satoko Baba, Shuji Masumoto
J. Med. Chem. **2013**, *56*, 8191-8195.

　　本学位論文は、第 2 章を上記の論文 1 を基に作成した。第 3 章は論文 2 および論文 3 を、第 4 章は論文 4 を基にそれぞれ作成した。

目次

略語表

Ac	acetyl
ANT	adenine nucleotide transporter
aq.	aqueous
Ar	aryl
ATR	attenuated total reflection
AUC	area under the curve
BA	bioavailability
Bn	benzyl
Boc	*tert*-butoxycarbonyl
BOPCl	bis(2-oxo-3-oxazolidinyl)phosphinic chloride
br	broad
Bu	butyl
^{t}Bu	*tert*-butyl
calcd	calculated
cat	catalytic
CBR	central benzodiazepine receptor
CDI	1,1'-carbonyldiimidazole
Cmax	maximum drug concentration
Compd	compound
Cy	cyclohexyl
CYP	cytochrome P450
d	doublet
dba	dibenzylideneacetone
DBI	diazepam binding inhibitor
DMF	*N*, *N*-dimethylformamide
DMSO	dimethyl sulfoxide
EDCI	1-ethyl-3-(3-dimethylaminopropyl)carbodiimide
ESI	electrospray ionization
Et	ethyl
F%	% oral bioavailability
GABA	γ-aminobutyric acid
Hex	hexyl
HOBt	1-hydroxybenzotriazole

HRMS	high resolution mass spectrometry
IC_{50}	half-maximum inhibitory concentration
Inh	inhibition
ip	intraperitoneally
IR	infrared spectroscopy
iv	intravenous
J	coupling constant (in NMR spectrometry)
Ki	inhibition constant
lit.	literature value
m	multiplet
MBR	mitochondrial benzodiazepine receptor
Me	methyl
MHz	megahertz
mp	melting point
NBS	*N*-bromosuccinimide
NMDA	*N*-methyl-D-aspartic acid
NMR	nuclear magnetic resonance
PBR	peripheral benzodiazepine receptor
PET	positron emission tomography
PK	pharmacokinetics
Ph	phenyl
po	*per os*
^{i}Pr	isopropyl
quant.	quantitative
rt	room temperature
s	singlet
SAR	structure-activity relationships
SEM	standard error of the mean
SSRI	selective serotonim reuptake inhibitor
t	triplet
THF	tetrahydrofuran
TLC	thin layer chromatography
TsOH	*p*-toluenesulfonic acid
TSPO	translocator protein
VDAC	voltage-dependent anion channel

第 1 章　諸言

第 1 節　不安障害治療薬の現状

　近年、社会情勢の変化に伴うストレス増大により、うつ病、不安障害に代表される精神疾患の罹患率は増加の一途をたどっている。精神疾患の中でも、不安障害はもっとも一般的な疾患のひとつであり、患者数は全世界の人口の約一割にも達するといわれている [1]。

　不安とは、日常生活の中で経験する感情の一種であり、生命維持に必要な自己防衛的感情機能である。ところが遺伝的要因または環境的要因により、脳の特定部位での異常興奮が引き起こされることで日常生活に支障をきたすほどの過剰な恐怖および緊張を生じ、時に身体的兆候（めまい、発汗等）が引き起こされることがある。このように過剰で場合によっては持続的な恐怖、緊張を伴い病的症状となった状態について不安障害と診断される。不安障害は、全般性不安障害、社会不安障害、恐怖症、パニック障害、外傷後ストレス障害、強迫性障害等に分類されている [2b]。また不安障害は発症頻度が高く、うつ病あるいは統合失調症などの精神疾患と高頻度で併発する疾患である。

　不安障害に対する治療薬としては、ベンゾジアゼピン系薬剤と、セロトニン 1A 受容体作動薬、選択的セロトニン再取り込み阻害薬、セロトニン・ノルアドレナリン再取り込み阻害薬、三環系抗うつ薬といったセロトニン系薬剤とが挙げられる (Figure 1-1)。ベンゾジアゼピン系薬剤は、即効性かつ強力な抗不安作用を示す反面、薬物依存形成、過度の鎮静、筋弛緩、健忘などの副作用が問題点となっている [2,3]。一方で、セロトニン系薬剤は抗うつ作用など幅広い作用スペクトルを示すが、薬効発現までの期間が長いことが臨床現場で課題となっている [2]。このような背景により、即効性かつ幅広い作用スペクトルを示し副作用の少ない新規な不安障害治療薬の創製が望まれている。
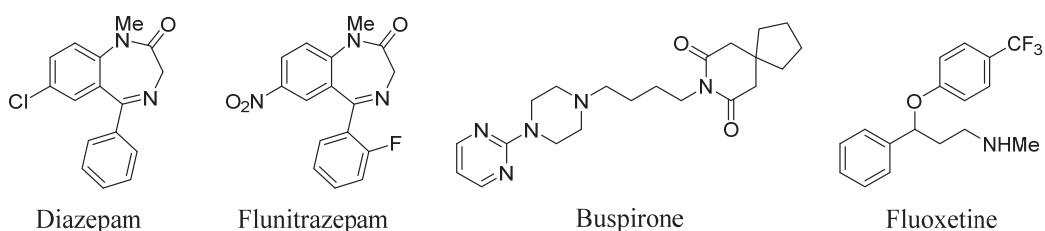

Figure 1-1. Chemical structures of representative antianxiety drugs

第 2 節　18 kDa translocator protein (TSPO) の機能および神経ステロイドの薬理作用

　　抗不安薬としてその即効性かつ強力な薬効から広く処方されているベンゾジアゼピン系薬剤の作用部位については、1977 年に Möhler らおよび Braestrup らの 2 つのグループによりラット及びヒトの脳内において特異的に結合する部位 (中枢型ベンゾジアゼピン受容体、CBR) の存在が明らかにされた [4]。本発見がベンゾジアゼピン系薬剤の生理機能の解明が進むきっかけとなり、GABA$_A$ 受容体に関する研究が進展することとなった。

　　ベンゾジアゼピン系薬剤が結合する CBR は、GABA$_A$ 受容体およびクロリドイオンチャネルと複合体を形成している。ベンゾジアゼピン系薬剤が CBR に作用することで、γ-aminobutyric acid (GABA) の GABA$_A$ 受容体への親和性が高まり、クロリドイオンチャネルの開口頻度の増大に伴いクロリドイオン流入が促進され、膜電位が過分極される [5]。この過分極により神経細胞の興奮が抑制され抗不安作用、抗痙攣作用などの薬理作用が発揮される。また、同時にベンゾジアゼピン系薬剤の CBR への結合により薬物依存形成、過度の鎮静、筋弛緩、健忘などの望ましくない薬理作用も引き起こされる。

　　一方で、CBR の発見と同時期に Braestrup らにより末梢器官にも diazepam 等のベンゾジアゼピン系薬剤の結合部位が存在することが明らかになった [6]。本結合部位は、CBR とは薬理学的ならびに分子的性質が異なり、GABA$_A$ 受容体とは共役していない [7]。この末梢器官におけるベンゾジアゼピン系薬剤の結合部位は、末梢臓器で初めに発見されたことから末梢型ベンゾジアゼピン受容体 (PBR) 、またミトコンドリア膜上に多く存在する [8] ことからミトコンドリア型ベンゾジアゼピン受容体 (MBR) 等複数の名称で呼ばれていたが、2006 年に Papadopoulos らにより 18kDa Translocater Protein (TSPO) に統一された [9]。

　　TSPO は、主にミトコンドリア外膜上に存在し、腎臓、心臓、肺、脾臓および副腎等の末梢臓器に加え、また中枢神経系のグリア細胞にも発現していることが明らかとなった [10]。TSPO は、32kDa voltage-dependent anion channel (VDAC)、30 kDa adenine nucleotide transporter (ANT) と複合体を形成している [11]。TSPO の生理機能としては、ステロイド生合成 [12]、細胞増殖 [13] およびアポトーシス [14]、免疫応答 [15] などへの関与が示唆されている。

　　著者は、TSPO の機能の中で中枢神経系でのステロイド生合成への関与に着目し、TSPO リガンドの新規な不安障害、うつ病を主とする精神疾患への治療薬としての可能性について検討することとした。

　　Popadopoulos らにより、Figure 1-2 に示すとおり TSPO が cholesterol の細胞質からミトコンドリア内への輸送に関与していること、ミトコンドリア内に取り込まれた cholesterol の側鎖が酵素により切断され、神経ステロイドの前駆体である pregnenolone が生成することが明らかにされた [16]。また cholesterol のミトコンドリア内への輸送が、神経ステロイドの生合成において律速段階となっていることも報告されており [17]、TSPO の活性化により、神経ステロイドの産生促進効果が期待される。

一方で、神経ステロイドの濃度変化と不安障害、うつ病との関連を示唆する臨床データも得られている。全般性不安障害の患者にて、神経ステロイドの検出量が健常人にくらべ減少していることや [18]、うつ病患者においても脊髄液中の神経ステロイド濃度が低下しており、SSRI の投与により神経ステロイド量の回復ならびに症状の改善が見られたとの報告がある [19]。

　また、神経ステロイドの投与によりげっし類のモデル動物にて抗うつ、抗不安作用を示すことや、小規模の臨床試験において神経ステロイド投与によりうつ病症状の改善が見られたとの報告もある [20]。神経ステロイドの生理機能解明のため、作用機序に関する研究も進んでおり、神経ステロイドが、GABA$_A$ 受容体、NMDA 受容体、セロトニン 3 受容体、σ1 受容体等へ作用することが明らかとなっている [21, 22]。このような背景より、神経ステロイドアナログの開発の試みも行われているが [23]、動態面および腎臓など末梢臓器での副作用など課題が多く承認には至っていない。

　上記のことから、TSPO に作用し、脳内グリア細胞にて神経ステロイドの産生を促進する化合物は、GABA$_A$ 受容体への間接的な作用により抗不安作用等の薬理作用を示すことが期待された。また、TSPO への作用により生合成が促進される神経ステロイドは、既述のとおり GABA$_A$ 受容体以外の精神疾患に関与する受容体、チャネルにも作用することが明らかになっており幅広い薬効スペクトルを示すことが期待される。さらに、臨床データとして全般性不安障害、外傷後ストレス障害の患者にて健常人に比べ血小板中の TSPO 密度の低下がみられているとの報告もある [24]。本臨床データは TSPO の機能低下と、不安障害の発症との関連性を示唆するもので、TSPO の活性化が不安症状の改善につながることが期待される。
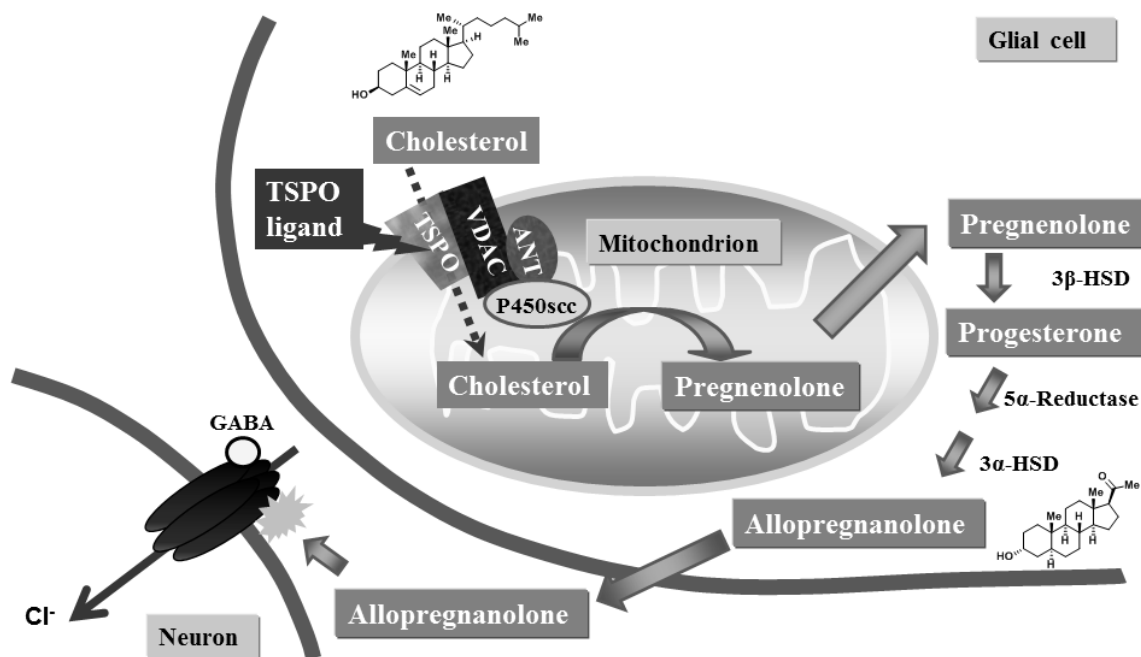

Figure 1-2. Rule of 18 kDa Translocator Protein (TSPO) in synthesis of neurosteroids

第 3 節　TSPO リガンドの開発状況

　これまで述べてきた背景により、TSPO への関心は高く、選択的な TSPO リガンドの開発研究が盛んに行われている。内因性の TSPO リガンドとしては、分子量 11 kDa のポリペプチドである Diazepame binding inhibitor (DBI)[25] あるいは Protoporphyrin IX、Heme といったポルフィリン[26] が挙げられる。DBI は、CBR に対しインバースアゴニストとして作用することが知られている。また、TSPO に作用し、ステロイド生合成を促進することが報告されている[25c]。Protoporphyrin IX、Heme はそれぞれ TSPO に対して Ki = 14.5 nM、40.6 nM と比較的強い結合活性を示す[26a]。

　既存の TSPO 合成リガンドについて代表的なものを Figure 1-3 に示す。ベンゾジアゼピン構造を有する Ro5-4864[27] は、Diazepam の 5 位ベンゼン環上に塩素基が導入された化合物である。この置換基導入により、げっし類にて TSPO 選択性が向上しており、ラット TSPO に対して Ki = 12 nM と強い結合活性を示す。しかしながら、ヒト TSPO に対する結合活性は、Ki = 400 nM と弱く種差がみられた[7a]。イソキノリン誘導体 PK11195[28] は、TSPO アンタゴニストであり、結合試験におけるラジオリガンドとして用いられている。

　これら 2 つのリガンドに関しては、結合部位に関する詳細について検討されており、結合部位に違いが見られることが報告されている。PK-11195 は TSPO にのみ結合部位として作用しているのに対し、Ro5-4864 では TSPO と VDAC の両サブユニットがリガンドの結合に関与することを示唆する実験データが得られている[29]。TSPO のアミノ酸配列についても報告されており、ヒト型に対して、マウス 82%、ラット 77% と比較的高い相同性を示すことが明らかとなっている[7a]。PK11195 は、ヒト、ラットの TSPO に対し、Ki = 4 nM、2 nM といずれにも高い結合活性を示している[7a]。Ro5-4854 と PK11195 との種差等の活性プロファイルの差はその結合様式の違いによってもたらされていると考えられる。

　DAA1097[30] は、1992 年に Okuyama らによって発表されたフェノキシアニリド誘導体である。DAA1097 は、神経ステロイド産生量を増加させることが確認されており、またマウス明暗箱試験およびラット高架式十字迷路試験にて、低用量から経口活性を示し、鎮静等のベンゾジアゼピン系薬剤でみられる副作用と十分な乖離があることが報告されている。

　Romeo らによって 1992 年に報告されたインドール誘導体 FGIN-1-27[31] は、選択的な TSPO アゴニストとして、グリア細胞およびラット脳内にて神経ステロイドの生合成を促進する[31d]。また、FGIN-1-27 は、ラット高架式十字迷路試験、ラット Vogel 型葛藤試験において、抗不安作用が確認されており、この抗不安作用が PK11195 にて拮抗されることが明らかとなっている[31c, d]。

　臨床試験において抗不安作用が報告されている AC-5216[32] については、ラット Vogel 型葛藤試験、マウス明暗箱試験、マウス社交性試験において抗不安作用が確認され、またラット強制水泳試験にて抗うつ作用を示した。さらに、ベンゾジアゼピン系薬剤で問題となっている筋弛緩作用、健忘等の副作用とも十分な乖離を示している[32a]。

DAA1097、AC-5216 の薬理作用に関する報告より、TSPO 活性化による神経ステロイドを介した間接的な GABA$_A$ 受容体への作用により、ベンゾジアゼピン系薬剤でみられる副作用を軽減しうる可能性が期待される。

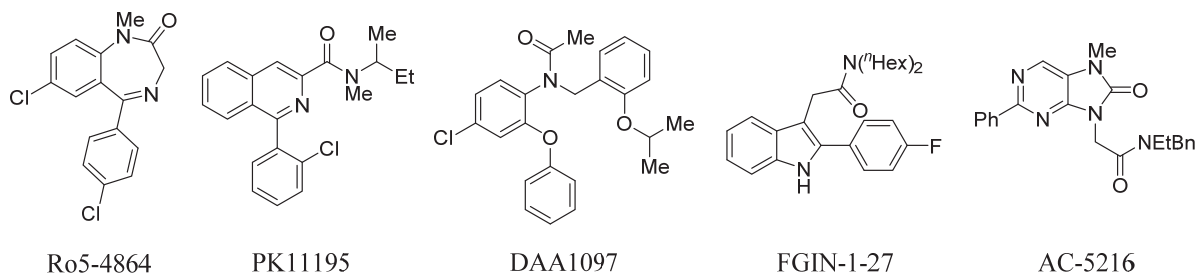| Ro5-4864 | PK11195 | DAA1097 | FGIN-1-27 | AC-5216 |

Figure 1-3. Chemical structures of representative TSPO ligands

第 4 節　新規 TSPO リガンドの探索研究

　本研究は、TSPO に着目した新規な精神疾患治療薬の創製を目的として行われた。またこれまでに多くの高活性な TSPO リガンドが報告されている一方で経口吸収性等薬物動態面における課題が明らかとなっている。このような薬物動態面での課題を克服し、TSPO 活性と薬物動態プロファイルを両立しうる化合物を取得するため検討を行った。

　第 2 章では、三環性ベンズイミダゾロン誘導体の合成、TSPO 結合活性、構造活性相関および薬物動態プロファイルについて述べる。著者は、Ro5-4864 のジアゼピノン環を開環することでデザインしたベンズイミダゾロン誘導体が、TSPO 結合活性を示すことを見出した [33]。そこで、本誘導体をもとに TSPO 活性向上が期待され、構造面での高い新規性が見込まれた三環性ベンズイミダゾロン誘導体への展開を図った (Figure 1-4)。
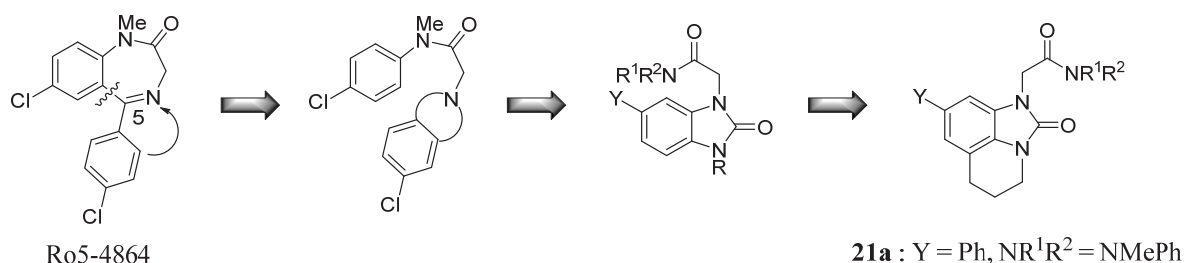

Figure 1-4. Design of benzimidazolone and tricyclic benzimidazolone derivatives as novel TSPO ligands

Ro5-4864

21a : Y = Ph, NR^{1}R^2 = NMePh

　三環性ベンズイミダゾロン誘導体における探索研究により PK11195 を上回る TSPO 結合活性を示し、CBR との選択性も十分である化合物を数多く取得した。しかし、多くの三環性ベンズイミダゾロン誘導体が、肝代謝を反映するラット肝 S-9 分画を用いた代謝実験にて、30 分後の残存率が低く肝臓で酵素による代謝を受けやすい傾向が明らかとなった。本代謝実験にて 30 分後の残存率が 12% であった化合物 **21a** について、ラット薬物動態試験を実施したが、生物学的利用率 (BA) は、1.2% と非常に低い値であったことから、ラット肝 S-9 分画を用いた代謝実験における 30 分後の残存率、すなわち代謝安定性の改善が、経口での薬効発現を目指す上で必須であると考えられた。そこで代謝安定性を改善すべく、アミド置換基、アリール置換基、さらに母骨格部分の変換を行ったが TSPO 活性と代謝安定性を両立した化合物を見出すには至っていない。本誘導体の構造活性相関に加え、代謝安定性と構造、物性パラメーターとの相関についての考察を述べる。

　第 3 章では、ベンズイミダゾロン骨格から新たに展開したベンズオキサゾロン誘導体について述べる。三環性ベンズイミダゾロン誘導体での検討結果を踏まえ、代謝安定性の観点からベンズイミダゾロン誘導体に含まれ、酸化代謝を受けやすいとされる N^3-アルキル構造を酸素原子に変

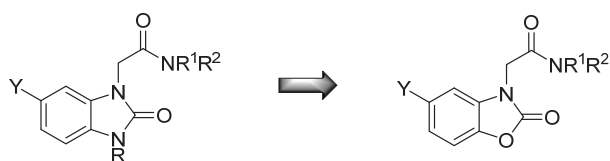換したベンズオキサゾロン誘導体を合成し、TSPO 結合活性、構造活性相関および薬物動態プロファイルについて考察した (Figure 1-5)。

Figure 1-5. Design of benzoxazolone derivatives as novel TSPO ligands

前述のとおり、Ro5-4864 において、diazepam の 5 位ベンゼン環上への塩素基の導入により TSPO への選択性が向上している。そこで Ro5-4864 の 5 位ベンゼン環上の塩素基に相当する Y 置換基の TSPO 結合活性に与える影響が予想されたことから、アリール置換基 Y の置換位置についての検討を行った。また、これまでに報告されている TSPO リガンドのファーマコファーより [34]、アミド部分も水素結合受容基として TSPO 結合活性に重要な官能基と考えられたことから、リンカー鎖長やアミド部分窒素上の置換基種について詳細な検討を行い、Figure 1-6 に示すような構造活性相関を得た。
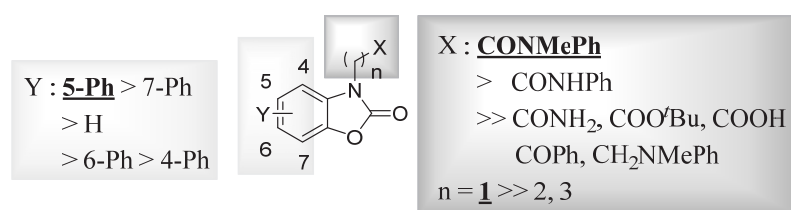
Figure 1-6. SAR of benzoxazolone derivatives for TSPO binding activity

三環性ベンズイミダゾロン誘導体における検討にて母骨格ベンジル位の代謝を抑制する目的で炭素鎖から酸素原子へ変換をしたところ代謝安定性が低下したことから、母骨格電子密度の代謝安定性への影響に着目し、置換基の電気陰性度を指標に、代謝安定性改善に向けた誘導体合成を実施した。Hammett の電子陰性度定数 (σ) を指標として導入置換基を選別し、TSPO 結合活性と代謝安定性をはじめとする薬物プロファイルがともに良好な有望化合物 **86b**、**88d** を見出した (Figure 1-7)。

代謝安定性の結果についての解析により、ベンズオキサゾロン環上の 5 位置換基においては、σ 値と相関がみられ、σ 値が大きいほど代謝安定性が高くなる傾向がみられた。一方で、AlogD7.4 と代謝安定性については明確な相関は得られなかった。

また、化合物 **88a** を用いた検討により、TSPO リガンド投与群での薬理作用が、神経ステロイドの生合成に関わる酵素に対し阻害作用を有する trilostane の投与により拮抗されたことから、薬理作用発現に神経ステロイドが関与していることが示唆された。

第 4 章では、化合物 **86b** の副作用に関する検討から明らかとなった協調運動障害の発現に関して考察し、副作用面での課題を解決し、動物モデルにて抗不安作用を示した開発候補化合物 **88d** を見出すに至るまでの検討結果、ならびに化合物 **88d** の薬理学的特性、薬物動態的特性について述べる。

　TSPO 結合活性、代謝安定性および水への溶解度等の薬物動態パラメーターをもとに化合物 **86b** を選抜し、薬物動態試験、ラット不安モデルでの薬理評価さらに中枢性副作用についての確認のためロータロッド試験を実施した。その結果、化合物 **86b** は、CBR への作用がほとんど認められないにも関わらず、抗不安作用を示した血中濃度に近い化合物濃度にて、協調運動障害を誘発した。副作用発現の原因を探るべく、化合物 **86b** について各種受容体、チャネル、トランスポーター、酵素についての結合試験を実施したところ、ラット中枢性ナトリウムチャネルへの作用が見られた (IC_{50} = 0.70 μM) 。中枢性ナトリウムチャネルの阻害により、協調運動障害が誘発されることがこれまでにも報告されていたことから、このチャネルへの作用が協調運動障害発症の一つの作用機序ととらえ、第 3 章で合成したベンズオキサゾロン誘導体について、中枢性ナトリウムチャネルのバトラコトキシン結合部位への作用についてスクリーニングを実施した。

　その結果、中枢性ナトリウムチャネルへの作用が弱く (IC_{50} > 10 μM)、TSPO 結合活性、代謝安定性および水への溶解度等の薬物動態パラメーターが良好な化合物 **88d** を見い出した。本化合物について、薬物動態試験、ラット不安モデルでの薬理評価、ベンゾジアゼピン系薬剤でみられる副作用について検討を行った結果、**88d·HCl** は、1.0 mg/kg の経口投与にて抗不安作用を示し、また副作用とも十分な乖離があることが確認された。化合物 **88d** は、TSPO 結合活性や、代謝安定性において種差検討も実施しており、新規な不安障害治療薬となりうることが期待される。

88a　　　　　　　　**86b**　　　　　　　　**88d**
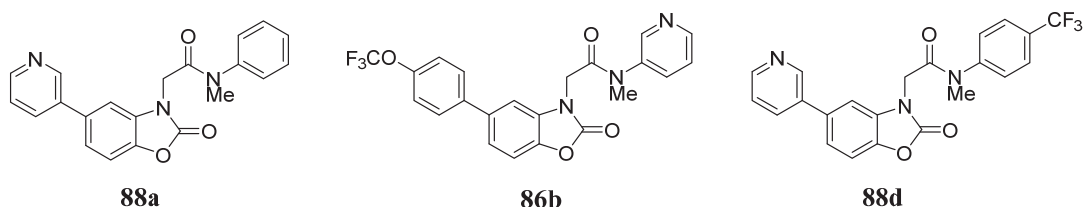
Figure 1-7. Chemical structures of representative benzoxazolone derivatives as novel TSPO ligands

　次章より、本研究の詳細について述べる。

第 2 章　三環性ベンズイミダゾロン誘導体の合成と構造活性相関

第 1 節　誘導体のデザイン

　著者は、ベンゾジアゼピン骨格を有しながら 5 位ベンゼン環上パラ位への塩素基の導入により TSPO への選択的な作用が見出されている Ro5-4864 に着目し、ジアゼピノン環の開環により新規誘導体であるベンズイミダゾロン誘導体への展開を図った (Figure 2-1)[33] 。
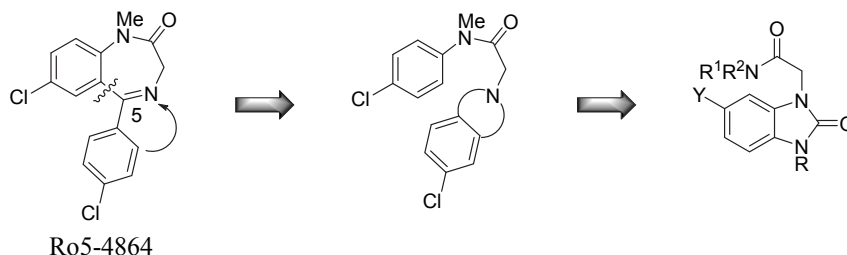
Figure 2-1. Design of benzimidazolone derivatives as novel TSPO ligands

　第 3 章にて詳細を述べるが、ベンズイミダゾロン誘導体を含む二環性誘導体について以下のような TSPO の結合活性に関する構造活性相関情報を得ている。Figure 2-2 に示すように、母骨格上のアリール基 (Y) と水素結合受容基として作用するアミド基 (X) が TSPO 結合活性に必要であり、これらの置換位置も活性に影響を及ぼしている。
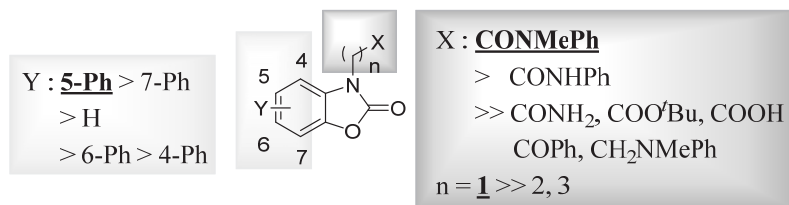
Figure 2-2. SAR of benzoxazolone derivatives for TSPO binding activity

　ここで、ベンズイミダゾロン誘導体について N^3 位にアルキル基を導入した化合物 **2** で TSPO 結合活性が向上したこと、ベンズオキサゾロン誘導体にて 7 位への置換基導入により TSPO 活性がある程度維持されている結果が得られていた。このような 2 つの知見から TSPO 活性の向上が期待され、また構造面でより高い新規性が見込まれた三環性ベンズイミダゾロン誘導体への展開について検討した (Figure 2-3) 。

1 : R = H　　TSPO Inh. at 100 nM =　86%
2 : R = Me　　TSPO Inh. at 100 nM = 100%
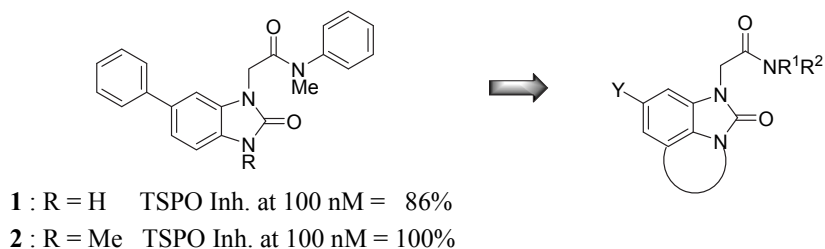
Figure 2-3. Design of novel tricyclic benzimidazolone derivatives as novel TSPO ligands

第 2 節　誘導体の合成

　評価化合物の合成は、その変換部位 (アミド置換基またはアリール置換基) に応じて、カルボン酸誘導体、ブロモアリール誘導体を鍵中間体として使い分けて行った。それぞれの中間体は、共通の化合物から導くことができ、本中間体は三環性ベンズイミダゾロン骨格を構築後、酢酸エステルユニットを導入することで得ることとした (Figure 2-4) 。
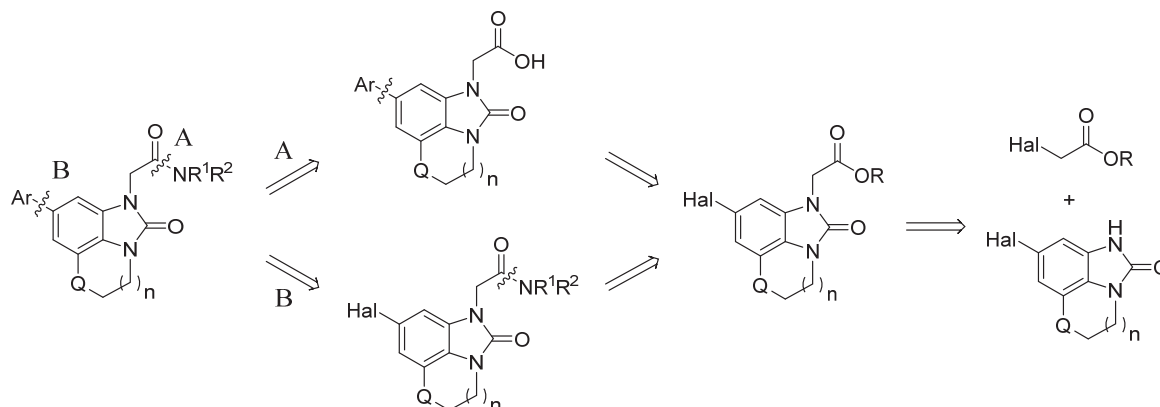
Figure 2-4. Synthetic strategy for tricyclic benzimidazolone derivatives

　8-フェニルジヒドロイミダゾキノリノン誘導体の合成法を Scheme 2-1 に示す。テトラヒドロキノリンをカルバメートとした後、ブロモ化、ニトロ化を位置選択的に行い化合物 **11** を得た。化合物 **11** を、酢酸溶媒中、80 ℃ にて鉄還元を行いニトロ基の還元に続く環化反応を進行させイミダゾキノリノン骨格を構築した。酢酸エステルを導入後、鈴木－宮浦カップリング [35] で 8－フェニル置換体 **17** とし、酸性条件下にて脱保護を行うことで得られたカルボン酸 **19** とアミンとの縮合反応により目的物であるアミド化合物 **21a-k** を得た。三環のうち一つの環を 6 員環から 7 員環へと環拡大した 9-フェニルジヒドロイミダゾベンズアゼピノン誘導体については、2,3,4,5-tetrahydro-*1H*-benzo[*b*]azepine **(6)** を出発原料とし 8-フェニルジヒドロイミダゾキノリノン誘導体と同様の合成法にて化合物 **22** を得た。

　8 位の置換基変換にはエステル **15** を酸性条件下で脱保護しカルボン酸 **23** とした後、縮合によりアミド体 **24** を得、8 位に鈴木－宮浦カップリング、Stille カップリング [36]、Buchwald らにより報告されているアミノ化 [37] によりアリール置換基を導入し、化合物 **25-32** を得た。さらに、**24** のパラジウム触媒存在下での水素添加反応により **33** を合成した (Scheme 2-2) 。
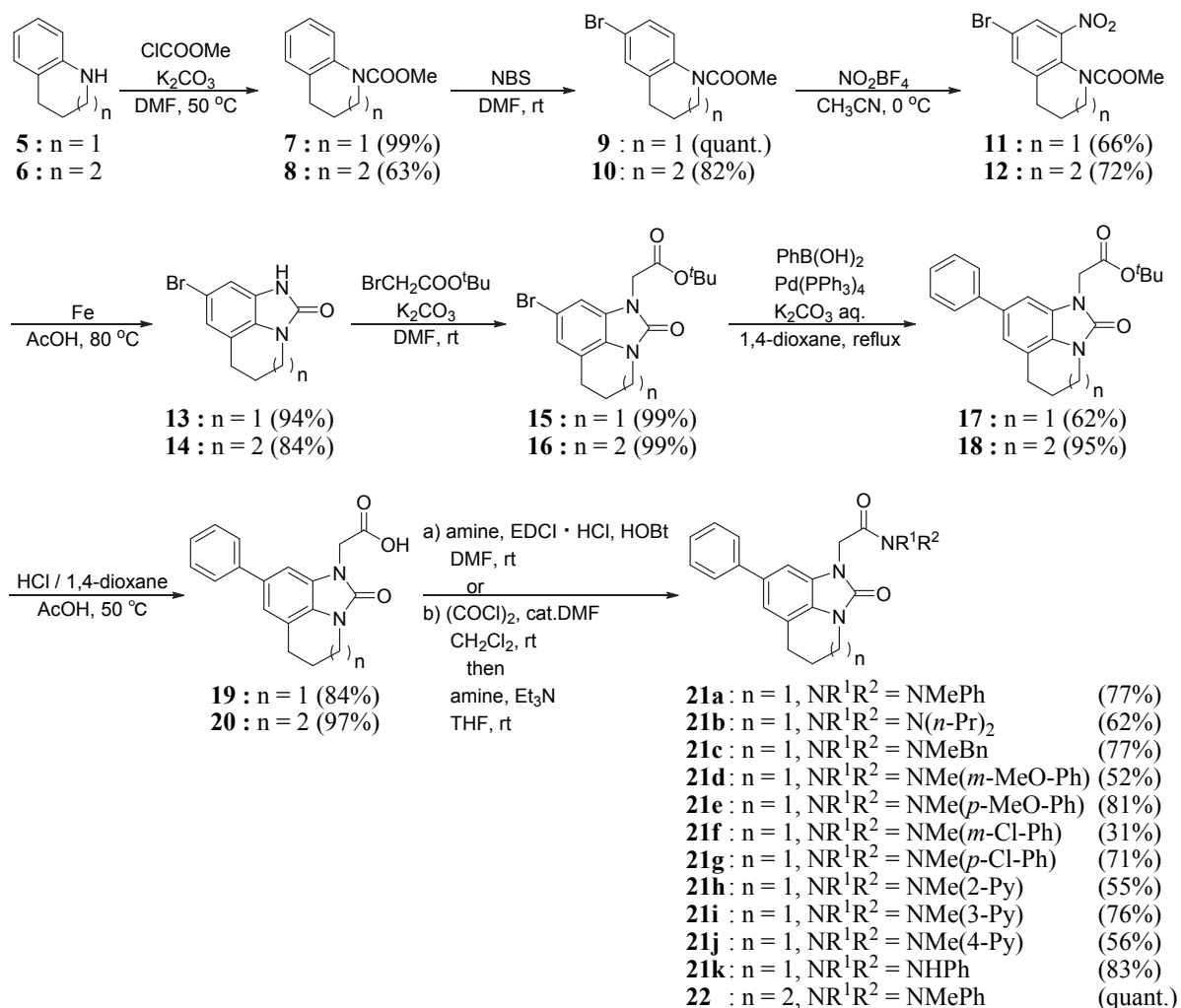
Scheme 2-1. Synthesis of dihydroimidazoquinolinone and dihydroimidazobenzepinone derivatives

25 : Y = *m*-MeO-Ph (49%)
26 : Y = *p*-MeO-Ph (77%)
27 : Y = *m*-CF₃-Ph (89%)
28 : Y = *p*-CF₃-Ph (84%)
29 : Y = 2-Py (22%)
30 : Y = 3-Py (quant.)
31 : Y = 4-Py (40%)
32 : Y = 3-PyNH (53%)
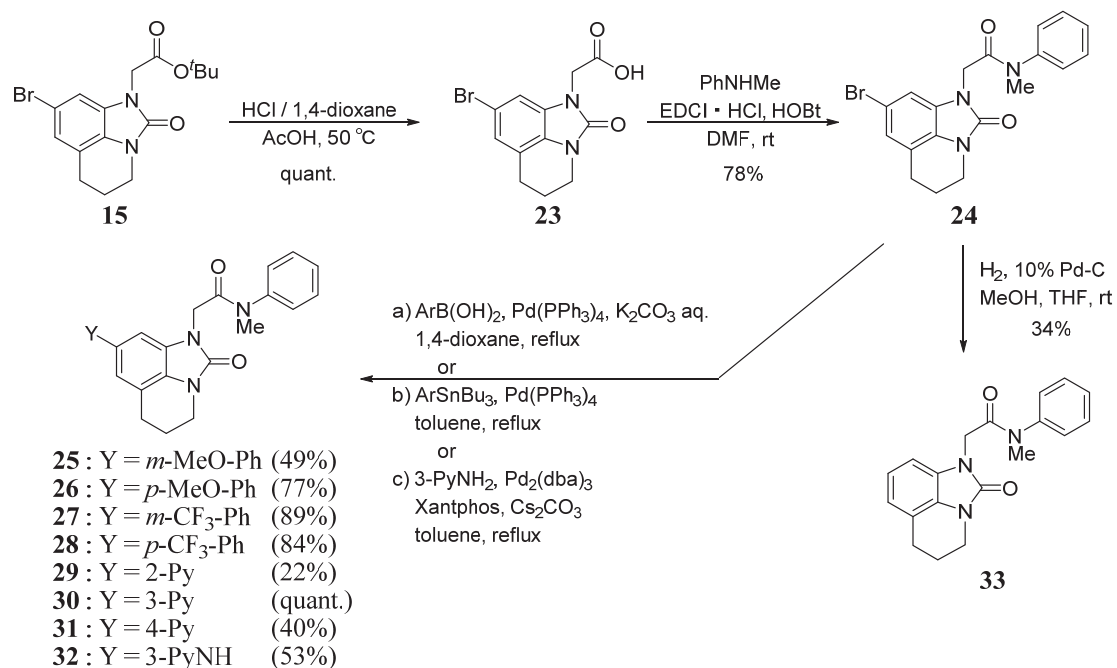
Scheme 2-2. Synthesis of dihydroimidazoquinolinone derivatives **25-33**

ジヒドロイミダゾベンズオキサジノン誘導体 **43** の合成法について Scheme 2-3 に示す。出発原料である 2-amino-3-nitrophenol **(34)** のフェノール基をアルキル化し酢酸エステルユニットを導入後、NBS によるブロモ化により化合物 **36** とした。化合物 **36** を酸触媒存在下、トルエン中加熱することで環化させオキサジノン環を構築した。鈴木－宮浦カップリングによりフェニル基を導入した際、塩基性条件下であったためオキサジノン環の開環反応が起きた。そこで再度酸触媒存在下、トルエン中加熱することで環化反応を行い化合物 **38** とした。化合物 **38** に対し LiAlH$_4$ による還元反応を行い、ニトロ基とオキサジノン環中のアミド基を同時に還元し化合物 **39** へと変換した。CDI (1,1'-carbonyldiimidazole) によるカルボニル化によりジヒドロイミダゾベンズオキサジノン骨格を形成し、酢酸エステルを導入後、酸性条件下にて脱保護を行うことで得られたカルボン酸 **42** と *N*-methylaniline との縮合反応により目的物を得た。
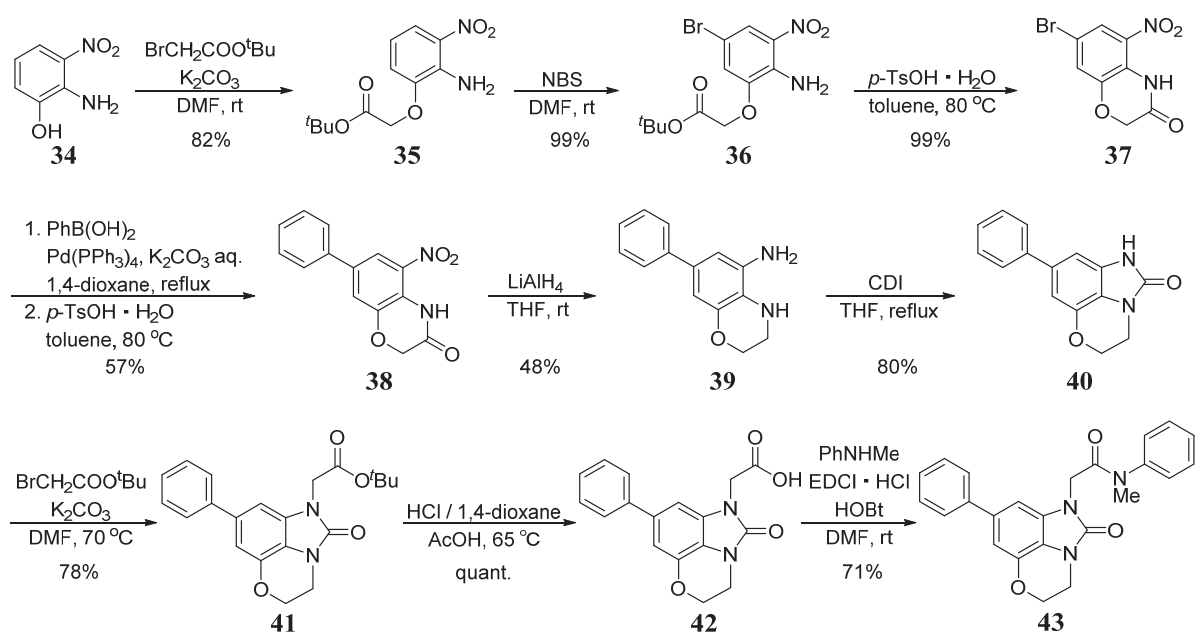
Scheme 2-3. Synthesis of dihydroimidazobenzoxazinone derivative **43**

　続いて、Scheme2-1～Scheme 2-3 にて合成した三環性ベンズイミダゾロン誘導体の TSPO 結合活性、代謝安定性および構造活性相関について述べる。

第 3 節　薬理学的評価および考察

　合成した三環性ベンズイミダゾロン誘導体の TSPO 結合活性、CBR 結合親和性、ラット肝 S-9 分画を用いた代謝安定試験結果について Table 2-1～Table 2-3 にまとめた。TSPO に対する親和性については、ラット腎臓粗ミトコンドリア膜標品を用いて、[³H]-PK11195 の特異的な結合に対する化合物の結合阻害実験を行い評価し、表中には TSPO に対する親和性を Ki 値 (nM) として示した。CBR に対する親和性については、ラット大脳皮質膜標品を用いて、[³H]-flumazenil の特異的な結合に対する化合物の結合阻害実験を行い評価し、表中には、化合物 10 μM での [³H]-flumazenil に対する結合阻害率 (%) を示した。

　Table 2-1 に示すとおり化合物 **21a** は、PK11195 よりも強く 1 μM 以下の TSPO 結合親和性を示した。三環性骨格の 1 つの環を 6 員環から 7 員環とした化合物 **22** において、約 3 倍の活性向上が見られた (Ki = 0.32 nM) 。また、第 4 節にて詳細に述べるが、代謝安定性を改善する試みの一つとしてベンジル位の酸化代謝が抑制されることを期待し [38]、メチレンリンカーを酸素原子に置換させた **43** は非常に強い活性を示したが、目的とする代謝安定性改善は達成されなかった。骨格の異なる 3 化合物とも高い TSPO 活性を示したが、より良好な代謝安定性を示した化合物 **21a** をリード化合物とし、TSPO 活性と薬物動態プロファイルを両立する化合物を得ることを目標にアミド部分、アリール置換基の変換を行った。

Table 2-1. In vitro profile of tricyclic benzimidazolone derivatives
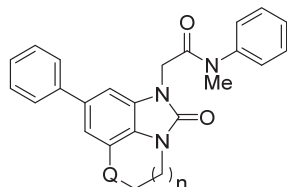
Compd	n	Q	TSPO Ki[a] (nM)	CBR inhibition[b] (%)	Metabolic stability[c] (remaining%)
21a	1	CH_2	0.94	14	12
22	2	CH_2	0.32	30	1
43	1	O	0.21	68	1
PK11195			1.7		
Ro5-4864			6.8		

[a] Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. [b] Percent inhibition of [³H]-flumazenil specific binding at 10 μM of the compound. [c] Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 °C. The initial concentration of each compound was 1.0 μM.

13

Table 2-2. In vitro profile of dihydroimidazoquinolinone derivatives with various amide moieties
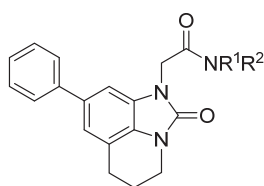
Compd	R^1	R^2	TSPO Ki^a (nM)	CBR inhibition^b (%)	Metabolic stability^c (remaining%)	AlogD7.4^e
21a	Me	Ph	0.94	14	12	3.89
21b	*n*-Pr	*n*-Pr	1.7	11	1	4.06
21c	Me	Bn	0.23	72	1	3.90
21d	Me	*m*-MeO-Ph	0.28	6	0	3.88
21e	Me	*p*-MeO-Ph	0.73	3	0^d	3.88
21f	Me	*m*-Cl-Ph	0.44	3	0	4.56
21g	Me	*p*-Cl-Ph	0.13	4	0	4.56
21h	Me	2-Py	2.5	34	0	3.28
21i	Me	3-Py	2.1	36	0	2.74
21j	Me	4-Py	15	24	0	2.74
21k	H	Ph	33	27	62	3.69

^{a} Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. ^{b} Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound. ^{c} Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 ^{o}C. The initial concentration of each compound was 1.0 μM. ^{d} Metabolic stability determined at 10 μM of the compound. ^{e} Predicted with PipelinePilot, version 8.0.1.

　アミド部分の置換基変換による TSPO 結合活性や代謝安定性の結果について Table 2-2 に示す。アリール基をフェニル基に固定しアミド部分を、アルキル-フェニルタイプから、ジアルキルタイプ、アルキル-ベンジルタイプへと変換したところ、対応する化合物 **21b**、**21c** において、Ki = 1.7 nM、0.23 nM とともに高い TSPO 結合活性を示し、特に **21c** については、化合物 **21a** に比べ 4 倍程度の活性向上が見られた。しかし、代謝安定性についていずれの化合物も低下したこと、アルキル-ベンジルタイプの化合物 **21c** では CBR に対する結合活性が若干強くなったことから **21a** に代表されるアルキル-フェニルタイプについて置換基の導入検討を行うこととした。電子供与性基としてメトキシ基、電子吸引性基としてクロル基を導入したところ、いずれも化合物 **21a** の TSPO 結合活性を上回った。メトキシ基についてはメタ位 (**21d**)、クロル基についてはパラ位 (**21g**) への置換基導入にてより活性の向上が顕著であった。代謝安定性向上を期待し電子吸引性基を導入した誘導体 (**21f** 、**21g**)、ベンゼン環パラ位へ置換基を導入した誘導体 (**21e**、**21g**) ともに代謝安定性改善には至らなかった。そこで、次に脂溶性の軽減、水溶性向上を目指しアミド部分のフェニル基からピリジル基への変換体について評価した。薬理評価結果よりピリジン誘導体 (**21h-j**) では塩基性窒素の位置が TSPO 活性に大きな影響を与えることが明らかとなった。すなわち 4-ピリジル体 **21j** では活性が **21 a** に比べ 10 倍以上低下した一方で、化合物

21h、**21i** では 2 倍程度の活性減弱は見られたものの高い TSPO 活性が維持された。しかしいずれのピリジン誘導体においても代謝安定性は改善されなかった。一方で、in vitro の代謝実験において、代謝体の分子量より脱メチル体と推定される化合物が検出されたことから 2 級アミド体である化合物 **21k** を合成し評価したところ、代謝安定性については大幅に改善されたが論文報告通り [39] その TSPO 活性は大きく減弱した (Ki = 33 nM)。以上のとおり、アミド部分の検討にて TSPO 活性の向上は達成できたが、課題であった代謝安定性については TSPO 活性が減弱した 2 級アミドを除いて改善した化合物が得られなかった。続いて、アリール置換基部分について検討を実施した。

Table 2-3. In vitro profile of dihydroimidazoquinolinone derivatives with various substituents at the C-8

Compd	Y	TSPO Ki^a (nM)	CBR inhibition^b (%)	Metabolic stability^c (remaining%)	AlogD7.4^e
33	H	2.5	0	0^d	2.38
24	Br	0.057	5	0^d	3.12
21a	Ph	0.94	14	12	3.89
25	*m*-MeO-Ph	0.11	9	1	3.92
26	*p*-MeO-Ph	0.30	5	2^d	3.88
27	*m*-CF$_3$-Ph	0.23	3	0	4.83
28	*p*-CF$_3$-Ph	0.16	0	0	4.84
29	2-Py	2.7	48	0	3.17
30	3-Py	3.3	47	0^d	2.74
31	4-Py	0.55	41	3	2.76
32	3-PyNH	2.0	70	41	2.81

^{a} Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. ^{b} Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound. ^{c} Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 ^{o}C. The initial concentration of each compound was 1.0 μM. ^{d} Metabolic stability determined at 10 μM of the compound. ^{e} Predicted with PipelinePilot, version 8.0.1.

　アリール置換基部分についての薬理評価結果について Table 2-3 にまとめた。中間体であるブロモ体 **24** では、予想に反し高い TSPO への結合活性を示した。また、ベンゼン環へのメトキシ基やトリフルオロメチル基の導入により高活性の化合物 **25-28** を得た。電子供与性基であるメトキシ基、電子吸引性基であるトリフルロメチル基を導入した誘導体においてともに TSPO 活性が向上していることから、8 位のベンゼン環上の電子密度は TSPO 活性に大きな影響を与えていないと推測された。ベンゼン環をピリジン環に変換した化合物 **29-31** では高い TSPO 活性が維

持され特に 4-ピリジン体 **31** では **21a** よりも強い活性を示した (Ki = 0.55 nM)。 このように、8 位置換基変換により高活性の化合物を多く得られたものの、アミド部分の変換体と同様いずれの化合物も低い代謝安定性を示した。アミノリンカーを挟んで 3-ピリジン環が置換した **32** において代謝安定性の改善が見られたものの、化合物 **32** は一部の代謝酵素に対し強い酵素阻害能 (CYP2C19、79% inh. at 1.0 μM) を有しており、他の薬物との相互作用が懸念されることからこれ以上の評価は実施しなかった。

第 4 節　薬物動態プロファイル改善に向けた取組みおよび考察

　　三環性ベンズイミダゾロン誘導体における検討では、PK11195 の結合活性を上回りまた CBR に対する選択性も十分であるリード化合物 **21a** を取得した。化合物 **21a** についてラットを用いた薬物動態試験を実施したところ、生物学的利用能 (BA) が非常に低い結果 (1.2%) であった (Table 2-4)。BA は、Figure 2-5 に示すとおり消化管管腔からの吸収、小腸代謝、肝臓代謝の 3 要素の積として算出される。Table 2-5 にまとめた in vitro の薬物動態評価結果より、化合物 **21a** は肝代謝を反映するラット肝 S-9 分画を用いた代謝実験において、30 分後の残存率が 12% と肝臓で酵素による代謝を受けやすいことがわかった。また Caco-2 細胞を用いた透過係数が吸収に十分な値を示していることから、BA が低い主な要因としては肝臓において酵素による代謝を受けやすいことが示唆された。

Table 2-4. Pharmacokinetic properties of **21a**^{a}

Dose (mg/kg)	Dose (mg/kg)	AUC (μg·h/mL)	CL (mL/min/kg)	T1/2 (min)	Vdss (L/kg)	Cmax (ng/mL)	Tmax (min)	F (%)
1 (iv)	1	6.66	134	18.3	3.5			
10 (po)	10	8.30				26.0	15	1.2

^{a}Each value represents the mean of two or three rats

Table 2-5. In vitro harmacokinetic properties of **21a**

logP*	Solubility pH 7.4	(μg/kg) pH 2.5	Metabolic stability^{c} (remaining%)	Caco-2 permeability Pappi (nm/sec)
>4.0	1	3	12	138

$$Bioavailability\,(BA) = Fa \times Fg \times Fh$$
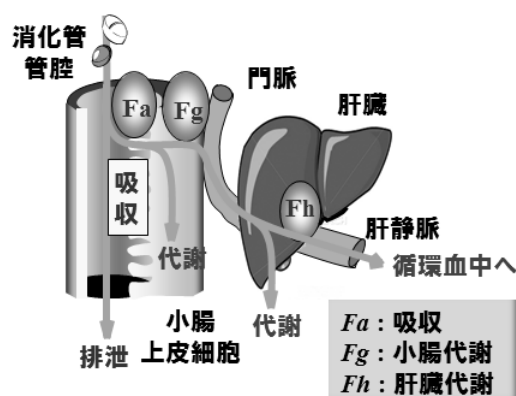
Figure 2-5. Factors of bioavailability

化合物 **21a** の薬物動態試験の結果を踏まえ代謝安定性改善に向け、以下の 5 つの項目について取り組みを行った。結果としては、第 3 節にて既に述べたとおり、十分な BA が期待できるまで代謝安定性が向上した誘導体はほとんど得られてはいないが、他の誘導体に三環性ベンズイミダゾロン誘導体で得られた情報を生かすため、各パラメーターとの相関に関する検証や得られた代謝安定性についてのデータと置換基種または構造との関連性の有無について考察を行った。

1) 代謝実験の結果の活用

　脱メチル体と推定される化合物が in vitro での代謝実験により検出されたことから、2 級アミド誘導体 **21k** について評価したところ、代謝安定性は大きく向上した (62%) ものの活性 (Ki = 33 nM) との両立が達成できなかった。

2) 代謝推定部位の構造変換

　酸化代謝を受けやすいと推定とされたベンジル位を酸素リンカーに変換したが改善は見られなかった (**21a**, 12%; **43**, 1%) 。

3) 脂溶性低減

　AlogD7.4 と代謝安定性との相関を Figure 2-6 に示したがその相関は得られなかった。
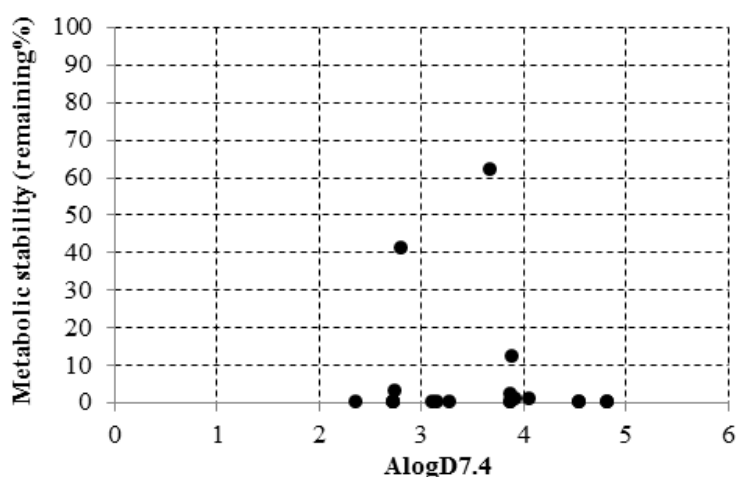
Figure 2-6. Metabolic stability vs AlogD7.4

4) 置換基導入による代謝反応の抑制

　電子供与性基が置換したベンゼン環上のパラ位は一般的に酸化代謝を受けやすいことが知られており[38]代謝をうけやすい部位への置換基導入にて代謝反応が抑制されることを期待したが改善はみられなかった (**21e**, 0%; **21g**, 0%; **26**, 2%; **28**, 0%)。

5) 電子吸引性基の導入によるベンゼン環の酸化反応抑制

　ベンゼン環上の電子密度を下げることで、CYP P450 との相互作用を弱め代謝反応が抑制されることが知られており[38]、三環性ベンズイミダゾロン誘導体についても検証したが期待した代謝安定性の改善効果は見られなかった(**21f**, 0%; **21g**, 0%; **27**, 0%; **28**, 0%)。

18
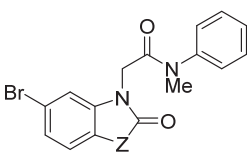
以上示したとおり三環性ベンズイミダゾロン誘導体における検討により、構造面の新規性が高くまた報告されている既存の TSPO リガンドに比べても非常に高い TSPO 結合活性を示す誘導体を数多く取得することができた。しかしながら、不安障害治療薬をめざす上では経口剤としての開発は必須であり、三環性ベンズイミダゾロン誘導体においては、TSPO 活性を維持し代謝安定性といった薬物動態面での課題が改善された誘導体を得ることはできなかった。そこで、三環性ベンズイミダゾロン誘導体での検討にて TSPO 活性、代謝安定性について得られた知見を活用し新たな化合物のデザイン、誘導体合成に取り組むこととした。

第 3 章　ベンズオキサゾロン誘導体の合成と構造活性相関

第 1 節　誘導体のデザイン

　三環性ベンズイミダゾロン誘導体での結果を受け、代謝安定性向上を目的とし中心骨格の変換について検討を行った。三環性ベンズイミダゾロン誘導体に含まれる N^3-アルキル構造は酸化代謝を受けやすいことが報告されており [38c]、アミド部やアリール置換基の変換が代謝安定性改善にほとんど寄与しなかった点から N^3-アルキル構造についての変換を試みた。ベンズイミダゾロン骨格の N-メチル基を酸素原子、硫黄原子へと変換したベンズオキサゾロン誘導体、ベンズチアゾロン誘導体について評価したところ TSPO 結合活性はほぼ同等でいずれも代謝安定性の向上が見られた (Table 3-1)。

Table 3-1. TSPO binding affinity and metabolic stability

Compd	Z	TSPO Inh. (%)[a] at 100 nM	Metabolic stability[b] (remaining%)
3	NMe	98	0
4	S	98	10
80	O	100	23

[a] Percent inhibition of [^{3}H]-PK11195 specific binding at 100 nM of the compound. [b] Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 ^{o}C. The initial concentration of each compound was 1.0 μM.

　Table 3-1 の結果をふまえ、代謝安定性の観点からよりベンズオキサゾロン誘導体を選抜し、さらなる検討に着手した (Figure 3-1)。
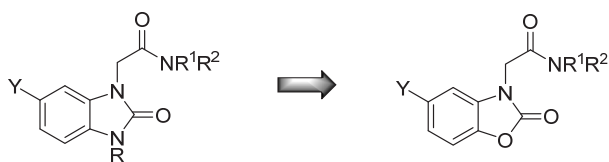
Figure 3-1. Design of benzoxazolone derivatives

第 2 節　誘導体の合成

　評価化合物の合成は、三環性ベンズイミダゾロン誘導体と同様にその変換部位に応じて、カルボン酸誘導体、ブロモアリール誘導体を鍵中間体として使い分けて行った。それぞれの中間体は、共通の化合物から導くことができ、共通中間体はベンズオキサゾロン骨格を構築後、酢酸エステルユニットの導入にて導くこととした (Figure 3-2) 。
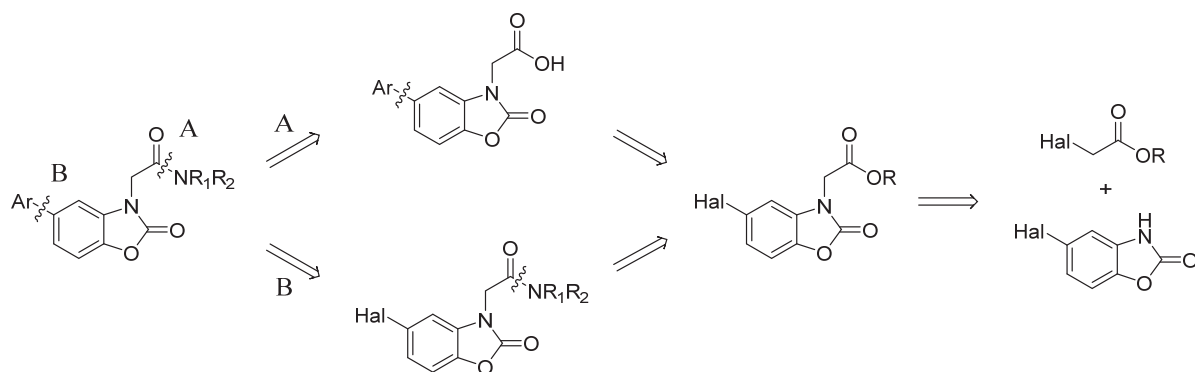
Figure 3-2. Synthetic strategy for benzoxazolone derivatives

　Scheme 3-1 に、5-フェニルベンズオキサゾロン誘導体の合成法を示す。化合物 **44** を出発原料とし CDI によるカルボニル化にてベンズオキサゾロン環を構築し化合物 **45** を得た。化合物 **45** に酢酸エステルを導入後、酸性条件下にて脱保護を行うことで得られたカルボン酸とアミンとの縮合反応により目的物とするアミド誘導体 **48a-l** を得た。

48a : $NR^1R^2 = NMePh$	(82%)
48b : $NR^1R^2 = NH_2$	(98%)
48c : $NR^1R^2 = NHPh$	(55%)
48d : $NR^1R^2 = N(n\text{-}Pr)_2$	(62%)
48e : $NR^1R^2 = NMeBn$	(54%)
48f : $NR^1R^2 = NMe(m\text{-}MeO\text{-}Ph)$	(50%)
48g : $NR^1R^2 = NMe(p\text{-}MeO\text{-}Ph)$	(81%)
48h : $NR^1R^2 = NMe(m\text{-}Cl\text{-}Ph)$	(30%)
48i : $NR^1R^2 = NMe(p\text{-}Cl\text{-}Ph)$	(72%)
48j : $NR^1R^2 = NMe(2\text{-}Py)$	(42%)
48k : $NR^1R^2 = NMe(3\text{-}Py)$	(39%)
48l : $NR^1R^2 = NMe(4\text{-}Py)$	(86%)
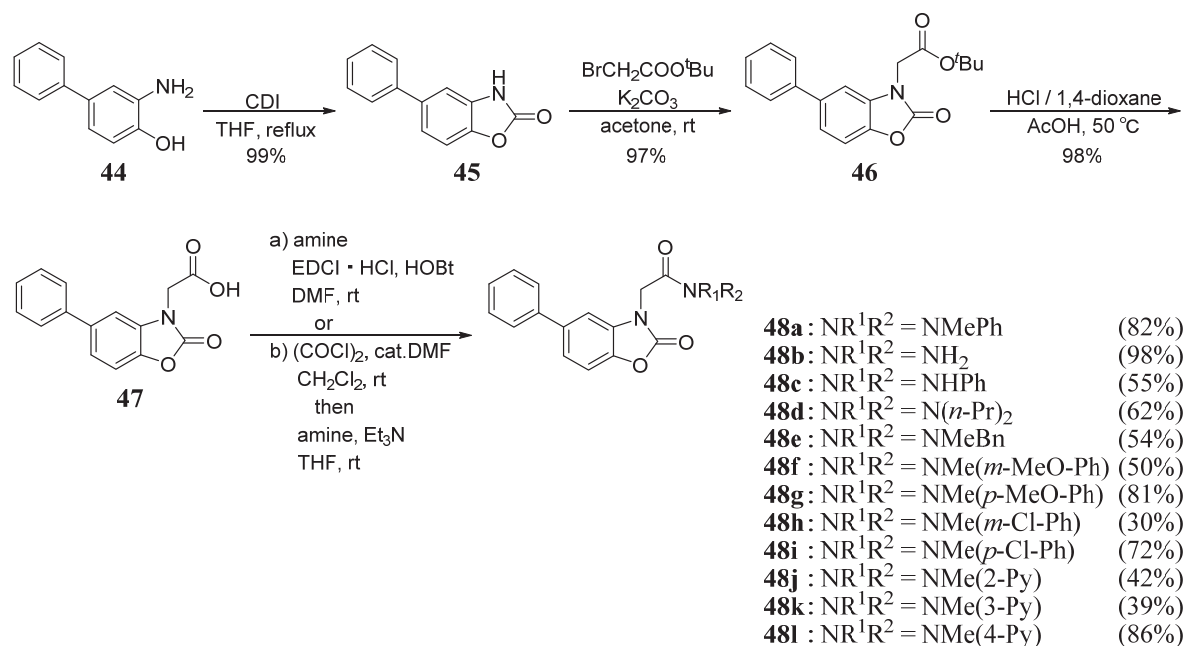
Scheme 3-1. Synthesis of 5-phenyl benzoxazolone derivatives **48a-l**

縮合反応後にアミド部分に変換が必要な 5-フェニルベンズオキサゾロン誘導体 **48m-o** につい
ては、Scheme 3-2 に示すとおりアルコール部分をブロモ基へと変換後ジメチルアミンのアルキル
化反応にて合成した。
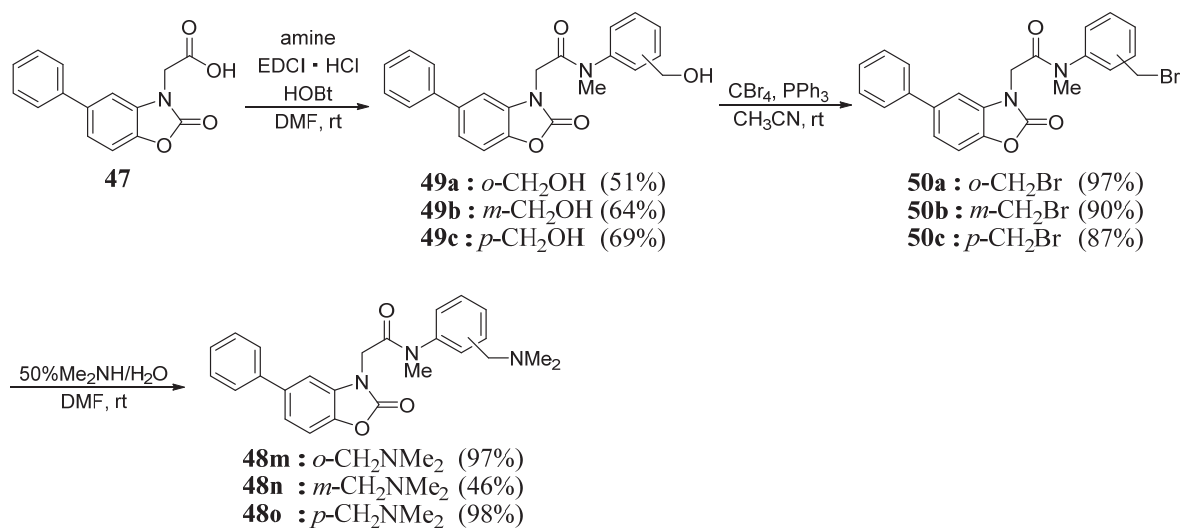
Scheme 3-2. Synthesis of 5-phenyl benzoxazolone derivatives **48m-o**

　アセトアミド部分の変換体については Scheme 3-3 に示すとおり合成した。すなわち、化合物
45 に対し、炭酸カリウム存在下アルキル化することで化合物 **51**、**52** を合成した。また、ケトン
体 **53** についてもブロモアセトフェノンを用いて同様のアルキル化反応にて目的物を得た。
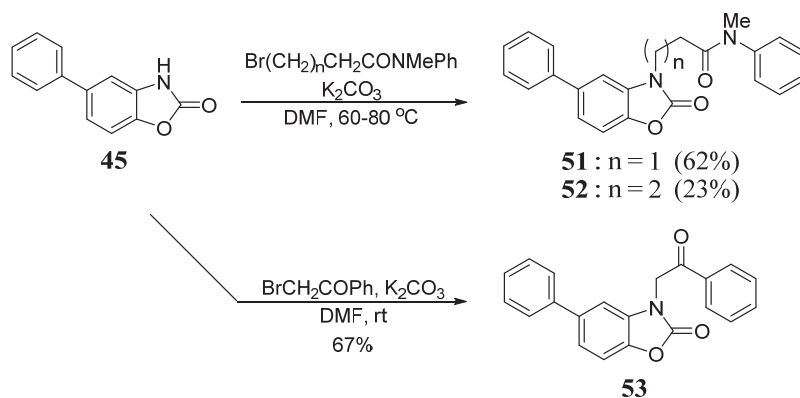
Scheme 3-3. Synthesis of benzoxazolone derivatives **51-53**

　アセトアミド基をアミノ基へと変換した化合物 **54** は化合物 **48a** のボラン還元によって得た。
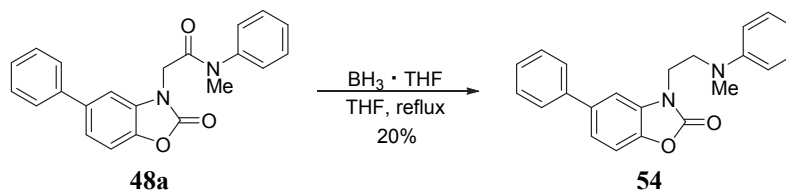
Scheme 3-4. Synthesis of benzoxazolone derivative **54**

ベンゼン環が 4 位、6 位に置換した誘導体の合成について Scheme 3-5 に示す。出発原料である 3-phenylphenol (55) のニトロ化[40]により 3 種の位置異性体 56 (14%)、57 (26%)[41]、58 (41%)[42] を得た。カラム精製にてそれぞれ単離し、4 –フェニルベンズオキサゾロン誘導体については、化合物 56 から目的物を得た。すなわち ニトロ基をパラジウム触媒存在下、水素添加反応にて還元し、化合物 59 を得た。続いて CDI によるカルボニル化にてベンズオキサゾロン環を構築し化合物 61 とした。化合物 61 に酢酸エステルを導入後、酸性条件下にて脱保護を行うことで得られたカルボン酸 65 と N-methylaniline との縮合反応により目的物 67 を得た。ベンゼン環が 6 位に置換した誘導体 68 については、57 を出発原料として水素添加反応にて化合物 60[43] とした後、67 と同様の方法にて合成した。
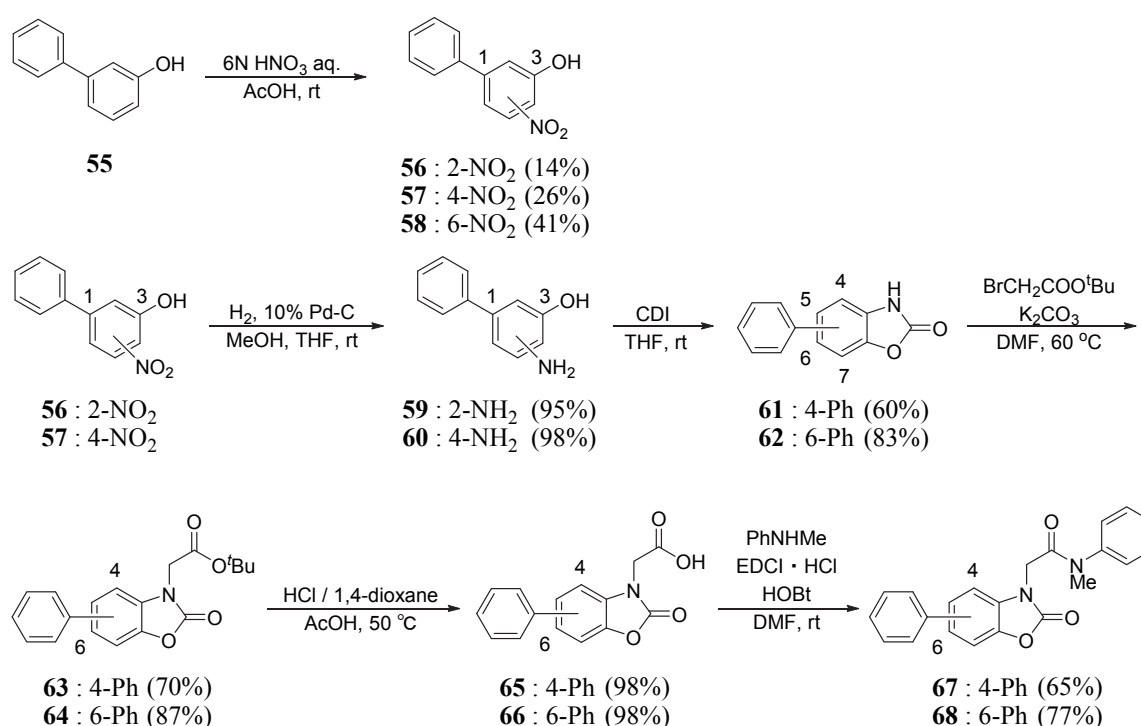
Scheme 3-5. Synthesis of 4-phenyl benzoxazolone derivative and 6-phenyl benzoxazolone derivative

ベンゼン環が 7 位に置換した誘導体 75 は、2-bromo-6-nitrophenol (69) を出発原料とし、鉄還元後、CDI によるカルボニル化にてベンズオキサゾロン骨格を構築した。化合物 71 に酢酸エステルを導入後、酸性条件下にて脱保護を行うことで得られたカルボン酸 73 と N-methylaniline との縮合反応により化合物 74 を得た。化合物 74 とフェニルボロン酸との鈴木-宮浦カップリング反応により化合物 75 を合成した (Scheme 3-6) 。
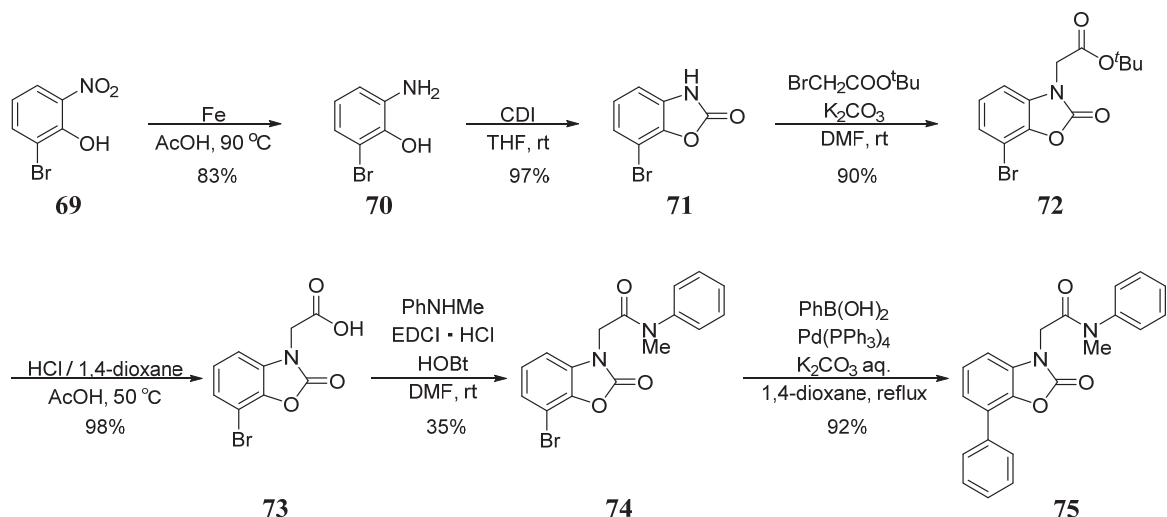
Scheme 3-6. Synthesis of 7-phenyl benzoxazolone derivative **75**

Schemes 3-7、3-8 に 5 位置換基変換誘導体の合成を示す。化合物 **76** のロジウム炭素を用いた水素添加反応[44]によりニトロ基を選択的に還元し化合物 **77**[45] を得た。続いて、CDI によるカルボニル化にてベンズオキサゾロン骨格を構築した。化合物 **78** に酢酸エステルを導入後、酸性条件下にて脱保護を行うことで得られたカルボン酸 **80** と *N*-methylaniline との縮合反応により化合物 **81** を合成した。化合物 **81** とボロン酸試薬またはスズ試薬とのカップリング反応により目的とする誘導体を得た。

82	: Y = *m*-MeO-Ph	(53%)
83	: Y = *p*-MeO-Ph	(80%)
84	: Y = *m*-CF$_3$-Ph	(73%)
85a	: Y = *p*-CF$_3$-Ph	(58%)
86a	: Y = *p*-CF$_3$O-Ph	(66%)
87	: Y = 2-Py	(73%)
88a	: Y = 3-Py	(80%)
89a	: Y = 4-Py	(68%)
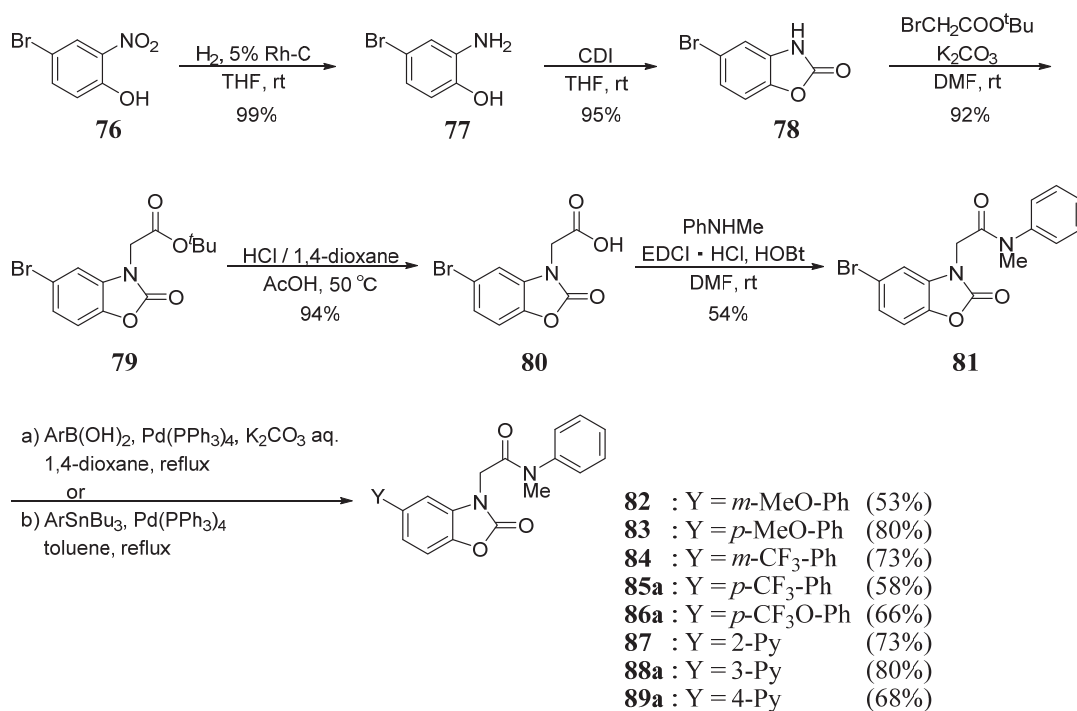
Scheme 3-7. Synthesis of benzoxazolone derivatives with various substituents at the C-5 position

ベンズオキサゾロン誘導体の 5 位置換基変換体合成をより効率的に行うため、中間体 **81** をボロン酸エステルへと変換した **91** を合成した [46]。化合物 **91** の活用によりボロン酸試薬が入手できないものについてボロン酸試薬の調製を経ずにブロモアリール化合物を用いて目的とする誘導体へと導くことができるため、効率的な化合物合成が可能となった。また、2-ピリジル体 **87** の合成には、カップリング反応にてスズ試薬を用いていたが、中間体 **91** を用いることで有害なスズ試薬の利用を回避し目的物を合成することが可能となった。
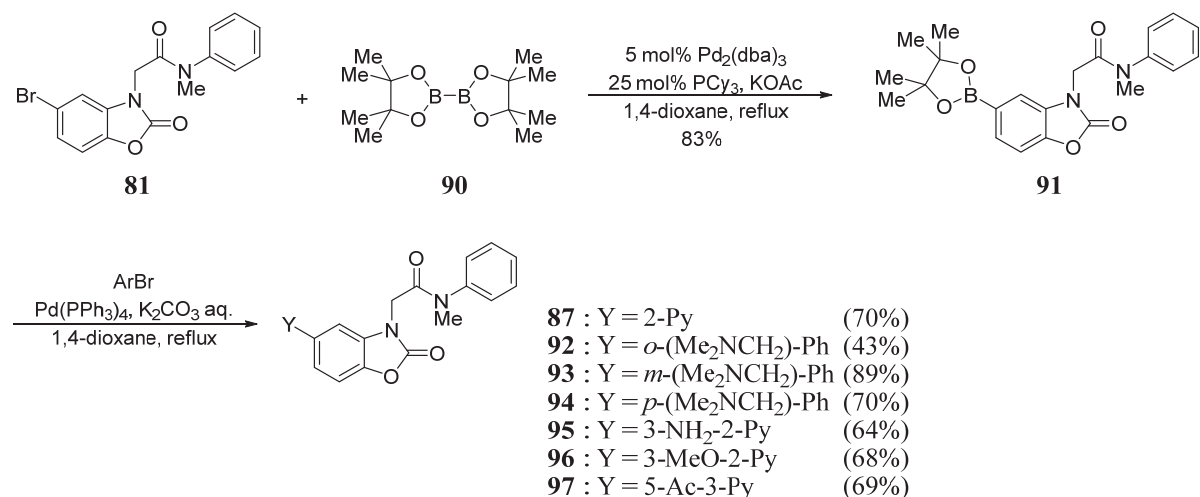
Scheme 3-8. Synthesis of benzoxazolone derivatives with various substituents at the C-5 position

　化合物 **98-100** の合成については、Scheme 3-9 に示すとおり行った。中間体 **81** の Ullmann カップリング反応 [47] により **98** を、Buchwald らによって報告された条件でのアミノ化反応 [37] により化合物 **99** を合成した。また、5 位脱ブロモ体 **100** については、**81** のパラジウム触媒存在下での水素添加反応により合成した。
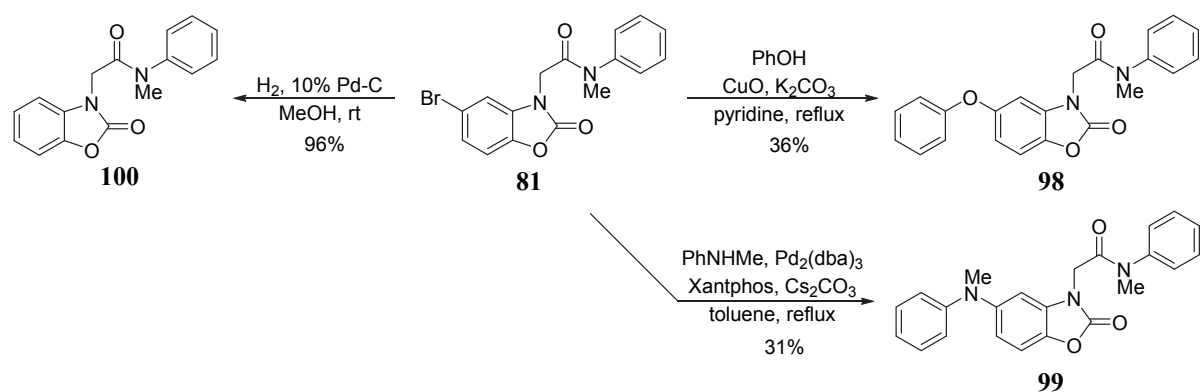
Scheme 3-9. Synthesis of benzoxazolone derivatives with various substituents at the C-5 position

25

5 位置換基としてニトリル基が置換した誘導体 **102** は、以下のとおり合成した (Scheme 3-10)。
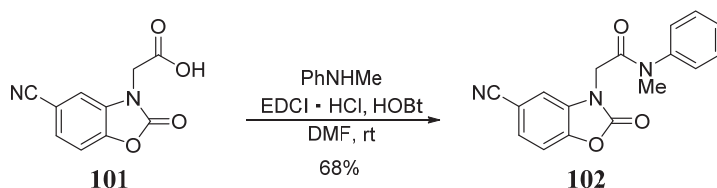
Scheme 3-10. Synthesis of benzoxazolone derivative **102**

アミド置換基と 5 位置換基についてそれぞれ変換した誘導体の合成について Scheme 3-11 に示す。中間体である **79** とボロン酸試薬との鈴木-宮浦カップリング反応により化合物 **103-106** を合成した後、酸性条件下にて脱保護を行うことで得られたカルボン酸 **107-110** とアミンとの縮合反応により誘導体 **85b**、**86b**、**88b-e**、**89b-e** を合成した。
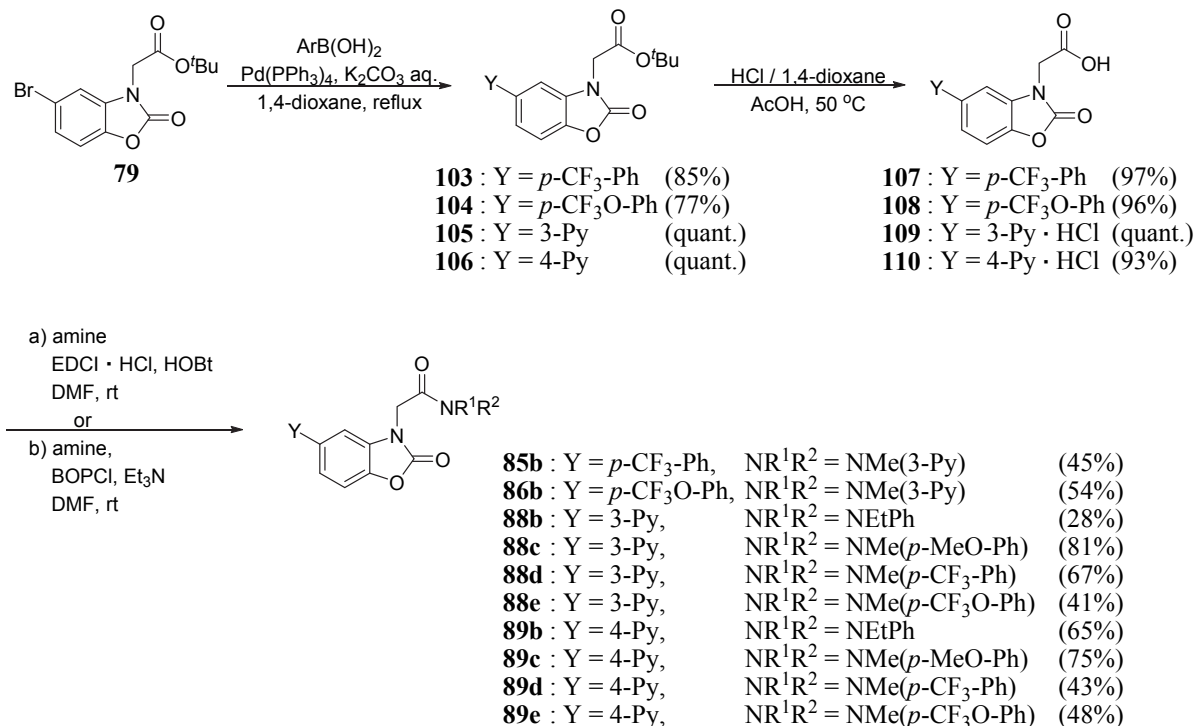
Scheme 3-11. Synthesis of benzoxazolone derivatives **85b**, **86b**, **88b-e** and **89b-e**

第 3 節　薬理学的評価および考察

　　合成したベンズオキサゾロン誘導体の TSPO 結合活性、構造活性相関および薬物動態プロファイルについて考察する。

　　水素結合受容基部分の検討結果について以下の Table 3-2 に示す。ベンズオキサゾロン骨格の窒素上置換基として、アセトアミド構造を有する化合物 **48a** が強い活性を示す一方で、酢酸エステル **46**、酢酸 **47**、ケトン **53** ではその TSPO 活性はほぼ消失した。アセトアミド部分に関しては、TSPO との作用において、水素結合受容基としての役割とアミド窒素原子上の置換基による疎水性ポケットへの相互作用という 2 種の異なる作用に関わっていると考えられる[34]。アセトアミド構造の水素結合受容基としての TSPO 親和性への寄与を裏付ける結果として、アミド構造のカルボニル基を除去した化合物 **54** で TSPO 活性が減弱することや、ベンズオキサゾロン骨格とアミド構造との炭素鎖長を変えることで TSPO 活性が大きく変化するといったデータが得られている。また、アセトアミド部分の疎水性ポケットとの相互作用については、カルバモイル体 **48b**、2 級アミド体 **48c** にて活性が減弱する点から 3 級アミドが適していると考えられる[39]。

Table 3-2. TSPO binding affinity of benzoxazolone derivatives with various hydrogen bond acceptor parts
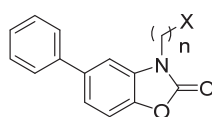
Compd	n	X	TSPO Inh. (%)^a at 100 nM
48a	1	CONMePh	98 (Ki^b = 1.6 nM)
46	1	COO^{t}Bu	0
47	1	COOH	0
53	1	COPh	14
54	1	CH$_2$NMePh	0
48b	1	CONH$_2$	3
48c	1	CONHPh	19
51	2	CONMePh	6
52	3	CONMePh	0

^{a} Percent inhibition of [^{3}H]-PK11195 specific binding at 100 nM of the compound. ^{b} Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound.

　　次に、ベンズオキサゾロン骨格のベンゼン環上のアリール置換基の位置についての最適化を行った。Ro5-4864 において、5 位ベンゼン環パラ位への塩素基の導入により TSPO への選択的な作用が見出されている。よって、ベンズオキサゾロン骨格上のアリール置換基が、TSPO との相互作用において重要であると考えられた。Table 3-3 に示すとおり、ベンゼン環の置換位置により

その TSPO 結合活性は大きく変動し、5 位置換体 **48a** においてもっとも強い活性を示すことが明らかとなった。

Table 3-3. TSPO binding affinity of benzoxazolone derivatives with different substation site
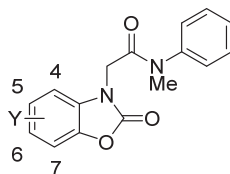
Compd	Y	TSPO Ki^a (nM)
100	H	11
67	4-Ph	120
48a	5-Ph	1.6
68	6-Ph	29
75	7-Ph	9.0

^{a} Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound.

アセトアミド部分、アリール置換基の置換位置に関する検討により得られた、TSPO 結合活性に関する構造活性相関について Figure 3-3 にまとめる。
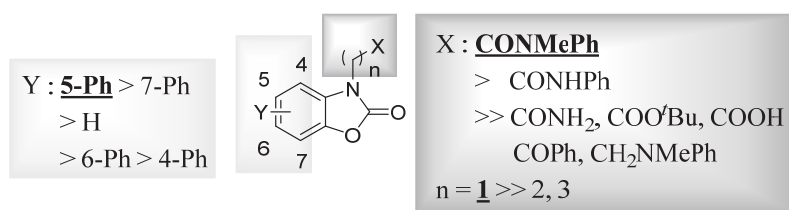
Figure 3-3. SAR of benzoxazolone derivatives for TSPO binding activity

上記の結果をもとに 5-アリールベンズオキサゾロンを中心とした最適化検討に取り組むこととした。アミド部分の置換基変換による検討結果を Table 3-4 に示す。アリール基をフェニル基に固定しアミド部分を、アルキル-フェニルタイプから、ジアルキルタイプ、アルキル-ベンジルタイプへと変換したところ、対応する化合物 **48d**、**48e** において、Ki = 17 nM、13 nM と約 10 倍程度の TSPO 結合活性の低下が見られた。よってアルキル-フェニルタイプについて置換基の導入検討を行うこととした。電子供与性基としてメトキシ基、電子吸引性基としてクロル基を導入したところ、TSPO 結合活性の向上が見られた。メトキシ基についてはメタ位 **(48f)**、クロル基についてはパラ位 **(48i)** への置換基導入にてより活性の向上が顕著であった。代謝安定性向上に効果があると考えられた電子吸引性基の導入 **(48h**、**48i)** や、ベンゼン環上パラ位への置換基導入 **(48g**、**48i)** ともに代謝安定性改善には至らず、次に脂溶性の軽減、水溶性向上を目指しアミド部のフェニル基からピリジル基への変換体について評価した。ピリジン誘導体では塩基性窒素の位

置が TSPO 活性に影響を与えることが明らかとなった。すなわち 4-ピリジル体 **48l** では活性が 100 倍以上低下した (Ki = 270 nM) 一方で、化合物 **48j**、**48k** では 15 倍程度の活性減弱 (Ki = 23 nM、28 nM) となった。アミノ基の導入においてもその置換位置により活性変化に差が見られ、オルト、メタ、パラ位の順に活性が減弱した。しかしいずれのアミド部の変換においても代謝安定性改善には至らなかった。アミド部分の置換基検討にて TSPO 結合活性が向上した誘導体は得られたものの、課題であった代謝安定性については TSPO 活性が大きく減弱した 2 級アミド体 **48c** を除いて改善された化合物は得られなかった。

Table 3-4. In vitro profiles of benzoxazolone derivatives with various amide moieties
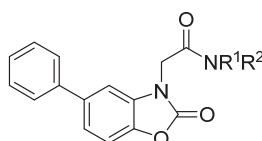
Compd	R^1	R^2	TSPO Ki[a] (nM)	CBR inhibition[b] (%)	Metabolic stability[c] (remaining%)
48a	Me	Ph	1.6	5	1
48d	*n*-Pr	*n*-Pr	17	2	0[d]
48e	Me	Bn	13	36	1
48f	Me	*m*-MeO-Ph	0.90	0	0
48g	Me	*p*-MeO-Ph	2.2	0	5[d]
48h	Me	*m*-Cl-Ph	0.79	0	0
48i	Me	*p*-Cl-Ph	0.21	0	0
48j	Me	2-Py	23	9	0
48k	Me	3-Py	28	29	0
48l	Me	4-Py	270	2	N.T.[e]
48m	Me	*o*-(Me$_2$NCH$_2$)-Ph	2.0	0	0
48n	Me	*m*-(Me$_2$NCH$_2$)-Ph	12	N.T.[e]	N.T.[e]
48o	Me	*p*-(Me$_2$NCH$_2$)-Ph	88	N.T.[e]	N.T.[e]
48c	H	Ph	220	N.T.[e]	36

[a] Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. [b] Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound. [c] Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 ^{o}C. The initial concentration of each compound was 1.0 μM. [d] Examinations were conducted at 10 μM of the compound. [e] Not tested

続いて、アリール置換基部分について検討を行った (Table 3-5)。中間体であるブロモ体 **81** やシアノ基が置換した **102** では、予想に反し高い TSPO への結合活性を示した。また、ベンゼン環へのメトキシ基、トリフルオロメチル基、トリフルオロメトキシ基の導入により高活性の化合物 **82-84**、**85a**、**85b** を得た。電子供与性基であるメトキシ基、電子吸引性基であるトリフルロメチル基の導入はともに TSPO 活性向上に寄与しており、ベンゼン環上の電子密度は TSPO 活性に大きな影響を与えていないと推測された。ベンゼン環へ水溶性置換基としてアミノ基を導入し

た **92-94** については、その置換位置により TSPO 活性に大きな差がみられ、メタ位、パラ位に関しては TSPO 結合活性は維持されていた (**93**, Ki = 5.8 nM; **94**, Ki = 11 nM) 。ベンゼン環をピリジン環に変換した誘導体では 3-ピリジン体 **88a** (Ki = 11 nM)、4-ピリジン体 **89a** (Ki = 5.3 nM) で活性の減弱がみられたが、代謝安定性において若干の改善がみられた (**88a**, 24%; **89a**, 27%) 。ベンゼン環からピリジン環への変換により代謝安定性が改善された化合物が得られたことから、ピリジン環上に置換基を持つ誘導体 **95-97** について誘導体を合成したが、代謝安定性が向上した誘導体は得られなかった。リンカーを挟んでベンゼン環が置換した **98**、**99** においても活性は維持されたものの代謝安定性の改善はみられなかった。

Table 3-5. In vitro profiles of benzoxazolone derivatives with various substituents at the 5-position
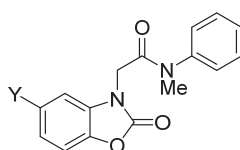
Compd	Y	TSPO Ki^a (nM)	CBR Inhibition^c (%)	Metabolic stability^d (remaining%)
100	H	11	3	49
81	Br	0.18	9	23
102	CN	0.91	0	84
48a	Ph	1.6	5	1
82	m-MeO-Ph	0.33	3	9^e
83	p-MeO-Ph	0.29	0	1
84	m-CF$_3$-Ph	0.48	0	6^e
85a	p-CF$_3$-Ph	0.68	0	26
86a	p-CF$_3$O-Ph	0.65	0	67
92	o-(Me$_2$NCH$_2$)-Ph	62	N.T.^{f}	N.T.^{f}
93	m-(Me$_2$NCH$_2$)-Ph	5.8	3	5
94	p-(Me$_2$NCH$_2$)-Ph	11	29	N.T.^{f}
87	2-Py	1.8	10	0
88a	3-Py	11	14	24
89a	4-Py	5.3	8	27
95	3-NH$_2$-2-Py	87%^{b}	8	0
96	3-MeO-2-Py	97%^{b}	10	0
97	5-Ac-3-Py	71%^{b}	2	0
98	PhO	3.8	2	1
99	PhNMe	0.36	10	1

^{a} Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. ^{b} Percent inhibition of [^{3}H]-PK11195 specific binding at 100 nM of the compound. ^{c} Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound. ^{d} Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 ^{o}C. The initial concentration of each compound was 1.0 μM. ^{e} Metabolic stability determined at 10 μM of the compound. ^{f} Not tested

Table 3-6. In vitro profiles of benzoxazolone derivatives

Compd	Y	R^1	R^2	TSPO Ki^a (nM)	CBR Inh.^{b} (%)	MS^c (remaining%)	Sol.^{d} (μg/mL) pH7.4	pH2.5
48a	Ph	Me	Ph	1.6	5	1	<1.0	1.0
48j	Ph	Me	2-Py	23	9	0	1.0	3.0
48k	Ph	Me	3-Py	28	29	0	45	180
48l	Ph	Me	4-Py	270	2	N.T.^{e}	N.T.^{e}	N.T.^{e}
85a	*p*-CF$_3$-Ph	Me	Ph	0.68	0	26	<1.0	<1.0
85b	*p*-CF$_3$-Ph	Me	3-Py	9.8	6	24	<1.0	<1.0
86a	*p*-CF$_3$O-Ph	Me	Ph	0.65	0	67	<1.0	<1.0
86b	*p*-CF$_3$O-Ph	Me	3-Py	4.9	0	32	<1.0	4.0
88a	3-Py	Me	Ph	11	14	24	5.0	440
88b	3-Py	Et	Ph	4.4	21	0	N.T.^{e}	N.T.^{e}
88c	3-Py	Me	*p*-MeO-Ph	19	0	9	N.T.^{e}	N.T.^{e}
88d	3-Py	Me	*p*-CF$_3$-Ph	8.6	0	38	1.0	420
88e	3-Py	Me	*p*-CF$_3$O-Ph	26	0	N.T.^{e}	N.T.^{e}	N.T.^{e}
89a	4-Py	Me	Ph	5.3	8	27	1.0	730
89b	4-Py	Et	Ph	3.0	5	24	2.0	860
89c	4-Py	Me	*p*-MeO-Ph	17	2	47	<1.0	130
89d	4-Py	Me	*p*-CF$_3$-Ph	6.6	0	35	3.0	>1000
89e	4-Py	Me	*p*-CF$_3$O-Ph	23	N.T.^{e}	N.T.^{e}	N.T.^{e}	N.T.^{e}

^{a} Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. ^{b} Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound. ^{c} Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 ^{o}C. The initial concentration of each compound was 1.0 μM. ^{d} The solubility was determined by HPLC using the supernatant obtained after shaking of the buffered solution (pH 7.4 and 2.5) (0.4 mL) containing 1 mg of tested compound, followed by centrifugation. ^{e} Not tested

　アミド置換基、アリール置換基について最適な組み合わせを持つ誘導体を見出すべく、アリール置換基としては代謝安定性と TSPO 活性ともに良好であった 4 種の置換基を選別し、アリール基またはアミド基のいずれかに水溶性置換基を含むよう誘導体合成を実施した (Table 3-6)。シアノ基が置換した誘導体 **102** は、TSPO 結合活性も強く、代謝安定性も良好であったが、課題である水溶性向上のため、アミド部分に水溶性置換基を導入したところ活性が大きく減弱したことからそれ以上の検討は中断した。化合物 **85a**、**86a** は、低水溶性が課題であったことからアミド部分として、水溶性向上が期待できる 3-ピリジル基を導入した。一方で、アリール置換基としてピリジル基をもつ **88a**、**89a** は TSPO 活性の向上が必要であったため、アミド置換基のベンゼン環上に置換基を導入した誘導体を中心に合成した。

化合物 **85a**、**86a** のアミド部分に 3-ピリジル基を導入した **85b**、**86b** はいずれも TSPO 活性は減弱したが、**86b** については、代謝安定性、水溶性に関しても経口吸収性が期待できる結果が得られたため有望化合物として評価を進めることとした。

　3-ピリジル体 **88a** のアミド部分変換誘導体 **88b-e** については、**88d** を除いて TSPO 活性または代謝安定性が低下することが明らかとなった。化合物 **88d** については、**86b** に比べ活性面で劣ったものの、薬物動態試験や抗不安モデル等の薬効評価を進めることとした。

　4-ピリジル体 **89a** のアミド部分変換体については、**89b**、**89d** にて、TSPO 活性と代謝安定性について十分なプロファイルを示した。しかしながら、4-ピリジル誘導体である **89a-b**、**89d** の 3 化合物はいずれも一部の代謝酵素 (CYP2C19、CYP3A4) に対し強い酵素阻害能を示し、薬物相互作用の懸念があったことから、これ以上の評価を進めることは断念した。

第 4 節　薬物動態プロファイル改善に向けた取組みおよび考察

　三環性ベンズオキサゾロン誘導体での検討において母骨格ベンジル位の代謝を抑制する目的で炭素鎖から酸素原子へ変換を実施したところ代謝安定性は低下したことから、母骨格電子密度の代謝安定性への影響が推定された (Figure 3-4)[38] 。そこで、ベンズオキサゾロン誘導体の検討を行うにあたり、置換基の電気陰性度を一つの指標として導入置換基の選別を行った。

21a
TSPO Ki = 0.94 nM
MS 12% remaining

43
TSPO Ki = 0.21 nM
MS 1% remaining
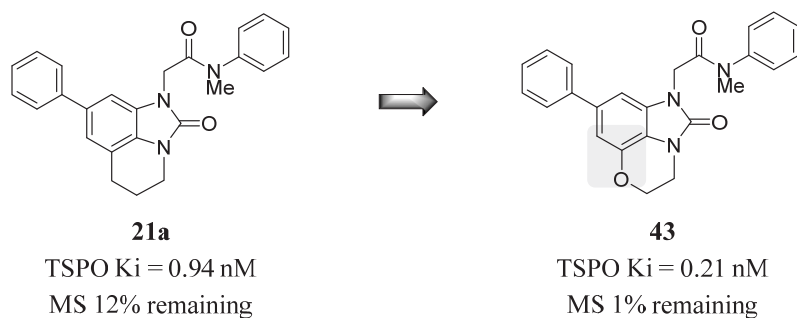
Figure 3-4. Results in tricyclic benzimidazolone derivative

　ベンズオキサゾロン誘導体の 5 位に導入した置換基の電気陰性度と代謝安定性との相関について考察を行った。ここで、Hammett の電子陰性度定数 (σ)[48] を用いて代謝安定性との相関性について考察を試みたところ、σ 値が 0.2 以上の化合物について比較的高い残存率を示すことが明らかとなった。一方で、脂溶性を反映する AlogD7.4 の値との相関についても検証を行ったが、相関性は得られなかった。Figure 3-5 で用いたデータについて Table 3-7 に記載した。

(a)　　　　　　　　　　　　　　　　　　　　(b)
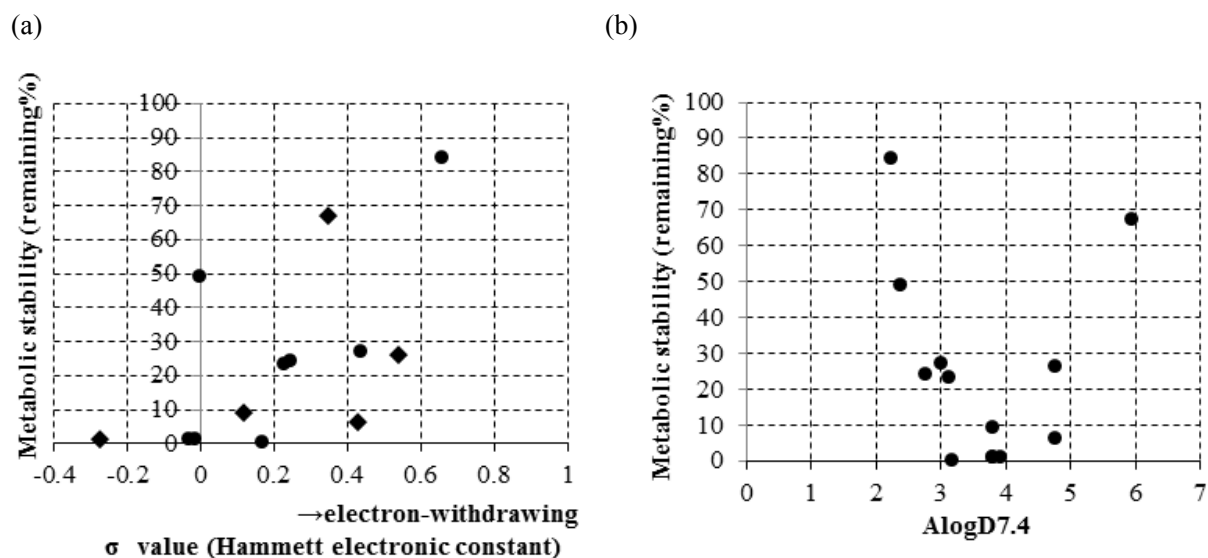
Figure 3-5. (a) Metabolic stability vs σ value (b) Metabolic stability vs AlogD7.4

Table 3-7. TSPO binding affinity, metabolic stability and σ value of benzoxazolone derivatives
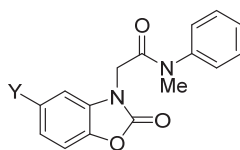
Compd	Y	TSPO Ki[a] (nM)	Metabolic stability[b] (remaining%)	AlogD7.4[d]	σ
100	H	11	49	2.38	0
81	Br	0.18	23	3.14	0.23[e]
102	CN	0.91	84	2.26	0.66[e]
48a	Ph	1.6	1	3.81	-0.01[e]
87	2-Py	1.8	0	3.19	0.17[e]
88a	3-Py	11	24	2.77	0.25[e]
89a	4-Py	5.3	27	3.02	0.44[e]
98	PhO	3.8	1	3.93	-0.03[e]
82	*m*-MeO-Ph	0.33	9[c]	3.82	0.12
83	*p*-MeO-Ph	0.29	1	3.82	-0.27
84	*m*-CF$_3$-Ph	0.48	6[c]	4.78	0.43
85a	*p*-CF$_3$-Ph	0.68	26	4.78	0.54
86a	*p*-CF$_3$O-Ph	0.65	67	5.95	0.35

[a] Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. [b] Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 °C. The initial concentration of each compound was 1.0 μM. [c] Examinations were conducted at 10 μM of the compound. [d] Predicted with PipelinePilot, version 8.0.1. [e] Hammett sigma constant at the *para*-position was used.

第 5 節　TSPO リガンドの作用機序についての考察

　神経ステロイド生合成において、cholesterol のミトコンドリア内への輸送が律速段階の一つとされており [17]、グリア細胞内のミトコンドリア膜上に存在する TSPO がコレステロールの細胞質からミトコンドリア内への輸送を調節していることが明らかとなっている [12, 16]。種々の TSPO リガンドについて、in vitro 試験にて pregnenolone 生成量が増加することが報告されている [31d]。一方で、神経ステロイドは $GABA_A$ 受容体の活性化により、抗不安作用を示すことが報告されていることから [32b]、TSPO リガンドは神経ステロイドの生合成促進により抗不安作用を示すと考えられる。

　既存の TSPO リガンドについても、実験動物にて抗不安作用を示すことが報告されている。FGIN-1-27 は、ラットでの高架式十字迷路試験において抗不安作用を示し、TSPO のアンタゴニストである PK11195 によってその効果がキャンセルされ、 CBR アンタゴニストである Flumazenil の投与では影響を受けないことからその作用は TSPO を介したものと考察されている [31d]。

　ここで、神経ステロイドの生合成経路の一部について Figure 3-6 に示した [21a]。また、trilostane は pregnenolone から progesterone への変換を触媒する 3β-HSD を阻害することが知られている [49]。よって、化合物 **88a** の薬効が、TSPO の刺激による神経ステロイドの生合成促進の結果として発現しているものであれば、trilostane の前投与により薬効がキャンセルされると考えた。
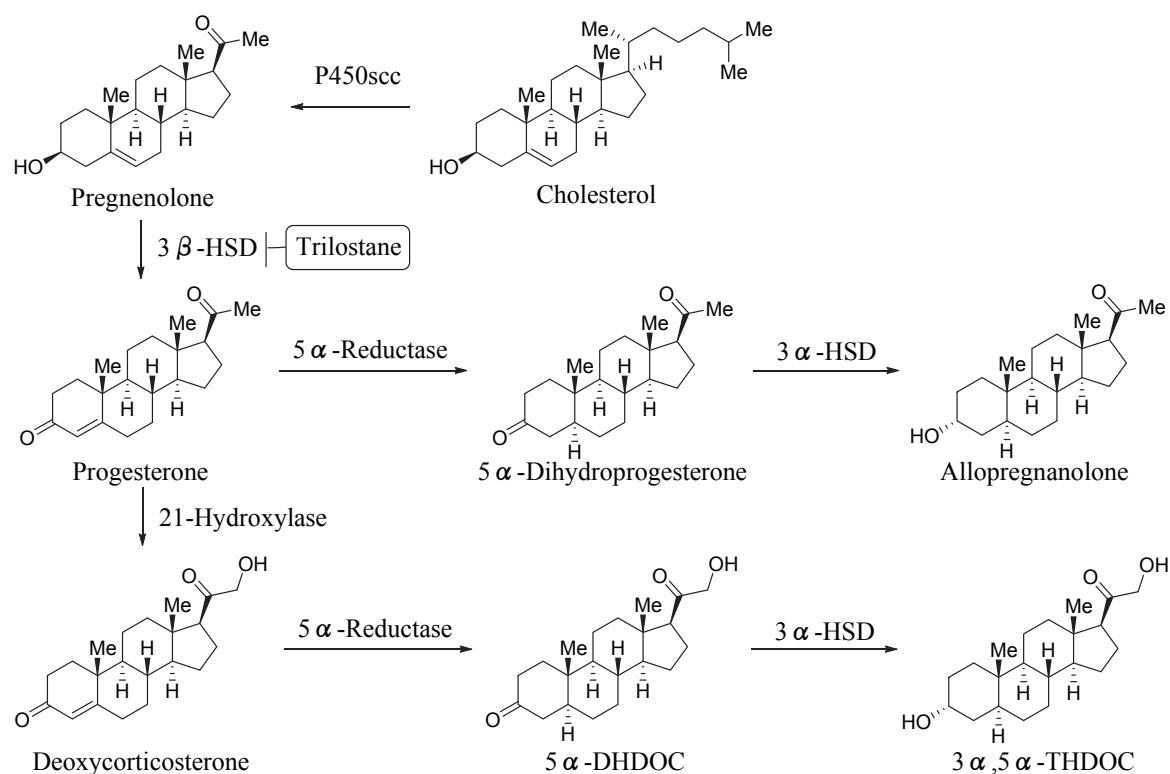
Figure 3-6. Biosynthesis of neurosteroids

化合物 **88a** についてラットを用いた薬物動態試験を行った。これらの結果より、クリアランスが高いなど若干の課題はあるものの薬効について確認するうえで、十分な脳内濃度が確保されていることが確認された (Table 3-8) 。

Table 3-8. Pharmacokinetic properties of **88a**[a]

Dose (mg/kg)	AUC (μg·h/mL)	CL (mL/min/kg)	T1/2 (min)	Vdss (L/kg)	Cmax (ng/mL)	Tmax (min)	F (%)	B/P[b]	Protein Binding Serum/Brain(%)
1 (iv)	6.04	166	17.3	4.4					80.8 / 91.1
10 (po)	8.37				50.4	60	13.9	0.63	

[a]Each value represents the mean of two or three rats
[b]B/P means brain/plasma AUC ratio after oral administration (10 mg/kg) of the HCl salt of **87a**

化合物 **88a** について Vogel 型葛藤試験 [50] を実施したところ抗不安活性が確認された (Figure 3-8) 。ここで、TSPO による神経ステロイド産生促進により期待される薬理作用として、神経ステロイドが NMDA 受容体に作用することが知られていることから [21]、NMDA 拮抗薬である MK-801 投与による認知障害が改善される可能性があると考えラット受動的回避反応試験[51]を実施したところ、Vogel 型葛藤試験と同様に薬効が検出された (Figure 3-9) 。

ここで、前述のとおり trilostane は Pregnenolone から Progesterone への変換を触媒する 3β-HSD を阻害することが知られている。よって、化合物 **88a** の薬理作用への神経ステロイドの関与について、trilostane により検証することとした。

化合物 **88a** については、ラット Vogel 型葛藤試験ならびにラット受動的回避反応試験といった 2 つの異なる動物モデルでその薬理作用が確認されているが、今回の拮抗試験についてはラット受動的回避反応試験を用いて実施した。その結果、trilostane の前投与により **88a** の薬効が消失されたことから、化合物 **88a** の薬理作用発現への神経ステロイドの関与が示唆された (Figure 3-9)。
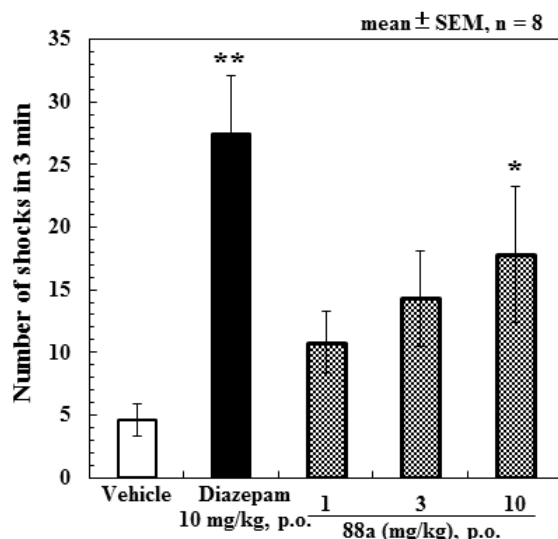
Figure 3-8. Effects of **88a** on the rat Vogel-type conflict model. ** $P < 0.01$, significantly different from the vehicle control group (Student's t-test). * $P < 0.05$, significantly different from the vehicle control group (Dunnett's multiple comparison test).

(a)   (b)
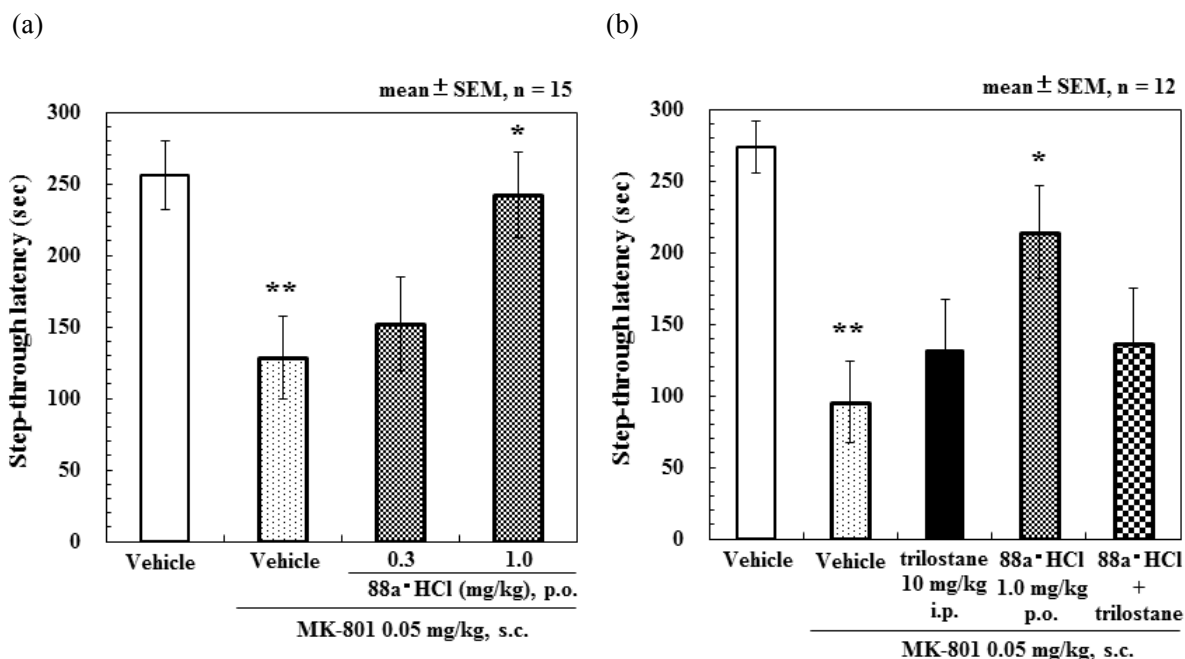
Figure 3-9. Effects of **88a·HCl** on the rat passive avoidance response. ** $P < 0.01$, significantly different from the vehicle control group (Wilcoxon test). * $P < 0.05$, significantly different from the vehicle control group (Steel's test).

　本章におけるベンズオキサゾロン誘導体の探索合成により良好な TSPO 結合活性、代謝安定性を示す有望化合物 **86**、**88d** を見出すことができたことから、次章にてこれらの化合物の動物モデルでの in vivo 評価結果等の薬理プロファイルについて考察する。

37

第 4 章　ベンズオキサゾロン誘導体の薬理プロファイル

第 1 節　*N*-Methyl-2-{2-oxo-5-[4-(trifluoromethoxy)phenyl]-1,3-benzoxazol-3(2*H*)-yl}-*N*-(pyridin-3-yl)acetamide の薬物動態試験、薬理作用評価および副作用評価

　化合物 **86b·HCl** について、ラット、マウスを用いた薬物動態試験を行った (Table 4-1) 。これらの結果より、十分な脳内濃度が確保されており、ラット、マウスともに同程度の血中濃度に達していることが確認された。中性領域でも胆汁酸を含む調製溶液中ではある程度の溶解度 (23 μg/mL in a pH 6.8 buffer with bile acids) を示したものの、高容量の投与にて低水溶性の影響によるものと思われる吸収飽和がみられた (Figure 4-1)。

　抗不安作用を確認すべく Vogel 型葛藤試験を実施したところ薬効が確認された (Figure 4-1)。

Table 4-1. Pharmacokinetic properties of **86b·HCl**[a]

Dose (mg/kg)	AUC (μg·h/mL)	CL (mL/min/kg)	T1/2 (min)	Vdss (L/kg)	Cmax (ng/mL)	Tmax (min)	F (%)	B/P[b]	Protein Binding Serum/Brain(%)
1 (iv)	38.1	26.2	116	3.4					96.1 / 99.7
10 (po)	172.6				329	30	45	1.3	
100(po)	934.0				571	60			
300(po)	730.1				530	240			

[a]Each value represents the mean of two or three rats
[b]B/P means brain/plasma AUC ratio after oral administration (10 mg/kg) of the HCl salt of **86b**

(a)

(b)
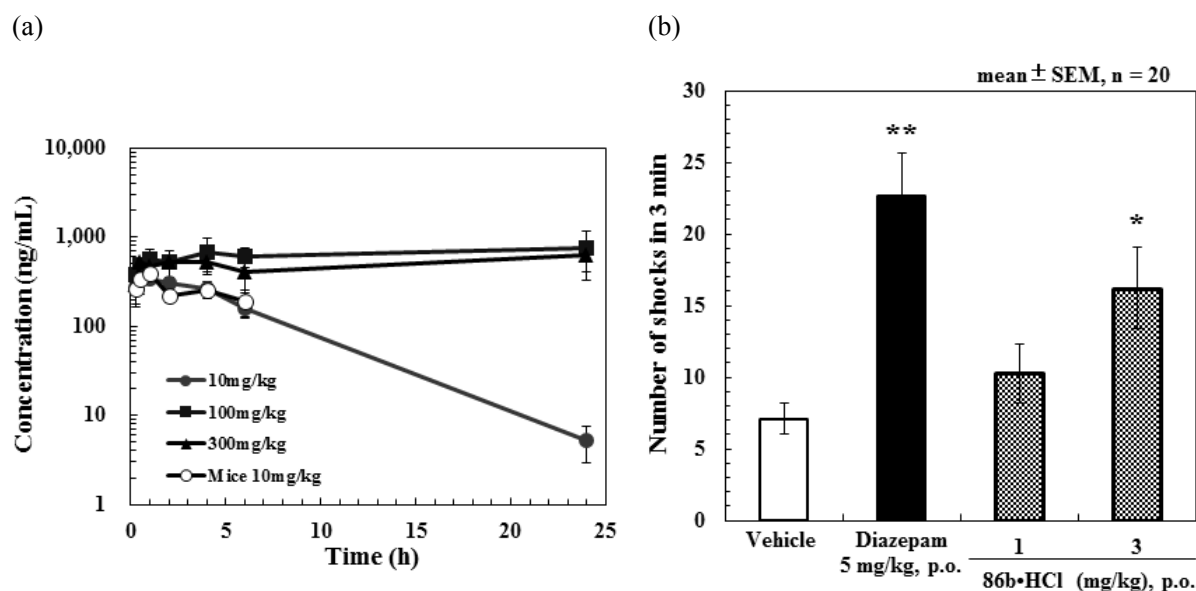
Figure 4-1. (a) Pharmacokinetic properties of **86b·HCl** (b) Effects of **86b·HCl** on the rat Vogel-type conflict model. ** $P < 0.01$, significantly different from the vehicle control group (Student's *t*-test). * $P < 0.05$, significantly different from the vehicle control group (Dunnett's multiple comparison test).

ここで、ベンゾジアゼピン系薬剤でみられる中枢性副作用について確認を行った。化合物 **86b** は、CBR との選択性が十分であることから (IC50 > 10 μM)、協調運動に対しては影響を与えないと考えた。しかし **86b·HCl** を 300 mg/kg マウスに経口投与しロータロッド試験を実施したところ協調運動の低下が観察された (Figure 4-2)。Figure 4-1 (a) に示すとおり **86b·HCl** の用量とラット血中濃度の相関関係の検討にて、10 mg/kg から 300 mg/kg へ 30 倍量投与量を増やした場合にも、血中濃度の増加は 2 倍にも達せず吸収段階にて飽和状態にあることが示唆された。また 10mg /kg 投与においてラットとマウスにてほぼ同等の血中濃度推移を示すことを確認している。これらの結果より投与量としては抗不安活性発現とベンゾジアゼピン系副作用の一つである協調運動障害の発現に 100 倍の差が得られているが、実際の血中濃度に換算するとその差は十分でないことが明らかとなった。
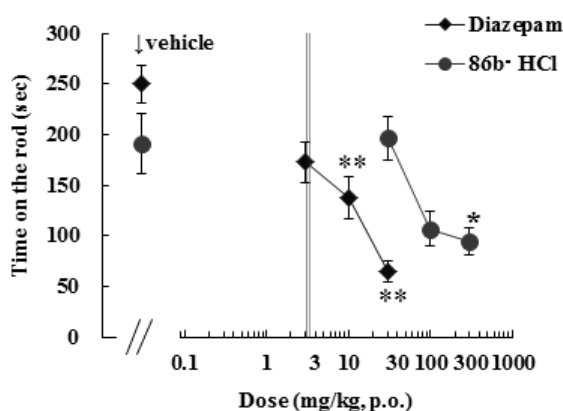
Figure 4-2. Effects of **86b·HCl** in the mice rota-rod test. * *P* < 0.05, ** *P* < 0.01, significantly different from the vehicle control group (Steel's test).

　化合物 **86b** の協調運動障害の原因を探るべく各種受容体、チャネル、トランスポーター、酵素に関する結合試験を実施した。

　Table 4-2 に示すとおり化合物 **86b** は 10 μM にて、ラットの中枢性ナトリウムチャネルへの作用に関してのみ 50% を超える作用を示した (IC$_{50}$ = 0.70 μM) 。本評価ではラットの脳由来のナトリウムチャネルに結合するバトラコトキシン結合部位についての結合親和性を算出している。化合物 **86b** は、CBR との選択性が十分であること(IC50 > 10 μM)、中枢領域に存在するナトリウムチャネルの阻害により協調運動障害が報告されていることから [52]、**86b** の協調運動障害の原因は中枢性ナトリウムチャネルへの作用によるものであることが示唆された。

Table 4-2. Radioligand-bindin assays of **86b**[a]

Assay Name	Species	Inhibition (%)
Monoamine Oxidase MAO-A	human	8
Phosphodiesterase PDE4	human	48
Adenosine A_1	human	-2
Adenosine A_{2A}	human	0
Adrenergic α_{2A}	human	11
Adrenergic β_1	human	2
Adrenergic β_2	human	1
Angiotensin AT_1	human	-2
Angiotensin AT_2	human	11
Bombesin BB3	human	13
Calcium Channel L-Type, Dihydropyridine	rat	15
Calcium Channel N-Type	rat	0
Cannabinoid CB_1	human	13
Cholecystokinin CCK_1 (CCK_A)	human	-5
Cholecystokinin CCK_2 (CCK_B)	human	7
Corticotropin Releasing Factor CRF_1	human	26
Dopamine D_1	human	19
Estrogen ERβ	human	18
$GABA_A$, Agonist Site	rat	-10
$GABA_A$, Chloride Channel, TBOB	rat	10
$GABA_B$, Non-Selective	rat	-1
Galanin GAL1	human	-7
Glucocorticoid	human	0
Glutamate, Kainate	rat	1
Glutamate, NMDA, Phencyclidine	rat	-8
Glycine, Strychnine-Sensitive	rat	-8
Histamine H_3	human	-8
Melanocortin MC_4	human	3
Muscarinic M_2	human	16
Muscarinic M_3	human	4
Neuropeptide Y Y_1	human	36
Neuropeptide Y Y_2	human	9

Table 4-2. (Continued)

Assay Name	Species	Inhibition (%)
Nicotinic Acetylcholine	human	-15
Opiate μ (OP3, MOP)	human	24
Orphanin ORL$_1$	human	-4
Potassium Channel [K$_{ATP}$]	syrian hamster	-16
Serotonin 5-HT$_2$, Non-Selective	rat	12
Serotonin 5-HT$_3$	human	5
Serotonin 5-HT$_4$	guinea pig	12
Serotonin 5-HT$_6$	human	1
Sigma σ$_1$	human	6
Sodium Channel, Site 2	rat	108
Somatostatin sst2	human	-2
Tachykinin NK$_1$	human	-1
Transporter, Dopamine (DAT)	human	2
Transporter, Norepinephrine (NET)	human	-10
Vasopressin V$_{1A}$	human	4
Vasopressin V$_{1B}$	human	-1

[a]Compounds **86b** (10 μM, n = 2) was evaluated at Eurofins Panlabs Taiwan, Ltd. in the following 48 receptors, ion channels, and enzymes. Primary binding assays were provided by the commercial supplier Eurofins Panlabs Taiwan, Ltd. Further information about the assay is given on Eurofins Panlabs Taiwan's website (https://www.eurofinspanlabs.com/Panlabs).

　ベンズオキサゾロン誘導体について代表化合物の中枢性ナトリウムチャネルに対する結合活性を実施したので次節にて述べる。

第 2 節　中枢性ナトリウムチャネルについての評価および考察

　ベンズオキサゾロン誘導体について代表化合物を選抜し中枢性ナトリウムチャネルに対する結合活性を評価した。化合物 **86b** の 5 位アリール置換基とアミド置換基とでどちらがナトリウムチャネルへの作用により影響を与えているか検証するため **48k** と **86a** について評価を行った。その結果 5 位ベンゼン環上のパラトリフルオロメトキシ基が中枢性ナトリウムチャネルへの作用を強めていることが示唆された。3-ピリジル基を有する **88a**、**88d** はナトリウムチャネルへの作用は弱いことが分かった。そこで TSPO 結合活性と代謝安定性を考慮し **88d** について更なる評価を行った。

Table 4-3. In vitro profiles of benzoxazolone derivatives

Compd	Y	R	TSPO Ki^a (nM)	MS^b (remaining%)	Sol.^{c} (μg/mL) pH7.4	pH2.5	Na$^+$ channel (site2) IC50 (μM)^d
48a	Ph	Ph	1.6	1	<1.0	1.0	>1.0
48k	Ph	3-Py	28	0	45	180	>1.0
86a	p-CF$_3$O-Ph	Ph	0.65	67	<1.0	<1.0	0.19
86b	p-CF$_3$O-Ph	3-Py	4.9	32	<1.0	4.0	0.70
88a	3-Py	Ph	11	24	5.0	440	>1.0
88d	3-Py	p-CF$_3$-Ph	8.6	38	1.0	420	>10

^{a} Ki values represent the means of 1-3 separate experiments run in duplicate using 4 concentrations of each compound. ^{b} Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 ^{o}C. The initial concentration of each compound was 1.0 μM. ^{c} The solubility was determined by HPLC using the supernatant obtained after shaking of the buffered solution (pH 7.4 and 2.5) (0.4 mL) containing 1 mg of tested compound, followed by centrifugation. ^{d} IC$_{50}$ value represents displacement of [^3H]Batrachotoxin (5.0 nM) binding to rat brain by each compound.

第 3 節　*N*-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-[4-(trifluoromethyl)-phenyl]acetamide の薬物動態試験、薬理作用評価および副作用評価

　N-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-[4-(trifluoromethyl)-phenyl]acetamide (**88d**) について各種受容体、チャネル、トランスポーター、酵素に関する結合試験を実施した。その結果、化合物 **88d** は、Table 4-4 に示すとおり中枢性ナトリウムチャネル以外に対しても十分な選択性を示した。

Table 4-4. Radioligand-bindin assays of **88d**[a]

Assay Name	Species	Inhibition (%)
Monoamine Oxidase MAO-A	human	27
Phosphodiesterase PDE4	human	19
Adenosine A_1	human	3
Adenosine A_{2A}	human	19
Adrenergic α_{2A}	human	10
Adrenergic β_1	human	-3
Adrenergic β_2	human	-2
Angiotensin AT_1	human	-8
Angiotensin AT_2	human	2
Bombesin BB3	human	4
Calcium Channel L-Type, Dihydropyridine	rat	1
Calcium Channel N-Type	rat	-2
Cannabinoid CB_1	human	17
Cholecystokinin CCK_1 (CCK_A)	human	-6
Cholecystokinin CCK_2 (CCK_B)	human	10
Corticotropin Releasing Factor CRF_1	human	14
Dopamine D_1	human	3
Estrogen ERβ	human	-4
$GABA_A$, Agonist Site	rat	10
$GABA_A$, Chloride Channel, TBOB	rat	0
$GABA_B$, Non-Selective	rat	16
Galanin GAL1	human	3
Glucocorticoid	human	2
Glutamate, Kainate	rat	-3
Glutamate, NMDA, Phencyclidine	rat	-10

Table 4-4. (Continued)

Assay Name	Species	Inhibition (%)
Glycine, Strychnine-Sensitive	rat	9
Histamine H_3	human	5
Melanocortin MC_4	human	-1
Muscarinic M_2	human	-9
Muscarinic M_3	human	1
Neuropeptide Y Y_1	human	-5
Neuropeptide Y Y_2	human	0
Nicotinic Acetylcholine	human	-2
Opiate μ (OP3, MOP)	human	3
Orphanin ORL_1	human	-23
Potassium Channel [K_{ATP}]	syrian hamster	-1
Serotonin 5-HT_2, Non-Selective	rat	5
Serotonin 5-HT_3	human	3
Serotonin 5-HT_4	guinea pig	24
Serotonin 5-HT_6	human	5
Sigma $σ_1$	human	-3
Sodium Channel, Site 2	rat	-8
Somatostatin sst2	human	-2
Tachykinin NK_1	human	-10
Transporter, Dopamine (DAT)	human	-4
Transporter, Norepinephrine (NET)	human	12
Vasopressin V_{1A}	human	-1
Vasopressin V_{1B}	human	0

[a]Compounds **88d** (10 μM, n = 2) was evaluated at Eurofins Panlabs Taiwan, Ltd. in the following 48 receptors, ion channels, and enzymes. Primary binding assays were provided by the commercial supplier Eurofins Panlabs Taiwan, Ltd. Further information about the assay is given on Eurofins Panlabs Taiwan's website (https://www.eurofinspanlabs.com/Panlabs).

　化合物 **88d** の中性領域での水溶性は **86b** と同様に低かったが、酸性領域では溶解度が向上した。中性領域でも胆汁酸を含む調製溶液中ではある程度の溶解度 (17 μg/mL in a pH 6.8 buffer with bile acids) を示した。ラット薬物動態試験にて BA は 20% で、脳内濃度も薬効発現が期待される濃度に達しており (Table 4-5)、500 mg/kg までは用量に相関した血中濃度の増加が確認された。また 10mg /kg 投与においてラットとマウスにてほぼ同等の血中濃度推移を示すことも確認している。化合物 **88d** は 1 mg/kg の経口投与にて抗不安効果を示した (Figure 4-2)。

44

Table 4-5. Pharmacokinetic properties of **88d·HCl**[a]

Dose (mg/kg)	AUC (μg·h/mL)	CL (mL/min/kg)	T1/2 (min)	Vdss (L/kg)	Cmax (ng/mL)	Tmax (min)	F (%)	B/P[b]	Protein Binding Serum/Brain(%)
1 (iv)	33.0	30.3	71.4	1.6					90.5 / 98.2
10 (po)	66.8				473	15	20	1.2	
100(po)	239.5				1313	120			
500(po)	1223.1				4771	240			

[a]Each value represents the mean of two or three rats
[b]B/P means brain/plasma AUC ratio after oral administration (10 mg/kg) of the HCl salt of **88d**

(a)   (b)
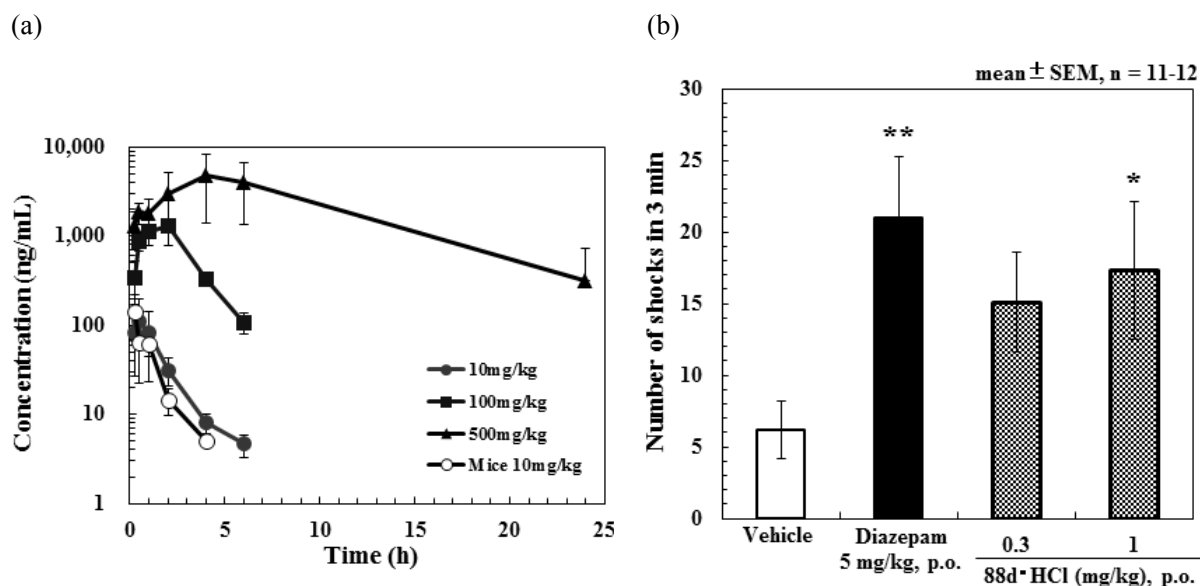
Figure 4-2. (a) Pharmacokinetic properties of **88d·HCl** (b) Effects of **88d·HCl** on the rat Vogel-type conflict model. ** $P < 0.01$, * $P < 0.05$, significantly different from the vehicle control group (Student's *t*-test).

　次に **88d** のベンゾジアゼピン系副作用について検証を行った (Figure 4-6) 。協調運動障害、健忘、鎮静についてジアゼパムでは薬効用量の数倍量での投与より各副作用が発現したが、**88d·HCl** は最高投与量においても作用はみられなかった。ヘキソバルビタール誘発の催眠作用に関してのみ 300 mg/kg にて有意な睡眠時間の延長がみられたものの、薬効発現用量と比較して十分な乖離は得られていると考えられる。

　化合物 **88d** については、種差についても検討しヒト TSPO に対しても Ki = 8.0 nM とラットとほぼ同等の活性を示している。代謝安定性については、ヒト肝 S-9 に対する安定性は、試験開始時の基質濃度 1.0 μM での評価において、30 分後の残存率は 84% とラットよりもより高い BA が期待される結果を得ている。またイヌによる大動物での安全性試験、ラットの 2 週間連投による安全性試験においても重篤な毒性等は見つかっておらず新規な精神疾患治療薬として期待される。

(a)

(b)

(c)

(d)
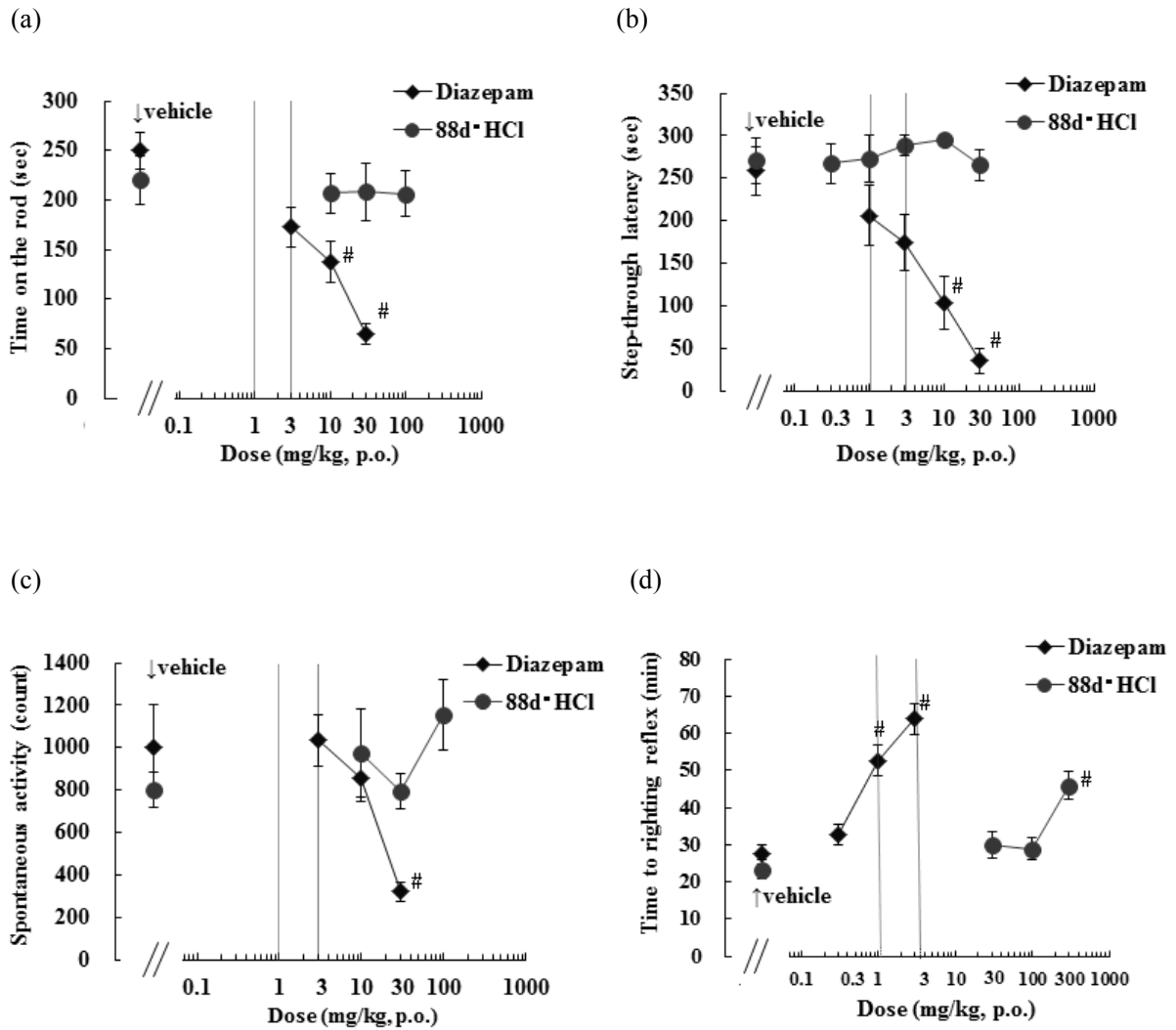
Figure 4-6. Effects of **88d·HCl** for safety study. (a) Mice rota-rod test. # $P < 0.01$, significantly different from the vehicle control group (Steel's test). (b) Rats passive avoidance response. # $P < 0.01$, significantly different from the vehicle control group (Steel's test). (c) Locomotor activity in mice. # $P < 0.01$, significantly different from the vehicle control group (Dunnett's multiple comparison test). (d) Hexobarbital-induced sleep in mice. # $P < 0.01$, significantly different from the vehicle control group (Steel's test).

　著者は、TSPO の中枢神経系でのステロイド生合成への関与に着目し、選択的 TSPO リガンドの創製を目的として本研究に着手し、その新規な不安障害、うつ病を主とする精神疾患への治療薬としての可能性について検討を行った。本研究により、TSPO に高選択的に作用し、動物モデルでの経口投与にて抗不安活性を示したベンズオキサゾロン骨格を有する *N*-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-[4-(trifluoromethyl)-phenyl]acetamide (**88d**) を見出した。本化合物は、ベンゾジアゼピン系薬剤で課題となっている中枢系副作用と十分な乖離が確認されており、既存薬の課題を改善した新規な不安障害をはじめとする精神疾患治療薬として期待される。

　第 2 章では、Ro5-4864 のジアゼピノン環開環によりデザインしたベンズイミダゾロン誘導体での知見をもとに TSPO 活性向上および構造面での高い新規性が見込まれた三環性ベンズイミダゾロン誘導体への展開を図り、誘導体合成を行った (Figure 4)。その結果、既存の TSPO リガンドを上回る TSPO 結合活性 (Ki < 1nM) を示す複数の化合物を見出した。しかしながら、リード化合物である **21a** を用いた薬物動態試験結果より、肝臓での代謝安定性が低く、経口吸収性に乏しいことが明らかとなった。アミド部位、アリール置換基の変換により代謝安定性の改善を試みたが、有望化合物の取得には至らなかった。本誘導体について、代謝安定性と各パラメーターとの相関について考察を行い、得られたデータを新規誘導体における探索研究において置換基選抜の際の指標として活用した。

　第 3 章では、三環性ベンズイミダゾロン誘導体で課題となった代謝安定性を中心に薬物動態パラメーターに着目し、ベンズイミダゾロン誘導体の N^3-アルキル構造を変換したベンズオキサゾロン誘導体について探索研究を実施した (Figure 5)。三環性ベンズイミダゾロン誘導体にて、母骨格の電子密度と代謝安定性の関係が示唆されたことから、Hammett の電気陰性度定数をひとつの指標として導入置換基の選別を行った。その結果、5 位アリール置換基上に電子吸引性官能基を導入した誘導体にて、強い TSPO 結合活性と代謝安定性が改善され良好な in vitro 薬物プロファイルを示す **86b** および **88d** を見出した。また、本章にて、ベンズオキサゾロン骨格を有する新規な TSPO リガンド **88a** を用いて、TSPO リガンドの作用機序に関して考察を行った。神経ステロイドの変換酵素 3β-HSD の阻害剤である trilostane の前投与により、**88a** の動物モデルでの薬理作用が拮抗されることが確認された。本結果は、TSPO リガンドによる薬理作用が神経ステロイドの産生促進を通じて発現していることを示唆するものである。

　第 4 章では、化合物 **86b** の薬理学プロファイルを明らかにした。化合物 **86b·HCl** は、抗不安作用が確認されたものの、CBR との選択性が十分であるにも関わらず、ベンゾジアゼピン系薬剤の副作用の一つである協調運動障害がみられた。さらに、PK 試験の結果も併せた解析により、その副作用を発現する血中濃度は、抗不安作用を示す濃度と十分乖離していないことが明らかとなった。本副作用発現の原因を探るべく、各種受容体、チャネル、トランスポーター、酵素に関する結合試験を実施した。その結果、中枢性ナトリウムチャネルへの結合 (IC50 = 0.70 μM) が確

認されたことからこの作用が協調運動障害発現の一因と推定した。そこで第 3 章で得られたベンズオキサゾロン誘導体について、中枢性ナトリウムチャネルについて評価を行ったところ、有望化合物の一つであった **88d** は、ナトリウムチャネルへの作用が非常に弱かったことから、副作用プロファイルを明らかにすべく検討を行った。その結果、化合物 **88d** は、低用量で抗不安活性が見られ、またベンゾジアゼピン系薬剤で課題となっている中枢性副作用との十分な乖離が確認された。化合物 **88d** は、TSPO 活性について、ヒトにおいてもラットと大きな種差がないことが in vitro 試験にて確認されており、新規な不安障害をはじめとする精神疾患治療薬として期待される。

　本研究にて取り組んだ代謝安定性改善に向けた検討については、TSPO リガンドの探索研究に限らず、一般的な創薬研究にて度々直面する課題の一つである。よって、本論文にて示した電気陰性度定数と代謝安定性との相関関係については幅広い創薬研究に活用できるものである。

　三環性ベンズイミダゾロン誘導体、ベンズオキサゾロン誘導体にて得られた構造活性相関情報は、今後の TSPO の作用部位の解明、ファーマコファーモデルの構築等 TSPO の研究に役立つと考えられる。

　さらに本研究にて見出された *N*-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-[4-(trifluoromethyl)-phenyl]acetamide (**88d**) は、TSPO への選択性が高いことから、既存薬でみられる副作用が回避された新規な精神疾患治療薬として期待されるとともに、本化合物の高い脳移行性等を含めた優れた薬物動態プロファイルにより、PET リガンドとしての診断や TSPO 機能解明に向けた取り組みへの活用も期待される [53]。

21a : Y = Ph, NR^{1}R^2 = NMePh

86b : Y = *p*-CF$_3$O-Ph, NR^{1}R^2 = NMe(3-Py)
88a : Y = 3-Py, NR^{1}R^2 = NMePh
88d : Y = 3-Py, NR^{1}R^2 = NMe(*p*-CF$_3$-Ph)
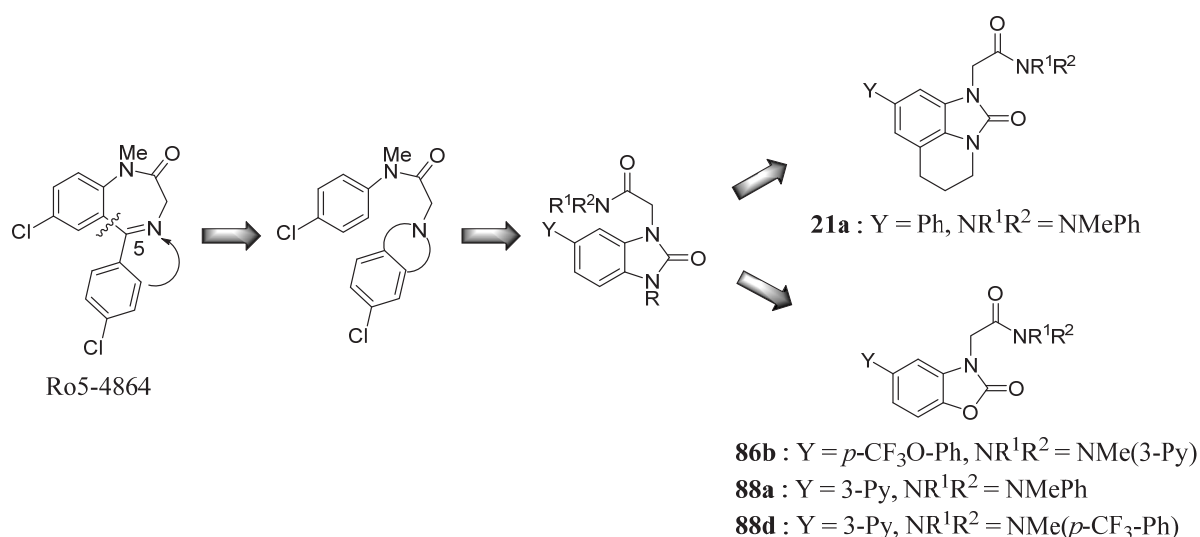
Figure 5. Development of tricyclic benzimidazolone and benzoxazolone derivatives

Chapter 6 Experimental Section

6-1 Chemistry

6-1-1 Genaral Information

Melting points were determined on Stanford Research Systems OptiMelt MPA100 without correction. NMR spectra were recorded at ambient temperature on a JEOL JNM-AL400 FT NMR spectrometer. Chemical shifts are expressed in δ values (ppm) relative to tetramethylsilane as an internal standard, and signals are expressed as s (singlet), d (doublet), t (triplet), m (multiplet), or br (broad). IR spectra were recorded on a JEOL JIR-SPX60 spectrometer as attenuated total reflection (ATR). High-resolution mass spectra (HRMS) were recorded on a Thermo Fisher Scientific LTQ orbitrap Discovery MS equipment. Elemental analysis was performed on a CE Instrument EA1110 and a Yokokawa analytical system IC7000. In general, reagents and solvents were used as obtained from commercial suppliers without further purification. Reaction progress was determined by thin layer chromatography (TLC) analysis on a Merck silica gel 60 F254 precoated glass plate. Visualization was done with UV light (254 nm) or iodine. Flash column chromatography was conducted using Merck silica gel 60 (70-230 mesh). All reactions were carried out under a nitrogen atmosphere unless otherwise mentioned.

6-1-2 Experiments in Chapter 2

Methyl 3,4-dihydroquinoline-1(2*H*)-carboxylate (7)

To a suspension of tetrahydroquinoline (18.0 mL, 143 mmol) and K_2CO_3 (79.3 g, 574 mmol) in DMF (100 mL) was added methylchloroformate (33.2 mL, 430 mmol) with cooling in an ice bath. The reaction mixture was stirred at 50 °C for 6 h and cooled to room temperature. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo to give **7** (27.2 g, 99%) as a yellow oil. This product was used in the following reaction without further purification.

^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.66 (1H, d, *J* = 7.6 Hz), 7.16 (1H, dd, *J* = 7.7. 7.7 Hz), 7.09 (1H, d, *J* = 7.3 Hz), 7.01 (1H, dd, *J* = 7.3, 7.3 Hz), 3.79 (3H, s), 3.76 (2H, t, *J* = 6.1 Hz), 2.77 (2H, t, *J* = 6.6 Hz), 1.99-1.90 (2H, m); ^{13}C-NMR (100 MHz, $CDCl_3$) δ: 155.4, 138.1, 130.0, 128.6, 125.9, 123.9, 123.7, 52.8, 44.8, 27.3, 23.4; IR (ATR) 1701, 1697, 1493, 1439, 1327 cm^{-1}; HRMS (ESI) m/z calcd for $C_{11}H_{14}NO_2$ [M+H][+] 192.1019; found 192.1015.

Methyl 2,3,4,5-tetrahydro-1*H*-1-benzazepine-1-carboxylate (8)

Compound **8** was prepared from 2,3,4,5-tetrahydro-1*H*-benzo[*b*]azepine (800 mg, 5.43 mmol) in a manner similar to that described for compound **7** as a white solid (706 mg, 63%).

Mp 113-115 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.25-7.10 (4H, m), 4.58-4.32 (1H, m), 3.79 and 3.65 (3H, each s), 2.87-2.58 (3H, m), 2.04-1.75 (3H, m), 1.55-1.27 (1H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 155.4, 142.1, 139.8, 129.8, 128.0, 127.1, 126.7, 52.8, 49.0, 34.6, 29.5, 26.3; IR (ATR) 1689, 1495, 1439, 1385, 1307 cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_{16}$NO$_2$ [M+H]$^+$ 206.1176; found 206.1173.

Methyl 6-bromo-3,4-dihydroquinoline-1(2*H*)-carboxylate (9)

To a solution of **7** (26.3 g, 138 mmol) in DMF (140 mL) was added *N*-bromosuccinimide (26.9 g, 151 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 2.5 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H$_2$O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo to give **9** (37.1 g, quant.) as a brown oil. This product was used in the following reaction without further purification.

^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.59 (1H, d, *J* = 8.5 Hz), 7.28-7.21 (2H, m), 3.79 (3H, s), 3.74 (2H, t, *J* = 6.1 Hz), 2.74 (2H, t, *J* = 6.6 Hz), 1.96-1.89 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 155.1, 137.2, 132.0, 131.2, 128.9, 125.4, 116.4, 53.0, 44.7, 27.2, 23.1; IR (ATR) 1701, 1483, 1441, 1321, 727 cm^{-1}; HRMS (ESI) m/z calcd for C$_{11}$H$_{13}$BrNO$_2$ [M+H]$^+$ 270.0124; found 270.0124.

Methyl 7-bromo-2,3,4,5-tetrahydro-1*H*-1-benzazepine-1-carboxylate (10)

Compound **10** was prepared from **8** (521 mg, 2.54 mmol) in a manner similar to that described for compound **9** as a colorless oil (590 mg, 82%).

^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.37-7.27 (2H, m), 7.21-6.96 (1H, m), 4.61-4.23 (1H, m), 3.79 and 3.65 (3H, each s), 2.83-2.53 (3H, m), 2.04-1.19 (4H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 155.1, 142.0, 141.2, 132.8, 132.6, 129.7, 120.4, 52.9, 48.9, 34.4, 29.3, 26.0; IR (ATR) 1701, 1487, 1441, 1383, 1300 cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_{15}$BrNO$_2$ [M+H]$^+$ 284.0281; found 284.0280.

Methyl 6-bromo-8-nitro-3,4-dihydroquinoline-1(2*H*)-carboxylate (11)

To a solution of nitronium tetrafluoroborate (5.17 g, 38.9 mmol) in CH$_3$CN (150 mL) was added dropwise a solution of **9** (7.51 g, 27.8 mmol) in CH$_3$CN (150 mL) with cooling in an ice bath, and the mixture was stirred with cooling in an ice bath for 1 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with aqueous saturated NaHCO$_3$ and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in

vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (5:1, v/v) as eluent to give **11** (5.77 g, 66%) as a yellow solid.

Mp 99-101 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.88 (1H, s), 7.49 (1H, s), 3.81 (3H, br s), 3.64-3.62 (2H, br m), 2.85-2.71 (2H, br m), 2.07-1.92 (2H, br m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 154.0, 144.6, 137.5, 135.1, 131.1, 125.8, 116.6, 52.9, 44.1, 27.3, 23.5; IR (ATR) 1701, 1483, 1439, 1321, 1190 cm^{-1}; HRMS (ESI) m/z calcd for C$_{11}$H$_{12}$BrN$_2$O$_4$ [M+H]$^+$ 314.9975; found 314.9974.

Methyl 7-bromo-9-nitro-2,3,4,5-tetrahydro-1*H*-1-benzazepine-1-carboxylate (12)

Compound **12** was prepared from **10** (6.09 g, 21.4 mmol) in a manner similar to that described for compound **11** as a yellow solid (4.89 g, 72%).

Mp 73-75 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.91-7.86 (1H, m), 7.63-7.60 (1H, m), 4.53-4.27 (1H, m), 3.77 and 3.57 (3H, each s), 3.00-2.71 (3H, m), 2.12-1.78 (3H, m), 1.52-1.32 (1H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 154.9, 153.8, 147.5, 147.1, 145.1, 145.1, 136.9, 136.9, 134.7, 134.4, 125.8, 125.6, 120.2, 120.1, 53.4, 53.1, 48.5, 48.3, 34.3, 34.1, 29.1, 28.7, 25.4, 25.3; IR (ATR) 1713, 1522, 1303, 1169, 1032 cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_{14}$BrN$_2$O$_4$ [M+H]$^+$ 329.0131; found 329.0129.

8-Bromo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-2(1*H*)-one (13)

To a solution of reduced iron (24.1 g, 431 mmol) in AcOH (250 mL) was added dropwise a solution of **11** (19.4 g, 61.6 mmol) in AcOH (200 mL) at 80 °C. The reaction mixture was stirred at 80 °C for 2 h and cooled to room temperature. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was diluted with EtOAc and H$_2$O. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **13** (14.6 g, 94%) as a brown solid.

Mp 235-236 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 10.81 (1H, s), 6.99 (1H, s), 6.94 (1H, s), 3.68 (2H, t, J = 5.7 Hz), 2.76 (2H, t, J = 6.0 Hz), 2.04-1.94 (2H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 153.3, 127.9, 126.8, 121.2, 120.9, 112.0, 109.0, 38.1, 22.9, 21.2; IR (ATR) 3143, 1707, 1657, 1641, 1491 cm^{-1}; HRMS (ESI) m/z calcd for C$_{10}$H$_{10}$BrN$_2$O [M+H]$^+$ 252.9971; found 252.9967.

9-Bromo-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-2(1*H*)-one (14)

Compound **14** was prepared from **12** (2.53 g, 7.69 mmol) in a manner similar to that described for compound **13** as a white solid (1.73 g, 84%).

Mp 147-149 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 11.01 (1H, br s), 6.99 (1H, d, J = 1.5 Hz), 6.94 (1H, d, J = 2.0 Hz), 3.75 (2H, t, J = 5.2 Hz), 2.93 (2H, t, J = 5.6 Hz), 2.53-2.48 (4H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 154.5, 130.2, 128.8, 126.0, 123.8, 112.1, 108.7, 44.3, 33.3, 27.2, 26.6; IR (ATR) 2864, 1686, 1610, 1471, 1149 cm^{-1}; HRMS (ESI) m/z calcd for C$_{11}$H$_{12}$BrN$_2$O [M+H]$^+$ 267.0128; found 267.0126.

***tert*-Butyl (8-bromo-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetate (15)**

To a suspension of **13** (28.9 g, 114 mmol) and K_2CO_3 (23.7 g, 171 mmol) in DMF (400 mL) was added *tert*-butyl bromoacetate (18.5 mL, 126 mmol) with cooling in an ice bath. The reaction mixture was stirred at 50 °C for 3 h and cooled to room temperature. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was triturated with hexane to give **15** (41.5 g, 99%) as a beige solid.

Mp 178-180 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.02 (1H, d, *J* = 1.7 Hz), 6.87 (1H, d, *J* = 1.7 Hz), 4.47 (2H, s), 3.85 (2H, t, *J* = 5.7 Hz), 2.82 (2H, t, *J* = 6.1 Hz), 2.15-2.07 (2H, m), 1.48 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.7, 153.0, 128.7, 125.6, 122.7, 121.0, 113.6, 108.8, 82.9, 43.0, 39.1, 28.0, 23.6, 21.7; IR (ATR) 1741, 1697, 1498, 1421, 1232 cm^{-1}; HRMS (ESI) m/z calcd for $C_{16}H_{19}BrN_2O_3Na$ [M+Na]$^+$ 389.0471; found 389.0473; Anal. Calcd for $C_{16}H_{19}BrN_2O_3 \cdot 0.10H_2O$: C, 52.07; H, 5.24; N, 7.59; Br, 21.65. Found: C, 52.43; H, 5.29; N, 7.45; Br, 21.25.

***tert*-Butyl (9-bromo-2-oxo-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-1(2*H*)-yl)acetate (16)**

Compound **16** was prepared from **14** (1.71 g, 6.40 mmol) in a manner similar to that described for compound **15** as a white solid (2.42 g, 99%).

Mp 172-174 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.02 (1H, s), 6.84 (1H, s), 4.48 (2H, s), 3.95 (2H, t, *J* = 5.4 Hz), 2.98 (2H, t, *J* = 4.8 Hz), 2.06-1.93 (4H, m), 1.48 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.5, 154.4, 130.9, 127.8, 126.4, 125.5, 113.6, 108.3, 83.0, 45.6, 43.0, 34.1, 28.0, 27.7, 27.1; IR (ATR) 1741, 1693, 1425, 1230, 1149 cm^{-1}; HRMS (ESI) m/z calcd for $C_{17}H_{22}BrN_2O_3$ [M+H]$^+$ 381.0808; found 381.0804.

General Procedure A for the Suzuki-Miyaura Coupling Reaction

To a suspension of the appropriate tricyclic benzimidazolone or benzoxazolone (1 equiv.) and boronic acid (1.2-1.5 equiv.) in 1 M K_2CO_3 solution (3 equiv.) and 1,4-dioxane was added Pd(PPh$_3$)$_4$ (3-5 mol%) in room temperature. The reaction mixture was stirred at reflux for 2 h and cooled to room temperature. Water was then added, and the mixture was extracted with EtOAc. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography to afford tricyclic benzimidazolone or benzoxazolone derivative.

***tert*-Butyl (2-oxo-8-phenyl-5,6-dihydro-4H-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetate (17)**

Compound **17** was prepared from **15** (1.50 g, 4.09 mmol) and phenylboronic acid (0.647 g, 5.31 mmol) according to the general procedure A as a beige solid (0.929 g, 62%).

Mp 202-203 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.52 (2H, d, J = 8.0 Hz), 7.41 (2H, m), 7.31 (1H, t, J = 7.1 Hz), 7.09 (1H, s), 6.91 (1H, s), 4.54 (2H, s), 3.90 (2H, t, J = 5.7 Hz), 2.90 (2H, t, J = 6.0 Hz), 2.20-2.12 (2H, m), 1.47 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 167.0, 153.5, 142.2, 135.2, 128.7, 128.1, 127.3, 126.7, 126.2, 119.6, 119.4, 104.6, 82.6, 43.1, 39.2, 28.0, 23.9, 22.0; IR (ATR) 1741, 1713, 1701, 1228, 1155 cm^{-1}; HRMS (ESI) m/z calcd for C$_{22}$H$_{25}$N$_2$O$_3$ [M+H]$^+$ 365.1860; found 365.1855; Anal. Calcd for C$_{22}$H$_{24}$N$_2$O$_3$·0.25H$_2$O: C, 71.62; H, 6.69; N, 7.59. Found: C, 71.50; H, 6.64; N, 7.71.

tert-Butyl (2-oxo-9-phenyl-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-1(2*H*)-yl)acetate (18)

Compound **18** was prepared from **16** (1.84 g, 4.83 mmol) and phenylboronic acid (0.706 g, 5.79 mmol) according to the general procedure A as a white solid (1.74 g, 95%).

Mp 141-143 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.54 (2H, d, J = 7.3 Hz), 7.45-7.38 (2H, m), 7.32 (1H, t, J = 7.1 Hz), 7.10 (1H, s), 6.89 (1H, s), 4.56 (2H, s), 4.03-3.94 (2H, m), 3.11-3.04 (2H, m), 2.09-1.97 (4H, m), 1.47 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.8, 154.8, 141.3, 134.8, 130.3, 128.7, 128.2, 127.1, 126.8, 124.9, 122.3, 103.9, 82.7, 45.7, 43.1, 34.6, 28.0, 27.9, 27.4; IR (ATR) 1741, 1697, 1234, 1153, 754 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{27}$N$_2$O$_3$ [M+H]$^+$ 379.2016; found 379.2009.

General procedure B for the synthesis of the acetic acid derivatives

To a solution of the appropriate acetate (1 equiv.) in AcOH was added 4 N HCl in 1,4-dioxane (4 equiv.). The reaction mixture was stirred at 50 $^{\circ}$C for 4 h and cooled to room temperature. The solvent was removed in vacuo, and the resulting solid was triturated with hexane to afford the acetic acid derivative.

(2-Oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetic acid (19)

Compound **19** was prepared from **17** (19.6 g, 53.8 mmol) according to the general procedure B as a brown solid (13.6 g, 84%).

Mp 216-218 $^{\circ}$C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 13.03 (1H, br s), 7.64-7.60 (2H, m), 7.45-7.40 (2H, m), 7.34-7.27 (2H, m), 7.17 (1H, s), 4.64 (2H, s), 3.78 (2H, t, J = 5.6 Hz), 2.87 (2H, t, J = 5.9 Hz), 2.11-2.01 (2H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 169.7, 152.8, 141.3, 133.4, 128.7, 128.3, 126.6, 126.6, 125.8, 119.5, 118.1, 104.6, 42.0, 38.6, 23.3, 21.6; IR (ATR) 2364, 1728, 1668, 1659, 1643 cm^{-1}; HRMS (ESI) m/z calcd for C$_{18}$H$_{16}$N$_2$O$_3$Na [M+Na]$^+$ 331.1053; found 331.1046; Anal. Calcd for C$_{18}$H$_{16}$N$_2$O$_3$·0.50H$_2$O: C, 68.13; H, 5.40; N, 8.83. Found: C, 67.76; H, 5.19; N, 8.78.

(2-Oxo-9-phenyl-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-1(2*H*)-yl)acetic acid (20)

Compound **20** was prepared from **18** (1.57 g, 4.15 mmol) according to the general procedure B as a yellow solid (1.30 g, 97%).

Mp 207-209 $^{\circ}$C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 13.05 (1H, br s), 7.69-7.64 (2H, m), 7.46-7.40 (2H, m), 7.37 (1H, d, J = 1.2 Hz), 7.31 (1H, t, J = 7.3 Hz), 7.20 (1H, s), 4.68 (2H, s), 3.89-3.81 (2H, m), 3.09-3.00 (2H, m), 2.03-1.88 (4H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 169.6, 154.2, 140.3, 133.2, 130.5, 128.7, 127.8, 126.8, 126.5, 124.5, 120.9, 104.1, 45.0, 42.0, 33.8, 27.5, 26.9; IR (ATR) 2931, 1732, 1662, 1655, 1227 cm^{-1}; HRMS (ESI) m/z calcd for $C_{19}H_{19}N_2O_3$ [M+H]$^{+}$ 323.1390; found 323.1385.

General Procedure C for the Synthesis of amide derivatives

To a solution of the appropriate acetic acid (1 equiv.) in DMF were added selected amine (2.5 equiv.), 1-hydroxybenzotriazole (1 equiv.) and 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide hydrochloride (EDCI·HCl) (3 equiv.) at room temperature. The reaction mixture was stirred at room temperature or 50 $^{\circ}$C for 1 h and cooled to room temperature. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H$_2$O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography to afford amide derivatives.

General Procedure D for the Synthesis of amide derivatives

To a solution of the appropriate acetic acid (1 equiv.) in DMF were added selected amine (1.2 equiv.), phosphoric acid bis(2-oxooxazolidide) chloride (BOPCl) (1.2 equiv.) and triethylamine (TEA) (2.5 equiv.) at room temperature. The reaction mixture was stirred at room temperature for 3 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H$_2$O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography to afford amide derivatives.

General Procedure E for the Synthesis of amide derivatives

To a suspension of the appropriate acetic acid (1 equiv.) in CH$_2$Cl$_2$ were added oxalyl choride (1.1 equiv.) and DMF with cooling in an ice bath, and then the mixture was stirred at room temperature for 1 h. The solvent was removed in vacuo, and then the residue was azeotropied with toluene. A solution of acid chloride thus obtained in THF was added to a solution of selected amine (1.2 equiv.) and triethylamine (1.5 equiv.) in THF at room temperature, and then the mixture was stirred at room temperature for 1 h. The reaction was quenched by adding aqueous saturated NaHCO$_3$, and then the mixture was extracted with EtOAc. The organic layer was washed with H$_2$O and brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography to afford amide derivatives.

N-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-N-phenylacetamide (21a)

Compound **21a** was prepared from **19** (462 mg, 1.50 mmol) and *N*-methylaniline (244 μL, 2.25 mmol) according to the general procedure C as a white solid (460 mg, 77%).

Mp 126-127 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.56-7.51 (2H, m), 7.51-7.45 (2H, m), 7.45-7.38 (3H, m), 7.37-7.27 (3H, m), 7.06 (1H, s), 6.89 (1H, s), 4.41 (2H, s), 3.84 (2H, t, *J* = 5.7 Hz), 3.30 (3H, s), 2.88 (2H, t, *J* = 6.0 Hz), 2.18-2.08 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.6, 153.5, 142.4, 142.3, 135.1, 130.2, 128.6, 128.5, 128.5, 127.3, 127.3, 126.6, 126.2, 119.5, 119.2, 104.8, 43.1, 39.1, 37.8, 23.9, 21.9; IR (ATR) 1713, 1666, 1493, 1423, 762 cm^{-1}; HRMS (ESI) m/z calcd for C$_{25}$H$_{24}$N$_3$O$_2$ [M+H]$^+$ 398.1863; found 398.1855; Anal. Calcd for C$_{25}$H$_{23}$N$_3$O$_2$·0.25H$_2$O: C, 74.70; H, 5.89; N, 10.45. Found: C, 74.65; H, 5.83; N, 10.51.

2-(2-Oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-N,N-dipropylacetamide (21b)

Compound **21b** was prepared from **19** (308 mg, 1.00 mmol) and dipropylamine (165 μL, 1.20 mmol) according to the general procedure C as a white solid (244 mg, 62%)

Mp 129-131 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53 (2H, dd, *J* = 8.2, 1.3 Hz), 7.42-7.37 (2H, m), 7.32-7.27 (1H, m), 7.08 (1H, d, *J* = 1.2 Hz), 7.02 (1H, d, *J* = 1.2 Hz), 4.69 (2H, s), 3.90 (2H, t, *J* = 5.9 Hz), 3.39-3.24 (4H, m), 2.90 (2H, t, *J* = 6.0 Hz), 2.21-2.10 (2H, m), 1.72-1.50 (4H, m), 0.99 (3H, t, *J* = 7.4 Hz), 0.86 (3H, t, *J* = 7.4 Hz); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.2, 153.5, 142.3, 135.2, 128.7, 128.6, 127.3, 126.6, 126.1, 119.5, 119.2, 105.4, 49.2, 48.1, 42.8, 39.2, 24.0, 22.3, 22.0, 20.8, 11.4, 11.2; IR (ATR) 1705, 1649, 1230, 764, 702 cm^{-1}; HRMS (ESI) m/z calcd for C$_{24}$H$_{30}$N$_3$O$_2$ [M+H]$^+$ 392.2333; found 392.2324; Anal. Calcd for C$_{24}$H$_{29}$N$_3$O$_2$: C, 73.63; H, 7.47; N, 10.73. Found: C, 73.27; H, 7.44; N, 10.71.

N-Benzyl-N-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4H-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetamide (21c)

Compound **21c** was prepared from **19** (462 mg, 1.50 mmol) and benzylmethylamine (290 μL, 2.25 mmol) according to the general procedure C as a white solid (473 mg, 77%)

Mp 173-175 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.55 (2H, d, *J* = 8.0 Hz), 7.46-7.38 (2H, m), 7.34-7.21 (5H, m), 7.13-7.06 (2H, m), 7.03 (1H, s), 4.77-4.58 (4H, m), 3.91 (1H, t, *J* = 5.7 Hz), 3.73 (1H, t, *J* = 5.7 Hz), 3.05 and 3.01 (3H, each s), 2.91 (1H, t, *J* = 6.0 Hz), 2.84 (1H, t, *J* = 6.0 Hz), 2.21-2.13 (1H, m), 2.11-2.03 (1H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 167.5, 166.6, 153.5, 153.1, 142.2, 142.1, 136.7, 136.0, 135.3, 135.2, 128.8, 128.7, 128.6, 128.5, 128.4, 128.3, 127.6, 127.5, 127.3, 127.3, 126.7, 126.6, 126.2, 126.1, 125.8, 125.8, 119.6, 119.5, 119.3, 119.2, 105.4, 105.2, 52.9, 51.5, 43.4, 43.0, 39.2, 39.0, 34.7, 34.2, 24.0, 23.9, 22.0, 21.9; IR (ATR) 1693, 1653, 1493, 756, 704 cm^{-1}; HRMS (ESI) m/z calcd for C$_{26}$H$_{26}$N$_3$O$_2$

[M+H]$^+$ 412.2020; found 412.2007; Anal. Calcd for C$_{26}$H$_{25}$N$_3$O$_2$: C, 75.89; H, 6.12; N, 10.21. Found: C, 75.66; H, 6.02; N, 10.26.

N-(3-Methoxyphenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-ij]quinolin-1(2*H*)-yl)acetamide (21d)

Compound **21d** was prepared from **19** (308 mg, 1.00 mmol) and 3-methoxy-*N*-methylaniline (165 mg, 1.20 mmol) according to the general procedure E as a yellow solid (220 mg, 52%).

Mp 166-168 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.56-7.50 (2H, m), 7.46-7.28 (4H, m), 7.06 (1H, s), 6.96-6.88 (3H, m), 6.84-6.80 (1H, m), 4.46 (2H, s), 3.89-3.79 (5H, m), 3.29 (3H, s), 2.88 (2H, t, *J* = 6.0 Hz), 2.18-2.08 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.6, 160.8, 153.4, 143.5, 142.3, 135.1, 130.8, 128.6, 128.5, 127.3, 126.6, 126.2, 119.5, 119.3, 119.1, 114.1, 112.7, 104.8, 55.4, 43.1, 39.0, 37.7, 23.9, 21.9; IR (ATR) 1705, 1674, 1489, 1421, 754 cm^{-1}; HRMS (ESI) m/z calcd for C$_{26}$H$_{26}$N$_3$O$_3$ [M+H]$^+$ 428.1969; found 428.1962; Anal. Calcd for C$_{26}$H$_{25}$N$_3$O$_3$: C, 73.05; H, 5.89; N, 9.83. Found: C, 72.72; H, 5.94; N, 9.75.

N-(4-Methoxyphenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetamide (21e)

Compound **21e** was prepared from **19** (308 mg, 1.00 mmol) and *N*-methyl-*p*-anisidine (165 mg, 1.20 mmol) according to the general procedure E as a brown solid (348 mg, 81%).

Mp 172-174 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53 (2H, d, *J* = 7.3 Hz), 7.44-7.38 (2H, m), 7.31 (1H, t, *J* = 7.4 Hz), 7.28-7.24 (2H, m), 7.05 (1H, s), 6.97 (2H, d, *J* = 8.8 Hz), 6.88 (1H, s), 4.39 (2H, s), 3.89-3.80 (5H, m), 3.27 (3H, s), 2.88 (2H, t, *J* = 5.9 Hz), 2.18-2.09 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.9, 159.4, 153.5, 142.4, 135.1, 135.1, 128.6, 128.6, 128.4, 127.4, 126.6, 126.2, 119.5, 119.1, 115.3, 104.8, 55.6, 43.0, 39.1, 37.9, 23.9, 21.9; IR (ATR) 1691, 1662, 1504, 1425, 1238 cm^{-1}; HRMS (ESI) m/z calcd for C$_{26}$H$_{26}$N$_3$O$_3$ [M+H]$^+$ 428.1969; found 428.1965; Anal. Calcd for C$_{26}$H$_{25}$N$_3$O$_3$·0.25H$_2$O: C, 72.29; H, 5.95; N, 9.73. Found: C, 72.60; H, 5.87; N, 9.79.

N-(3-Chlorophenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl) acetamide (21f)

Compound **21f** was prepared from **19** (308 mg, 1.00 mmol) and 3-chloro-*N*-methylaniline (170 mg, 1.20 mmol) according to the general procedure C as a white solid (135 mg, 31%).

Mp 161-162 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.54 (2H, d, *J* = 7.8 Hz), 7.46-7.23 (7H, m), 7.08 (1H, s), 6.91 (1H, s), 4.45 (2H, s), 3.83 (2H, t, *J* = 5.4 Hz), 3.28 (3H, s), 2.89 (2H, t, *J* = 6.0 Hz), 2.19-2.09 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.5, 153.2, 143.5, 142.2, 135.4, 135.2, 131.1, 128.6, 128.3, 127.3, 126.7, 126.1, 125.6, 125.5, 123.6, 119.6, 119.3, 104.8, 43.3, 39.1, 37.9, 23.9, 21.9; IR (ATR) 1697,

1686, 1674, 760, 694 cm^{-1}; HRMS (ESI) m/z calcd for $C_{25}H_{23}ClN_3O_2$ [M+H]$^+$ 432.1473; found 432.1467; Anal. Calcd for $C_{25}H_{22}ClN_3O_2$: C, 69.52; H, 5.13; N, 9.73; Cl, 8.21. Found: C, 69.15; H, 5.15; N, 9.80; Cl, 8.12.

N-(4-Chlorophenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl) acetamide (21g)

Compound **21g** was prepared from **19** (308 mg, 1.00 mmol) and 4-chloro-*N*-methylaniline (145 μL, 1.20 mmol) according to the general procedure E as a pale yellow solid (306 mg, 71%).

Mp 178-180 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53 (2H, d, J = 7.6 Hz), 7.46-7.38 (4H, m), 7.31 (1H, t, J = 7.1 Hz), 7.28-7.22 (2H, m), 7.07 (1H, s), 6.89 (1H, s), 4.41 (2H, s), 3.83 (2H, t, J = 5.6 Hz), 3.27 (3H, s), 2.88 (2H, t, J = 6.0 Hz), 2.19-2.07 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.5, 153.3, 142.2, 140.9, 135.2, 134.3, 130.3, 128.6, 128.5, 128.4, 127.3, 126.6, 126.2, 119.6, 119.3, 104.8, 43.1, 39.1, 37.9, 23.9, 21.9; IR (ATR) 1695, 1670, 1489, 1425, 754 cm^{-1}; HRMS (ESI) m/z calcd for $C_{25}H_{23}ClN_3O_2$ [M+H]$^+$ 432.1473; found 432.1466; Anal. Calcd for $C_{25}H_{22}ClN_3O_2$: C, 69.52; H, 5.13; N, 9.73; Cl, 8.21. Found: C, 69.23; H, 5.15; N, 9.75; Cl, 8.22.

N-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-(pyridin-2-yl) acetamide (21h)

Compound **21h** was prepared from **19** (462 mg, 1.50 mmol) and 2-(methylamino)pyridine (185 μL, 1.80 mmol) according to the general procedure C as a white solid (331 mg, 55%).

Mp 171-172 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.50 (1H, d, J = 2.9 Hz), 7.79 (1H, t, J = 7.0 Hz), 7.54 (2H, d, J = 8.0 Hz), 7.44-7.19 (5H, m), 7.08 (1H, s), 7.01 (1H, s), 4.82 (2H, s), 3.87 (2H, t, J = 5.7 Hz), 3.44 (3H, s), 2.89 (2H, t, J = 6.0 Hz), 2.19-2.10 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 167.5, 155.1, 153.6, 148.7, 142.3, 138.6, 135.2, 128.7, 128.6, 127.3, 126.6, 126.2, 121.9, 119.4, 119.1, 119.1, 105.1, 44.2, 39.1, 35.4, 23.9, 22.0; IR (ATR) 1697, 1660, 1587, 1419, 1313 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{23}N_4O_2$ [M+H]$^+$ 399.1816; found 399.1810; Anal. Calcd for $C_{24}H_{22}N_4O_2$·0.25H$_2$O: C, 71.53; H, 5.63; N, 13.90. Found: C, 71.80; H, 5.65; N, 13.94.

N-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-(pyridin-3-yl) acetamide (21i)

Compound **21i** was prepared from **25** (308 mg, 1.00 mmol) and *N*-methy-3-pyridinamine (108 mg, 1.00 mmol) according to the general procedure C as a white solid (302 mg, 76%).

Mp 165-167 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.65 (1H, br s), 8.58 (1H, d, J = 2.7 Hz), 7.73 (1H, d, J = 8.0 Hz), 7.56-7.50 (2H, m), 7.47-7.37 (3H, m), 7.31 (1H, t, J = 7.3 Hz), 7.08 (1H, s), 6.93 (1H, s), 4.41 (2H, s), 3.86-3.78 (2H, m), 3.32 (3H, s), 2.88 (2H, t, J = 6.0 Hz), 2.19-2.08 (2H, m); ^{13}C-NMR (100

MHz, CDCl₃) δ: 166.7, 153.2, 149.5, 148.5, 142.1, 139.0, 135.3, 134.9, 128.6, 128.2, 127.3, 126.7, 126.1, 124.6, 119.7, 119.4, 104.8, 43.2, 39.1, 38.1, 23.9, 21.9; IR (ATR) 1709, 1666, 1493, 1423, 764 cm⁻¹; HRMS (ESI) m/z calcd for $C_{24}H_{23}N_4O_2$ [M+H]⁺ 399.1816; found 399.1810; Anal. Calcd for $C_{24}H_{22}N_4O_2 \cdot 0.25H_2O$: C, 71.53; H, 5.63; N, 13.90. Found: C, 71.43; H, 5.64; N, 13.87.

N-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4H-imidazo[4,5,1-ij]quinolin-1(2H)-yl)-N-(pyridin-4-yl) acetamide (21j)

Compound **21j** was prepared from **19** (308 mg, 1.00 mmol) and 4-(methylamino)pyridine (108 mg, 1.00 mmol) according to the general procedure C as a white solid (225 mg, 56%).

Mp 141-143 °C (iPrOH); ¹H-NMR (400 MHz, CDCl₃) δ 8.68 (2H, d, J = 5.9 Hz), 7.53 (2H, d, J = 7.1 Hz), 7.45-7.38 (2H, m), 7.34-7.23 (3H, m), 7.09 (1H, s), 6.92 (1H, s), 4.60 (2H, s), 3.83 (2H, t, J = 5.7 Hz), 3.37 (3H, s), 2.89 (2H, t, J = 6.0 Hz), 2.19-2.07 (2H, m); ¹³C-NMR (100 MHz, CDCl₃) δ: 166.4, 153.2, 151.5, 149.8, 142.1, 135.3, 128.7, 128.2, 127.3, 126.7, 126.1, 120.9, 119.7, 119.4, 104.8, 43.4, 39.1, 37.2, 23.9, 21.9; IR (ATR) 1693, 1672, 1587, 1491, 1429 cm⁻¹; HRMS (ESI) m/z calcd for $C_{24}H_{23}N_4O_2$ [M+H]⁺ 399.1816; found 399.1807; Anal. Calcd for $C_{24}H_{22}N_4O_2 \cdot 0.25H_2O$: C, 71.53; H, 5.63; N, 13.90. Found: C, 71.80; H, 5.56; N, 13.94.

2-(2-Oxo-8-phenyl-5,6-dihydro-4H-imidazo[4,5,1-ij]quinolin-1(2H)-yl)-N-phenylacetamide (21k)

Compound **21k** was prepared from **19** (462 mg, 1.50 mmol) and aniline (164 μL, 1.80 mmol) according to the general procedure C as a white solid (479 mg, 83%).

Mp 223-224 °C (MeOH); ¹H-NMR (400 MHz, CDCl₃) δ 8.49 (1H, br s), 7.58-7.47 (4H, m), 7.45-7.38 (2H, m), 7.36-7.23 (3H, m), 7.17 (2H, d, J = 6.1 Hz), 7.09 (1H, t, J = 7.4 Hz), 4.67 (2H, s), 3.95 (2H, t, J = 5.7 Hz), 2.94 (2H, t, J = 6.0 Hz), 2.24-2.16 (2H, m); ¹³C-NMR (100 MHz, CDCl₃) δ: 165.7, 154.0, 141.6, 137.4, 136.0, 129.0, 128.8, 127.9, 127.3, 127.0, 126.1, 124.6, 120.2, 120.1, 120.0, 105.1, 46.9, 39.4, 23.9, 22.0; IR (ATR) 1687, 1558, 1497, 1238, 700 cm⁻¹; HRMS (ESI) m/z calcd for $C_{24}H_{22}N_3O_2$ [M+H]⁺ 384.1707; found 384.1700; Anal. Calcd for $C_{24}H_{21}N_3O_2$: C, 75.18; H, 5.52; N, 10.96. Found: C, 75.07; H, 5.61; N, 11.00.

N-Methyl-2-(2-oxo-9-phenyl-4,5,6,7-tetrahydroimidazo[4,5,1-jk][1]benzazepin-1(2H)-yl)-N-phenyl acetamide (22)

Compound **22** was prepared from **20** (129 mg, 0.400 mmol) and N-methylaniline (43.3 μL, 0.400 mmol) according to the general procedure C as a beige solid (164 mg, quant.).

Mp 157-159 °C (MeOH); ¹H-NMR (400 MHz, CDCl₃) δ 7.54 (2H, d, J = 7.1 Hz), 7.51-7.46 (2H, m), 7.46-7.29 (6H, m), 7.07 (1H, s), 6.88 (1H, s), 4.42 (2H, s), 3.96-3.87 (2H, m), 3.30 (3H, s), 3.09-3.01 (2H, m), 2.07-1.95 (4H, m); ¹³C-NMR (100 MHz, CDCl₃) δ: 166.4, 154.8, 142.4, 141.5, 134.7, 130.7, 130.2,

128.7, 128.5, 128.2, 127.2, 127.2, 126.8, 124.8, 122.1, 104.1, 45.6, 43.1, 37.8, 34.6, 27.9, 27.4; IR (ATR) 1707, 1662, 1425, 758, 700 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{26}N_3O_2$ [M+H]$^+$ 412.2020; found 412.2013; Anal. Calcd for $C_{26}H_{25}N_3O_2$: C, 75.89; H, 6.12; N, 10.21. Found: C, 75.78; H, 6.16; N, 10.23.

(8-Bromo-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetic acid (23)

Compound **23** was prepared from **15** (16.7 g, 45.5 mmol) according to the general procedure B as a yellow solid (14.1 g, quant.).

Mp 213-215 ^{o}C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 13.08 (1H, br s), 7.28 (1H, d, *J* = 1.7 Hz), 7.05 (1H, d, *J* = 1.7 Hz), 4.58 (2H, s), 3.74 (2H, t, *J* = 5.7 Hz), 2.79 (2H, t, *J* = 6.0 Hz), 2.05-1.95 (2H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 169.5, 152.4, 128.9, 125.3, 121.8, 121.2, 112.4, 109.0, 42.0, 38.5, 22.9, 21.2; IR (ATR) 1718, 1653, 1635, 1624, 1427 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{11}BrN_2O_3Na$ [M+Na]$^+$ 332.9845; found 332.9841.

2-(8-Bromo-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-methyl-*N*-phenylacetamide (24)

Compound **24** was prepared from **23** (11.2 g, 36.0 mmol) and *N*-methylaniline (4.68 mL, 43.2 mmol) according to the general procedure C as a yellow solid (11.3 g, 78%).

Mp 195-197 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53-7.46 (2H, m), 7.42 (1H, t, *J* = 7.1 Hz), 7.35 (2H, d, *J* = 7.8 Hz), 6.98 (1H, s), 6.84 (1H, s), 4.32 (2H, s), 3.80 (2H, t, *J* = 5.9 Hz), 3.31 (3H, s), 2.79 (2H, t, *J* = 6.0 Hz), 2.13-2.03 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.2, 153.1, 142.3, 130.2, 129.1, 128.6, 127.3, 125.6, 122.5, 120.9, 113.6, 108.9, 43.0, 39.0, 37.8, 23.6, 21.6; IR (ATR) 1705, 1662, 1497, 1421, 1406 cm^{-1}; HRMS (ESI) m/z calcd for $C_{19}H_{19}BrN_3O_2$ [M+H]$^+$ 400.0655; found 400.0652; Anal. Calcd for $C_{19}H_{18}BrN_3O_2 \cdot 0.25H_2O$: C, 56.38; H, 4.61; N, 10.38; Br, 19.74. Found: C, 56.11; H, 4.51; N, 10.25; Br, 20.03.

2-[8-(3-Methoxyphenyl)-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-methyl-*N*-phenylacetamide (25)

Compound **25** was prepared from **24** (200 mg, 0.500 mmol) and 3-methoxyphenylboronic acid (98.8 mg, 0.650 mmol) according to the general procedure A as a white solid (105 mg, 49%).

Mp 221-223 ^{o}C (MeOH); ^{1}H-NMR (CDCl$_3$) δ 7.52-7.46 (2H, m), 7.41 (1H, t, *J* = 7.2 Hz), 7.37-7.30 (3H, m), 7.12 (1H, d, *J* = 7.8 Hz), 7.08-7.04 (2H, m), 6.90-6.84 (2H, m), 4.40 (2H, s), 3.87 (3H, s), 3.84 (2H, t, *J* = 5.9 Hz), 3.30 (3H, s), 2.88 (2H, t, *J* = 6.0 Hz), 2.18-2.08 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.6, 159.9, 153.5, 143.9, 142.5, 138.1, 135.0, 130.2, 129.6, 128.5, 127.3, 126.4, 120.0, 119.5, 119.2, 113.3, 111.9, 104.8, 55.4, 43.1, 39.1, 37.8, 23.9, 22.0; IR (ATR) 1705, 1660, 1491, 1429, 1236 cm^{-1}; HRMS (ESI)

m/z calcd for $C_{26}H_{26}N_3O_3$ [M+H]$^+$ 428.1969; found 428.1960; Anal. Calcd for $C_{26}H_{25}N_3O_3\cdot0.25H_2O$: C, 72.29; H, 5.95; N, 9.73. Found: C, 72.09; H, 6.02; N, 9.39.

2-[8-(4-Methoxyphenyl)-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-methyl-*N*-phenylacetamide (26)

   Compound **26** was prepared from **24** (200 mg, 0.500 mmol) and 4-methoxyphenylboronic acid (98.8 mg, 0.650 mmol) according to the general procedure A as a white solid (165 mg, 77%).
Mp 138-140 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.51-7.38 (5H, m), 7.34 (2H, d, *J* = 7.3 Hz), 7.01 (1H, s), 6.96 (2H, d, *J* = 8.8 Hz), 6.84 (1H, s), 4.40 (2H, s), 3.85 (3H, s), 3.83 (2H, t, *J* = 5.9 Hz), 3.30 (3H, s), 2.87 (2H, t, *J* = 6.0 Hz), 2.16-2.08 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.7, 158.7, 153.5, 142.5, 135.0, 134.8, 130.2, 128.5, 128.5, 128.3, 127.3, 125.8, 119.5, 118.8, 114.1, 104.5, 55.4, 43.1, 39.1, 37.8, 24.0, 22.0; IR (ATR) 1695, 1668, 1497, 1242, 825 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{26}N_3O_3$ [M+H]$^+$ 428.1969; found 428.1960; Anal. Calcd for $C_{26}H_{25}N_3O_3\cdot0.75H_2O$: C, 70.81; H, 6.06; N, 9.53. Found: C, 70.59; H, 5.91; N, 9.55.

N-Methyl-2-{2-oxo-8-[3-(trifluoromethyl)phenyl]-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl}-*N*-phenylacetamide (27)

   Compound **27** was prepared from **24** (200 mg, 0.500 mmol) and 3-(trifluoromethyl)phenylboronic acid (123 mg, 0.650 mmol) according to the general procedure A as a white solid (207 mg, 89%).
Mp 259-260 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.77 (1H, s), 7.71 (1H, d, *J* = 7.3 Hz), 7.58-7.47 (4H, m), 7.42 (1H, t, *J* = 7.1 Hz), 7.36 (2H, d, *J* = 7.3 Hz), 7.06 (1H, s), 6.89 (1H, s), 4.42 (2H, s), 3.85 (2H, t, *J* = 5.6 Hz), 3.31 (3H, s), 2.89 (2H, t, *J* = 6.0 Hz), 2.20-2.09 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.6, 153.5, 143.1, 142.4, 133.5, 130.8 (m), 130.6, 130.2, 129.1, 128.7, 128.5, 127.3, 126.8, 124.0 (m), 123.2 (m), 122.9 (m), 119.8, 119.2, 104.8, 43.0, 39.1, 37.8, 23.9, 21.9; IR (ATR) 1716, 1660, 1421, 1329, 1117 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{23}F_3N_3O_2$ [M+H]$^+$ 466.1737; found 466.1726; Anal. Calcd for $C_{26}H_{22}F_3N_3O_2$: C, 67.09; H, 4.76; N, 9.03; F, 12.24. Found: C, 66.97; H, 4.78; N, 9.03; F, 12.26.

N-Methyl-2-{2-oxo-8-[4-(trifluoromethyl)phenyl]-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl}-*N*-phenylacetamide (28)

   Compound **28** was prepared from **24** (200 mg, 0.500 mmol) and 4-(trifluoromethyl)phenylboronic acid (123 mg, 0.650 mmol) according to the general procedure A as a white solid (196 mg, 84%).
Mp 207-209 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.68-7.61 (4H, m), 7.53-7.46 (2H, m), 7.42 (1H, t, *J* = 7.1 Hz), 7.36 (2H, d, *J* = 7.3 Hz), 7.07 (1H, s), 6.90 (1H, s), 4.41 (2H, s), 3.85 (2H, t, *J* = 5.7 Hz), 3.31 (3H, s), 2.89 (2H, t, *J* = 6.0 Hz), 2.20-2.10 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.5, 153.5, 145.8, 142.4, 133.5, 130.2, 128.7 (m), 128.6 (m), 127.5, 127.3, 126.9, 125.6 (m), 125.5, 125.5 (m), 119.7, 119.3,

104.9, 43.1, 39.1, 37.8, 23.9, 21.9; IR (ATR) 1708, 1660, 1323, 1111, 1065 cm^{-1}; HRMS (ESI) m/z calcd for C$_{26}$H$_{23}$F$_3$N$_3$O$_2$ [M+H]$^+$ 466.1737; found 466.1724; Anal. Calcd for C$_{26}$H$_{22}$F$_3$N$_3$O$_2$·0.50H$_2$O: C, 65.82; H, 4.89; N, 8.86; F, 12.01. Found: C, 65.69; H, 4.81; N, 8.87; F, 11.95.

N-Methyl-2-[2-oxo-8-(pyridin-2-yl)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-phenyl acetamide (29)

To a solution of **24** (200 mg, 0.500 mmol) and 2-(tributylstannyl)pyridine (0.192 mL, 0.600 mmol) in toluene (3.0 mL) was added Pd(PPh$_3$)$_4$ (28.9 mg, 0.025 mmol) at room temperature. The reaction mixture was stirred at reflux for 14.5 h and cooled to room temperature. Water was then added, and the mixture was extracted with EtOAc. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using EtOAc/MeOH (10:1, v/v) as eluent to give **29** (42.9 mg, 22%) as a white solid.

Mp 200-202 $^{\circ}$C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.65 (1H, d, *J* = 4.9 Hz), 7.74-7.65 (2H, m), 7.53-7.45 (3H, m), 7.44-7.33 (4H, m), 7.20-7.15 (1H, m), 4.44 (2H, s), 3.85 (2H, t, *J* = 5.7 Hz), 3.30 (3H, s), 2.90 (2H, t, *J* = 6.0 Hz), 2.18-2.09 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.6, 158.3, 153.6, 149.4, 142.4, 136.7, 133.1, 130.2, 128.7, 128.5, 127.7, 127.4, 121.4, 120.5, 119.4, 119.0, 104.7, 43.2, 39.1, 37.8, 24.0, 21.9; IR (ATR) 1704, 1666, 1425, 781, 706 cm^{-1}; HRMS (ESI) m/z calcd for C$_{24}$H$_{23}$N$_4$O$_2$ [M+H]$^+$ 399.1816; found 399.1805; Anal. Calcd for C$_{24}$H$_{22}$N$_4$O$_2$·0.50H$_2$O: C, 70.74; H, 5.69; N, 13.75. Found: C, 70.55; H, 5.49; N, 13.72.

N-Methyl-2-[2-oxo-8-(pyridin-3-yl)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-phenyl acetamide (30)

Compound **30** was prepared from **24** (200 mg, 0.500 mmol) and 3-pyridineboronic acid (79.9 mg, 0.650 mmol) according to the general procedure A as a white solid (199 mg, quant.).

Mp 130-132 $^{\circ}$C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.79 (1H, s), 8.55 (1H, d, *J* = 4.9 Hz), 7.87-7.79 (1H, m), 7.54-7.31 (6H, m), 7.05 (1H, s), 6.88 (1H, s), 4.42 (2H, s), 3.86 (2H, t, *J* = 5.7 Hz), 3.31 (3H, s), 2.90 (2H, t, *J* = 6.0 Hz), 2.20-2.10 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.4, 153.4, 147.4, 146.7, 142.3, 138.2, 135.3, 130.8, 130.2, 128.9, 128.6, 127.3, 127.0, 123.8, 120.0, 119.1, 104.7, 43.1, 39.1, 37.8, 23.9, 21.8; IR (ATR) 1691, 1659, 1643, 1491, 1431 cm^{-1}; HRMS (ESI) m/z calcd for C$_{24}$H$_{23}$N$_4$O$_2$ [M+H]$^+$ 399.1816; found 399.1810; Anal. Calcd for C$_{24}$H$_{22}$N$_4$O$_2$·1.50H$_2$O: C, 67.75; H, 5.92; N, 13.17. Found: C, 67.41; H, 5.84; N, 13.02.

N-Methyl-2-[2-oxo-8-(pyridin-4-yl)-5,6-dihydro-4H-imidazo[4,5,1-ij]quinolin-1(2H)-yl]-N-phenyl acetamide (31)

Compound **31** was prepared from **24** (100 mg, 0.250 mmol) and 4-pyridineboronic acid (36.9 mg, 0.300 mmol) according to the general procedure A as a white solid (39.7 mg, 40%).

Mp 238-239 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.62 (2H, d, J = 6.3 Hz), 7.57 (2H, d, J = 6.3 Hz), 7.54-7.48 (2H, m), 7.43 (1H, t, J = 7.1 Hz), 7.37 (2H, d, J = 7.6 Hz), 7.16 (1H, s), 6.99 (1H, s), 4.42 (2H, s), 3.86 (2H, t, J = 5.7 Hz), 3.32 (3H, s), 2.91 (2H, t, J = 6.0 Hz), 2.21-2.08 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.5, 153.4, 151.2, 148.1, 142.3, 130.8, 130.3, 129.0, 128.7, 128.2, 127.3, 122.1, 120.0, 119.3, 104.7, 43.1, 39.1, 37.8, 23.9, 21.8; IR (ATR) 1709, 1695, 1662, 1497, 1429 cm^{-1}; HRMS (ESI) m/z calcd for C$_{24}$H$_{23}$N$_4$O$_2$ [M+H]$^+$ 399.1816; found 399.1803; Anal. Calcd for C$_{24}$H$_{22}$N$_4$O$_2$·1.75H$_2$O: C, 67.04; H, 5.98; N, 13.03. Found: C, 66.77; H, 5.75; N, 12.83.

N-Methyl-2-[2-oxo-8-(pyridin-3-ylamino)-5,6-dihydro-4H-imidazo[4,5,1-ij]quinolin-1(2H)-yl]-N-phenylacetamide (32)

A mixture of **24** (200 mg, 0.500 mmol), 3-aminopyridine (70.6 mg, 0.750 mmol), Pd$_2$(dba)$_3$ (22.9 mg, 0.0250 mmol), Xantphos (43.4 mg, 0.0750 mmol) and Cs$_2$CO$_3$ (228 mg, 0.700 mmol) in toluene (4.0 mL) was heated at reflux for 11 h and cooled to room temperature. The reaction was quenched by adding aqueous saturated NaHCO$_3$, and then the mixture was extracted with CH$_2$Cl$_2$. The organic layer was washed with brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using EtOAc/MeOH (20:1, v/v) as eluent to give **32** (110 mg, 53%) as a yellow solid.

Mp 194-196 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.25 (1H, d, J = 2.7 Hz), 8.06 (1H, dd, J = 4.4, 1.2 Hz), 7.50-7.44 (2H, m), 7.40 (1H, t, J = 6.7 Hz), 7.32 (2H, d, J = 7.3 Hz), 7.20-7.15 (1H, m), 7.12-7.06 (1H, m), 6.65 (1H, s), 6.54 (1H, s), 4.32 (2H, s), 3.80 (2H, t, J = 5.7 Hz), 3.29 (3H, s), 2.77 (2H, t, J = 6.0 Hz), 2.14-2.04 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.6, 153.4, 142.3, 142.2, 140.4, 138.2, 135.4, 130.2, 128.8, 128.5, 127.3, 123.7, 123.3, 121.2, 120.1, 113.9, 100.6, 43.0, 39.0, 37.8, 23.8, 21.9; IR (ATR) 1695, 1662, 1506, 1429, 694 cm^{-1}; HRMS (ESI) m/z calcd for C$_{24}$H$_{24}$N$_5$O$_2$ [M+H]$^+$ 414.1925; found 414.1911; Anal. Calcd for C$_{24}$H$_{23}$N$_5$O$_2$·0.25H$_2$O: C, 68.96; H, 5.67; N, 16.76. Found: C, 69.21; H, 5.63; N, 16.47.

N-Methyl-2-(2-oxo-5,6-dihydro-4H-imidazo[4,5,1-ij]quinolin-1(2H)-yl)-N-phenylacetamide (33)

To a solution of **24** (100 mg, 0.250 mmol) in MeOH (5.0 mL) and THF (5.0 mL) was added 10% Pd/C (50% wet, 10.0 mg), and stirred at room temperature for 1 h under hydrogen atmosphere. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was recrystallized from iPrOH to give **33** (27.6 mg, 34%) as a white solid.

Mp 173-174 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.51-7.44 (2H, m), 7.40 (1H, t, J = 7.7 Hz), 7.32 (2H, d, J = 7.1 Hz), 6.95 (1H, dd, J = 7.7, 7.7 Hz), 6.83 (1H, d, J = 7.7 Hz), 6.71 (1H, d, J = 7.7 Hz), 4.37 (2H, s), 3.81 (2H, t, J = 5.9 Hz), 3.30 (3H, s), 2.83 (2H, t, J = 6.1 Hz), 2.14-2.06 (2H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.7, 153.3, 142.5, 130.2, 128.5, 128.1, 127.3, 126.6, 120.8, 119.5, 119.5, 105.5, 43.1, 39.0, 37.8, 23.9, 21.9; IR (ATR) 1699, 1660, 1497, 1421, 739 cm^{-1}; HRMS (ESI) m/z calcd for C$_{19}$H$_{20}$N$_3$O$_2$ [M+H]$^+$ 322.1550; found 322.1548; Anal. Calcd for C$_{19}$H$_{19}$N$_3$O$_2$·0.25H$_2$O: C, 70.03; H, 6.03; N, 12.89. Found: C, 69.90; H, 5.94; N, 12.82.

tert-Butyl (2-amino-3-nitrophenoxy)acetate (35)

To a suspension of 2-amino-3-nitrophenol (2.51 g, 16.3 mmol) and K$_2$CO$_3$ (3.15 g, 22.8 mmol) in DMF (15 mL) was added *tert*-butyl bromoacetate (2.55 mL, 17.3 mmol) with cooling in an ice bath. The reaction mixture was stirred at room temperature for 2.5 h. Water was then added, and the mixture was extracted with toluene. The organic layer was washed with H$_2$O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using Hexane/EtOAc (10:1, v/v) as eluent to give **35** (3.59 g, 82%) as an orange solid.

Mp 103-105 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.79 (1H, dd, J = 8.8, 1.2 Hz), 6.82 (1H, d, J = 7.8 Hz), 6.62-6.52 (3H, m), 4.58 (2H, s), 1.50 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 167.2, 146.8, 137.6, 132.0, 118.8, 115.8, 114.2, 83.0, 67.0, 28.0; IR (ATR) 3479, 1736, 1628, 1522, 11151 cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_{17}$N$_2$O$_5$ [M+H]$^+$ 269.1132; found 269.1127.

tert-Butyl (2-amino-5-bromo-3-nitrophenoxy)acetate (36)

To a solution of **35** (2.54 g, 9.47 mmol) in DMF (15 mL) was added *N*-bromosuccinimide (1.77 g, 9.94 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 3 h. Water was then added, and the mixture was extracted with Et$_2$O. The organic layer was washed with H$_2$O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **36** (3.26 g, 99%) as a brown solid.

Mp 73-74 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.95 (1H, d, J = 2.2 Hz), 6.89 (1H, s), 6.61 (2H, br s), 4.58 (2H, s), 1.51 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.6, 147.3, 136.8, 131.9, 120.8, 118.9, 105.5, 83.4, 67.0, 28.0; IR (ATR) 3369, 1743, 1516, 1209, 1151 cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_{16}$BrN$_2$O$_5$ [M+H]$^+$ 347.0237; found 347.0236.

7-Bromo-5-nitro-2*H*-1,4-benzoxazin-3(4*H*)-one (37)

To a solution of **36** (2.33 g, 6.71 mmol) in toluene (10 mL) was added *p*-toluenesulfonic acid monohydrate (0.100 g, 0.526 mmol) at room temperature The reaction mixture was stirred at 80 °C for 2 h, and cooled to room temperature. The reaction mixture was concentrated, and the residue was diluted with

CH_2Cl_2 and aqueous saturated $NaHCO_3$. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **37** (1.82 g, 99%) as a yellow solid.

Mp 179-180 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 10.50 (1H, s), 7.91 (1H, d, J = 2.2 Hz), 7.68 (1H, d, J = 2.2 Hz), 4.78 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 164.2, 146.2, 135.7, 124.6, 123.3, 120.9, 113.1, 66.7; IR (ATR) 2343, 1699, 1684, 1338, 1286 cm^{-1}; HRMS (ESI) m/z calcd for $C_8H_4BrN_2O_4$ [M-H]⁻ 270.9360; found 270.9352.

5-Nitro-7-phenyl-2*H*-1,4-benzoxazin-3(4*H*)-one (38)

To a suspension of **37** (1.30 g, 4.76 mmol) and phenylboronic acid (0.697 g, 5.71 mmol) in 2 M K_2CO_3 solution (7.14 mL, 14.3 mmol) and 1,4-dioxane (15 mL) was added Pd(PPh$_3$)$_4$ (275 mg, 0.238 mmol) at room temperature. The reaction mixture was stirred at reflux for 3 h and cooled to room temperature. 1 M HCl solution was then added, and the separated solid was collected by filtration to give [(4-amino-5-nitrobiphenyl-3-yl)oxy]acetic acid (921 mg, 67%) as a brown solid. To a suspension of [(4-amino-5-nitrobiphenyl-3-yl)oxy]acetic acid (900 mg, 3.12 mmol) in toluene (15 mL) was added *p*-toluenesulfonic acid monohydrate (0.119 g, 0.624 mmol) at room temperature The reaction mixture was stirred at reflux for 1.5 h, and cooled to room temperature. The reaction mixture was concentrated, and the residue was diluted with CH_2Cl_2 and aqueous saturated $NaHCO_3$. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **38** (718 mg, 85%) as a yellow solid.

Mp 163-165 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 10.47 (1H, s), 8.01 (1H, d, J = 2.2 Hz), 7.78-7.71 (3H, m), 7.52-7.45 (2H, m), 7.42 (1H, t, J = 7.1 Hz), 4.80 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 164.5, 145.7, 137.0, 135.9, 134.5, 129.1, 128.3, 126.5, 122.7, 119.9, 116.3, 66.6; IR (ATR) 3261, 1716, 1699, 1338, 1176 cm^{-1}; HRMS (ESI) m/z calcd for $C_{14}H_9N_2O_4$ [M-H]⁻ 269.0568; found 269.0561.

7-Phenyl-3,4-dihydro-2*H*-1,4-benzoxazin-5-amine (39)

To a solution of LiAlH$_4$ (387 mg, 10.2 mmol) in THF (15 mL) was added dropwise a solution of **38** (689 mg, 2.55 mm0l) in THF (25 mL) at 50 °C. The reaction mixture was stirred at 50 °C for 3 h and cooled to room temperature. The reaction was then quenched by dropwise addition of H_2O (0.400 mL), 15% NaOH solution (0.400 mL) and H_2O (1.20 mL) with cooling in an ice bath. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was purified by silica gel column chromatography using hexane/EtOAc (1:4, v/v) as eluent to give **55** (279 mg, 48%) as a brown oil.

^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.55-7.47 (2H, m), 7.41-7.34 (2H, m), 7.30-7.23 (1H, m), 6.65 (1H, d, J = 2.0 Hz), 6.60 (1H, d, J = 2.0 Hz), 4.23 (2H, t, J = 4.4 Hz), 3.46 (2H, t, J = 4.4 Hz); ^{13}C-NMR (100 MHz,

CDCl$_3$) δ: 145.9, 141.2, 136.5, 133.4, 128.5, 126.6, 126.5, 121.0, 107.6, 107.2, 65.2, 41.7; IR (ATR) 3334, 2870, 1487, 1340, 754 cm^{-1}; HRMS (ESI) m/z calcd for C$_{14}$H$_{15}$N$_2$O [M+H]$^+$ 227.1179; found 227.1176.

8-Phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-2(1*H*)-one (40)

To a solution of **39** (263 mg, 1.16 mmol) in THF (5.0 mL) was added 1,1'-carbonyldiimidazole (226 mg, 1.39 mmol) at room temperature. The mixture was stirred at reflux for 1 h and cooled to room temperature. The reaction was then quenched by adding 2 M HCl solution, and the mixture was extracted with EtOAc. The organic layer was washed with brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The residue was purified by silica gel column chromatography using EtOAc as eluent to give **40** (234 mg, 80%) as a brown solid.

Mp 195-197 $^{\circ}$C; ^{1}H-NMR (400 MHz, DMSO-*d$_6$*) δ 10.84 (1H, s), 7.60-7.53 (2H, m), 7.45-7.37 (2H, m), 7.30 (1H, t, *J* = 7.3 Hz), 6.83 (2H, s), 4.41 (2H, t, *J* = 4.5 Hz), 3.92 (2H, t, *J* = 4.5 Hz); ^{13}C-NMR (100 MHz, DMSO-*d$_6$*) δ: 153.0, 141.3, 140.4, 134.3, 128.8, 128.3, 126.8, 126.7, 117.2, 105.5, 101.0, 65.9; IR (ATR) 3000, 1686, 1660, 1490, 1410 cm^{-1}; HRMS (ESI) m/z calcd for C$_{15}$H$_{13}$N$_2$O$_2$ [M+H]$^+$ 253.0972; found 253.0965.

tert-Butyl (2-oxo-8-phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-1(2*H*)-yl)acetate (41)

Compound **41** was prepared from **40** (228 mg, 0.904 mmol) in a manner similar to that described for compound **15** as a beige solid (259 mg, 78%).

Mp 160-162 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.51 (2H, d, *J* = 7.6 Hz), 7.44-7.38 (2H, m), 7.32 (1H, t, *J* = 7.6 Hz), 6.88 (1H, s), 6.73 (1H, s), 4.54 (2H, s), 4.44 (2H, t, *J* = 4.6 Hz), 4.05 (2H, t, *J* = 4.6 Hz), 1.48 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.9, 152.6, 141.9, 140.9, 136.5, 129.2, 128.7, 127.4, 127.0, 115.8, 107.5, 100.8, 82.8, 65.8, 43.4, 39.7, 28.0; IR (ATR) 1741, 1707, 1653, 1234, 1157 cm^{-1}; HRMS (ESI) m/z calcd for C$_{21}$H$_{23}$N$_2$O$_4$ [M+H]$^+$ 367.1652; found 367.1648.

(2-Oxo-8-phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-1(2*H*)-yl)acetic acid (42)

Compound **42** was prepared from **41** (239 mg, 0.652 mmol) according to the general procedure B as a beige solid (202 mg, quant.): mp 226-228 $^{\circ}$C; ^{1}H-NMR (400 MHz, DMSO-*d$_6$*) δ 13.08 (1H, br s), 7.61 (2H, t, *J* = 4.3 Hz), 7.46-7.39 (2H, m), 7.31 (1H, t, *J* = 7.3 Hz), 7.16 (1H, d, *J* = 1.0 Hz), 6.91 (1H, d, *J* = 1.0 Hz), 4.64 (2H, s), 4.44 (2H, t, *J* = 4.6 Hz), 3.98 (2H, t, *J* = 4.6 Hz); ^{13}C-NMR (100 MHz, DMSO-*d$_6$*) δ: 169.6, 152.0, 141.0, 140.6, 134.5, 129.5, 128.8, 126.8, 126.8, 115.6, 105.8, 100.8, 65.8, 42.3, 39.2; IR (ATR) 2875, 1718, 1647, 1201, 1034 cm^{-1}; HRMS (ESI) m/z calcd for C$_{17}$H$_{15}$N$_2$O$_4$ [M+H]$^+$ 311.1026; found 311.1022.

N-Methyl-2-(2-oxo-8-phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-1(2*H*)-yl)-*N*-phenylacet amide (43)

Compound **43** was prepared from **42** (55.0 mg, 0.177 mmol) and *N*-methylaniline (19.2 μL, 0.177 mmol) according to the general procedure C as a white solid (50.1 mg, 71%).

Mp 166-167 $^\circ$C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.55-7.46 (4H, m), 7.45-7.38 (3H, m), 7.37-7.29 (3H, m), 6.85 (1H, s), 6.71 (1H, s), 4.46-4.35 (4H, m), 3.99 (2H, t, *J* = 4.6 Hz), 3.31 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.3, 152.6, 142.3, 142.0, 140.8, 136.3, 130.2, 129.5, 128.6, 128.5, 127.4, 127.3, 126.9, 115.8, 107.3, 101.0, 65.8, 43.4, 39.6, 37.8; IR (ATR) 1718, 1662, 1497, 1284, 1192 cm^{-1}; HRMS (ESI) m/z calcd for C$_{24}$H$_{22}$N$_3$O$_3$ [M+H]$^+$ 400.1656; found 400.1648; Anal. Calcd for C$_{24}$H$_{21}$N$_3$O$_3$: C, 72.16; H, 5.30; N, 10.52. Found: C, 71.80; H, 5.43; N, 10.37.

6-1-3 Experiments in Chapter 3

5-Phenyl-1,3-benzoxazol-2(3*H*)-one (45)

To a solution of 2-amino-4-phenylphenol (6.10 g, 32.9 mmol) in THF (150 mL) was added 1,1'-carbonyldiimidazole (6.41 g, 39.5 mml) at room temperature. The mixture was stirred at reflux for 2 h and cooled to room temperature. The reaction was then quenched by adding 2 M HCl solution, and the mixture was extracted with EtOAc. The organic layer was washed with brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo to give **45** (6.89 g, 99%) as a white solid.

Mp 156-157 $^\circ$C; ^{1}H-NMR (400MHz, CDCl$_3$) δ 8.98 (1H, br s), 7.56-7.52 (2H, m), 7.48-7.42 (2H, m), 7.40-7.32 (2H, m), 7.30 (1H, d, *J* = 1.7 Hz), 7.27 (1H, d, *J* = 8.3 Hz); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 155.6, 143.4, 140.4, 138.1, 129.7, 128.9, 127.5, 127.2, 121.9, 110.3, 108.7; IR (ATR) 3176, 1763, 1466, 1254, 949 cm^{-1}; HRMS (ESI) m/z calcd for C$_{13}$H$_8$NO$_2$ [M-H]$^-$ 210.0561; found 210.0556.

tert-Butyl (2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetate (46)

To a suspension of **45** (3.00 g, 14.2 mmol) and K$_2$CO$_3$ (2.94 g, 21.3 mmol) in acetone (30 mL) was added *tert*-butyl bromoacetate (2.31 mL, 15.6 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 1 day. The reaction mixture was filtrated, and the filtrate was concentrated. The resulting solid was triturated with Et$_2$O to give **46** (4.46 g, 97%) as a yellow solid.

Mp 99-100 $^\circ$C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.55-7.50 (2H, m), 7.45 (2H, dd, *J* = 7.4, 7.4 Hz), 7.40-7.32 (2H, m), 7.28 (1H, d, *J* = 8.5 Hz), 7.05 (1H, d, *J* = 1.7 Hz), 4.50 (2H, s), 1.47 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.6, 154.7, 142.1, 140.6, 138.0, 131.3, 128.9, 127.5, 127.2, 122.0, 110.3, 107.2, 83.5, 43.9, 28.0; IR (ATR) 1759, 1743, 1485, 1230, 1026 cm^{-1}; HRMS (ESI) m/z calcd for C$_{19}$H$_{20}$NO$_4$ [M+H]$^+$ 326.1387; found 326.1395; Anal. Calcd for C$_{19}$H$_{19}$NO$_4$·0.25H$_2$O: C, 69.18; H, 5.96; N, 4.25. Found: C, 69.32; H, 5.85; N, 4.64.

(2-Oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetic acid (47)

Compound **47** was prepared from **46** (4.00 g, 12.3 mmol) according to the general procedure B as a beige solid (3.24 g, 98%).

Mp 153-155 $^{\circ}$C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 13.38 (1H, br s), 7.72 (1H, s), 7.68 (2H, dd, J = 8.2, 1.1 Hz), 7.51-7.42 (4H, m), 7.37 (1H, t, J = 7.3 Hz), 4.73 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.8, 154.1, 141.5, 139.6, 136.6, 131.8, 128.9, 127.5, 126.8, 120.9, 110.0, 108.0, 42.9; IR (ATR) 2353, 1763, 1728, 1483, 1241 cm^{-1}; HRMS (ESI) m/z calcd for $C_{15}H_{12}NO_4$ [M+H]$^+$ 270.0761; found 270.0756; Anal. Calcd for $C_{15}H_{11}NO_4$: C, 66.91; H, 4.12; N, 5.20. Found: C, 66.77; H, 4.17; N, 5.24.

N-Methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)-*N*-phenylacetamide (48a)

Compound **48a** was prepared from **47** (500 mg, 1.86 mmol) and *N*-methylaniline (241 μL, 2.23 mmol) according to the general procedure C as a white solid   (547 mg, 82%).

Mp 123-125 $^{\circ}$C (*i*PrOH); ^{1}H-NMR (400MHz, CDCl$_3$) δ 7.56-7.49 (4H, m), 7.48-7.41 (3H, m), 7.39-7.32 (3H, m), 7.30 (1H, dd, J = 8.3, 1.7 Hz), 7.24 (1H, d, J = 8.3 Hz), 7.04 (1H, d, J = 1.2 Hz), 4.36 (2H, s), 3.32 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.8, 142.2, 142.0, 140.8, 137.9, 131.7, 130.4, 128.9, 128.8, 127.4, 127.3, 127.2, 121.8, 110.1, 107.5, 43.9, 37.8; IR (ATR) 1778, 1662, 1485, 1385, 1120 cm^{-1}; HRMS (ESI) m/z calcd for $C_{22}H_{19}N_2O_3$ [M+H]$^+$ 359.1390; found 359.1381; Anal. Calcd for $C_{22}H_{18}N_2O_3 \cdot 0.25H_2O$: C, 72.81; H, 5.14; N, 7.72. Found: C, 72.85; H, 5.05; N, 7.81.

2-(2-Oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (48b)

Compound **48b** was prepared from **47** (269 mg, 1.00 mmol) and 30% ammonia solution (568 mg, 10.0 mmol) according to the general procedure E as a white solid (262 mg, 98%).

Mp   208-209 $^{\circ}$C (*i*PrOH); ^{1}H-NMR (400MHz, DMSO-d_6) δ 7.78 (1H, br s), 7.67 (2H, d, J = 8.0 Hz), 7.56 (1H, s), 7.51-7.41 (4H, m), 7.41-7.34 (2H, m), 4.53 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 167.7, 154.2, 141.6, 139.7, 136.4, 132.3, 128.9, 127.4, 126.7, 120.7, 109.8, 107.8, 43.9; IR (ATR) 1792, 1686, 1483, 1383, 1252 cm^{-1}; HRMS (ESI) m/z calcd for $C_{15}H_{13}N_2O_3$ [M+H]$^+$ 269.0921; found 269.0920; Anal. Calcd for $C_{15}H_{12}N_2O_3$: C, 67.16; H, 4.51; N, 10.44. Found: C, 67.25; H, 4.64; N, 10.31.

2-(2-Oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)-*N*-phenylacetamide (48c)

Compound **48c** was prepared from **47** (108mg, 0.400 mmol) and aniline (43.7 μL, 0.480 mmol) according to the general procedure C as a white solid (75.7 mg, 55%).

Mp 231-234 $^{\circ}$C (*i*PrOH); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 10.43 (1H, s), 7.76-7.64 (2H, m), 7.62-7.56 (2H, m), 7.56-7.39 (6H, m), 7.39-7.27 (2H, m), 7.11-7.00 (1H, m), 4.81 (1H, s), 4.54 (1H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 169.3, 164.5, 154.2, 154.2, 153.4, 153.4, 141.5, 139.6, 139.3, 138.4, 136.4, 132.2, 132.1, 130.8, 128.8, 128.8, 128.7, 127.8, 127.3, 126.9, 126.8, 126.7, 126.7, 126.6, 125.8, 123.8, 123.5,

120.7, 119.1, 117.3, 109.8, 107.9, 51.6, 44.5; IR (ATR) 1755, 1686, 1560, 1481, 1246 cm^{-1}; HRMS (ESI) m/z calcd for $C_{21}H_{17}N_2O_3$ [M+H]$^+$ 345.1234; found 345.1228; Anal. Calcd for $C_{21}H_{16}N_2O_3 \cdot 0.25H_2O$: C, 72.30; H, 4.77; N, 8.03. Found: C, 72.54; H, 4.74; N, 8.29.

2-(2-Oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)-*N,N*-dipropylacetamide (48d)

Compound **48d** was prepared from **47** (269mg, 1.00 mmol) and dipropylamine (137 μL, 1.00 mmol) according to the general procedure C as a white solid (219 mg, 62%).

Mp 100-102 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.52 (2H, d, *J* = 8.3 Hz), 7.42 (2H, m), 7.37-7.29 (2H, m), 7.25 (1H, d, *J* = 7.3 Hz), 7.13 (1H, s), 4.65 (2H, s), 3.34-3.29 (4H, m), 1.70 (2H, tq, *J* = 7.4, 7.4 Hz), 1.57 (2H, tq, *J* = 7.4, 7.4 Hz), 1.01 (3H, t, *J* = 7.4 Hz), 0.88 (3H, t, *J* = 7.4 Hz); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.7, 154.9, 142.2, 140.7, 138.0, 131.8, 128.8, 127.4, 127.3, 121.8, 110.1, 108.0, 49.2, 48.2, 43.5, 22.3, 20.8, 11.3, 11.2; IR (ATR) 1790, 1772, 1647, 1485, 1147 cm^{-1}; HRMS (ESI) m/z calcd for $C_{21}H_{25}N_2O_3$ [M+H]$^+$ 353.1860; found 353.1859; Anal. Calcd for $C_{21}H_{24}N_2O_3 \cdot 0.25H_2O$: C, 70.66; H, 6.92; N, 7.85. Found: C, 71.04; H, 6.81; N, 7.73.

N-Benzyl-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (48e)

Compound **48e** was prepared from **47** (269 mg, 1.00 mmol) and benzylmethylamine (129 μL, 1.00 mmol) according to the general procedure C as a white solid (202 mg, 54%).

Mp 154-156 ^{o}C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53 (2H, dd, *J* = 7.2. 7.2 Hz), 7.48-7.20 (10H, m), 7.14 and 7.05 (1H, each d, each *J* = 1.7 Hz), 4.71 and 4.68 (2H, each s), 4.68 and 4.61 (2H, each s), 3.05 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.7, 165.1, 154.9, 154.8, 142.2, 140.7, 140.7, 139.8, 138.0, 137.9, 136.3, 135.6, 131.7, 129.3, 128.8, 128.3, 128.1, 127.8, 127.5, 127.4, 127.3, 126.0, 122.4, 122.4, 122.3, 122.0, 121.9, 121.8, 110.2, 110.2, 108.0, 107.8, 52.9, 51.6, 43.8, 43.6, 34.9, 34.1; IR (ATR) 1768, 1749, 1649, 1483, 1026 cm^{-1}; HRMS (ESI) m/z calcd for $C_{23}H_{21}N_2O_3$ [M+H]$^+$ 373.1547; found 373.1544; Anal. Calcd for $C_{23}H_{20}N_2O_3$: C, 74.18; H, 5.41; N, 7.52. Found: C, 74.42; H, 5.40; N, 7.69.

N-(3-Methoxyphenyl)-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (48f)

Compound **48f** was prepared from **47** (269 mg, 1.00 mmol) and 3-methoxy-*N*-methylaniline (165 mg, 1.20 mmol) according to the general procedure E as a white solid (193 mg, 50%).

Mp 158-160 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53 (2H, d, *J* = 8.0 Hz), 7.47-7.33 (4H, m), 7.32-7.21 (2H, m), 7.04 (1H, s), 6.96 (1H, d, *J* = 7.8 Hz), 6.91 (1H, d, *J* = 7.8 Hz), 6.86 (1H, s), 4.41 (2H, s), 3.85 (3H, s), 3.31 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 161.0, 154.8, 143.1, 142.2, 140.8, 137.9, 131.7, 131.1, 128.8, 127.4, 127.3, 121.8, 119.2, 114.3, 113.0, 110.1, 107.5, 55.5, 43.8, 37.7; IR (ATR) 1782, 1674, 1485, 1383, 1039 cm^{-1}; HRMS (ESI) m/z calcd for $C_{23}H_{21}N_2O_4$ [M+H]$^+$ 389.1496;

found 389.1491; Anal. Calcd for $C_{23}H_{20}N_2O_4$: C, 71.12; H, 5.19; N, 7.21. Found: C, 70.93; H, 5.26; N, 7.23.

N-(4-Methoxyphenyl)-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (48g)

Compound **48g** was prepared from **47** (269 mg, 1.00 mmol) and *N*-methyl-*p*-anisidine (165 mg, 1.20 mmol) according to the general procedure C as a white solid (316 mg, 81%).

Mp 137-139 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53 (2H, d, J = 7.3 Hz), 7.44 (2H, dd, J = 7.3, 7.3 Hz), 7.36 (1H, t, J = 7.3 Hz), 7.31-7.22 (4H, m), 7.04-6.98 (3H, m), 4.34 (2H, s), 3.85 (3H, s), 3.28 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.4, 159.6, 154.8, 142.2, 140.8, 137.9, 134.6, 131.7, 128.8, 128.4, 127.4, 127.3, 121.8, 115.5, 110.1, 107.5, 55.6, 43.8, 37.9; IR (ATR) 1770, 1670, 1508, 1248, 1022 cm^{-1}; HRMS (ESI) m/z calcd for $C_{23}H_{21}N_2O_4$ [M+H]$^+$ 389.1496; found 389.1492; Anal. Calcd for $C_{23}H_{20}N_2O_4$: C, 71.12; H, 5.19; N, 7.21. Found: C, 70.99; H, 5.24; N, 7.27.

N-(3-Chlorophenyl)-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (48h)

Compound **48h** was prepared from **47** (269 mg, 1.00 mmol) and 3-chloro-*N*-methylaniline (170 mg, 1.20 mmol) according to the general procedure C as a white solid (116 mg, 30%).

Mp 157-159 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.55-7.51 (2H, m), 7.47-7.41 (4H, m), 7.39-7.33 (2H, m), 7.31 (1H, dd, J = 8.3, 1.7 Hz), 7.28-7.23 (2H, m), 7.05 (1H, s), 4.38 (2H, s), 3.30 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.0, 154.7, 143.1, 142.2, 140.7, 138.0, 135.9, 131.6, 131.4, 129.2, 128.8, 127.6, 127.4, 127.3, 125.6, 121.9, 110.2, 107.5, 43.9, 37.8; IR (ATR) 1770, 1672, 1481, 1250, 1020 cm^{-1}; HRMS (ESI) m/z calcd for $C_{22}H_{18}ClN_2O_3$ [M+H]$^+$ 393.1000; found 393.0999; Anal. Calcd for $C_{22}H_{17}ClN_2O_3$: C, 67.26; H, 4.36; N, 7.13; Cl, 9.02. Found: C, 67.08; H, 4.39; N, 7.21; Cl, 8.83.

N-(4-Chlorophenyl)-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (48i)

Compound **48i** was prepared from **47** (269 mg, 1.00 mmol) and 4-chloro-*N*-methylaniline (145 μL, 1.20 mmol) according to the general procedure E as a white solid (283 mg, 72%).

Mp 83-85 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.55-7.41 (6H, m), 7.36 (1H, t, J = 7.3 Hz), 7.32-7.22 (4H, m), 7.03 (1H, s), 4.34 (2H, s), 3.29 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.0, 154.7, 142.2, 140.7, 140.5, 138.0, 134.9, 131.6, 130.7, 128.8, 128.6, 127.4, 127.3, 121.9, 110.2, 107.5, 43.8, 37.8; IR (ATR) 1768, 1670, 1483, 1250, 1090 cm^{-1}; HRMS (ESI) m/z calcd for $C_{22}H_{18}ClN_2O_3$ [M+H]$^+$ 393.1000; found 393.1001; Anal. Calcd for $C_{22}H_{17}ClN_2O_3$: C, 67.26; H, 4.36; N, 7.13; Cl, 9.02. Found: C, 67.03; H, 4.40; N, 7.22; Cl, 8.81.

N-Methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl)-N-(pyridin-2-yl)acetamide (48j)

Compound **48j** was prepared from **47** (539 mg, 2.00 mmol) and 2-(methylamino)pyridine (247 μL, 2.40 mmol) according to the general procedure C as a white solid (302 mg, 42%).

Mp 94-96 °C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.53-8.46 (1H, m), 7.87-7.80 (1H, m), 7.57-7.52 (2H, m), 7.43 (2H, dd, J = 7.8, 7.8 Hz), 7.38-7.22 (5H, m), 7.17 (1H, d, J = 1.7 Hz), 4.83 (2H, s), 3.43 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 166.2, 154.9, 154.8, 148.6, 142.2, 140.7, 139.0, 137.9, 131.9, 128.8, 127.4, 127.3, 122.1, 121.7, 118.6, 110.1, 107.8, 45.2, 35.5; IR (ATR) 1772, 1670, 1589, 1252, 1022 cm^{-1}; HRMS (ESI) m/z calcd for C$_{21}$H$_{18}$N$_3$O$_3$ [M+H]$^+$ 360.1343; found 360.1341; Anal. Calcd for C$_{21}$H$_{17}$N$_3$O$_3$·0.75H$_2$O: C, 67.64; H, 5.00; N, 11.27. Found: C, 68.00; H, 5.10; N, 11.48.

N-Methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl)-N-(pyridin-3-yl)acetamide (48k)

Compound **48k** was prepared from **47** (539 mg, 2.00 mmol) and N-methy-3-pyridinamine (216 mg, 2.00 mmol) according to the general procedure C as a white solid (280 mg, 39%).

Mp 112-114 °C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.70 (1H, d, J = 4.4 Hz), 8.66 (1H, s), 7.74 (1H, d, J = 8.0 Hz), 7.56-7.40 (5H, m), 7.39-7.22 (3H, m), 7.06 (1H, s), 4.34 (2H, s), 3.34 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.6, 150.0, 148.6, 142.1, 140.6, 138.6, 138.0, 134.9, 131.5, 128.8, 127.5, 127.3, 124.8, 122.0, 110.3, 107.5, 43.9, 38.1; IR (ATR) 1780, 1670, 1485, 1381, 1097 cm^{-1}; HRMS (ESI) m/z calcd for C$_{21}$H$_{18}$N$_3$O$_3$ [M+H]$^+$ 360.1343; found 360.1341; Anal. Calcd for C$_{21}$H$_{17}$N$_3$O$_3$·H$_2$O: C, 66.83; H, 5.07; N, 11.13. Found: C, 66.71; H, 5.15; N, 11.18.

N-Methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl)-N-(pyridin-4-yl)acetamide (48l)

Compound **48l** was prepared from **47** (539 mg, 2.00 mmol) and 4-(methylamino)pyridine (260 mg, 2.40 mmol) according to the general procedure C as a white solid (616 mg, 86%).

Mp 148-149 °C (iPrOH-Et$_2$O); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.74 (2H, d, J = 5.6 Hz), 7.54-7.50 (2H, m), 7.43 (2H, dd, J = 7.6, 7.6 Hz), 7.38-7.29 (4H, m), 7.27-7.24 (1H, m), 7.06 (1H, d, J = 1.7 Hz), 4.51 (2H, s), 3.38 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.8, 154.7, 151.9, 149.5, 142.1, 140.6, 138.1, 131.4, 128.8, 127.5, 127.3, 122.0, 121.1, 110.3, 107.5, 43.9, 37.2; IR (ATR) 1795, 1780, 1664, 1581, 1483 cm^{-1}; HRMS (ESI) m/z calcd for C$_{21}$H$_{18}$N$_3$O$_3$ [M+H]$^+$ 360.1343; found 360.1342; Anal. Calcd for C$_{21}$H$_{17}$N$_3$O$_3$: C, 70.18; H, 4.77; N, 11.69. Found: C, 70.09; H, 4.86; N, 11.68.

N-[2-(Hydroxymethyl)phenyl]-N-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl)acetamide (49a)

Compound **49a** was prepared from **47** (539 mg, 2.00 mmol) and 2-(methylaminophenyl)methanol (274 mg, 2.00 mmol) according to the general procedure C as a white solid (393 mg, 51%).

Mp 93-94 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.62 (1H, dd, J = 7.2, 1.8 Hz), 7.54-7.38 (6H, m), 7.35-7.25 (3H, m), 7.21 (1H, d, J = 8.3 Hz), 7.17 (1H, d, J = 1.7 Hz), 4.76 (1H, d, J = 12.7 Hz), 4.62 (1H, d, J = 12.7

Hz), 4.38 (1H, d, J = 16.8 Hz), 4.20 (1H, d, J = 16.8 Hz), 3.24 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.9, 155.1, 142.1, 140.6, 139.9, 138.3, 137.8, 131.8, 131.1, 129.8, 129.8, 128.8, 128.2, 127.3, 127.2, 121.7, 110.1, 107.8, 60.9, 43.8, 37.2; IR (ATR) 3419, 1778, 1664, 1483, 1383 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{21}$N$_2$O$_4$ [M+H]$^+$ 389.1496; found 389.1486.

N-[3-(Hydroxymethyl)phenyl]-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (49b)

Compound **49b** was prepared from **47** (539 mg, 2.00 mmol) and 3-(methylaminophenyl)methanol (274 mg, 2.00 mmol) according to the general procedure C as a white solid (495 mg, 64%).

Mp 140-141 ^{o}C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.53 (2H, d, J = 7.3 Hz), 7.47-7.41 (3H, m), 7.40-7.33 (3H, m), 7.29 (1H, dd, J = 8.3, 1.7 Hz), 7.20 (1H, d, J = 8.3 Hz), 7.17 (1H, d, J = 7.6 Hz), 7.09 (1H, d, J = 1.7 Hz), 4.74 (2H, s), 4.37 (2H, s), 3.30 (3H, s), 2.54 (1H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.2, 154.6, 143.8, 142.0, 140.6, 137.9, 131.5, 130.3, 128.8, 127.4, 127.3, 127.0, 125.7, 125.3, 121.8, 110.1, 107.7, 64.2, 44.0, 37.8; IR (ATR) 3439, 1767, 1657, 1481, 1397 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{21}$N$_2$O$_4$ [M+H]$^+$ 389.1496; found 389.1489.

N-[4-(Hydroxymethyl)phenyl]-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (49c)

Compound **49c** was prepared from **47** (241 mg, 0.897 mmol) and 4-(methylaminophenyl)methanol (123 mg, 0.897 mmol) according to the general procedure C as a white solid (241 mg, 69%).

Mp 161-162 ^{o}C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.56-7.48 (4H, m), 7.44 (2H, dd, J = 7.6, 7.6 Hz), 7.39-7.28 (4H, m), 7.24 (1H, d, J = 8.3 Hz), 7.04 (1H, s), 4.77 (2H, d, J = 5.6 Hz), 4.36 (2H, s), 3.31 (3H, s), 1.90 (1H, t, J = 5.6 Hz); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.2, 154.8, 142.2, 141.8, 141.2, 140.8, 137.9, 131.7, 128.8, 128.8, 127.4, 127.4, 127.3, 121.8, 110.2, 107.6, 64.4, 43.9, 37.8; IR (ATR) 2360, 1778, 1655, 1483, 1244 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{21}$N$_2$O$_4$ [M+H]$^+$ 389.1496; found 389.1488.

N-{2-[(Dimethylamino)methyl]phenyl}-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl) acetamide (48m)

To a suspension of **49a** (200 mg, 0.515 mmol) and carbon tetrabromide (427 mg, 1.29 mmol) in CH$_3$CN (7.0 mL) was added triphenylphosphine (338 mg, 1.29 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 30 min. The solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (2:3, v/v) as eluent to give *N*-[2-(Bromomethyl)phenyl]-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (**50a**) (226 mg, 97%) as a white solid. Compound **50a** (150 mg, 0.332 mmol) was dissolved in DMF (3.0 mL), then 50% methylamine solution　(150 mg, 1.66 mmol) was added to the solution with cooling in an ice bath, and the mixture was stirred at room temperature for 30 min. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H$_2$O and brine,

and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using CHCl$_3$/MeOH (50:1, v/v) as eluent. The solvent was removed in vacuo, and the resulting solid was recrystallized from *i*PrOH to give **48m** (134 mg, 97%) as a white solid.

Mp 134-135 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.54-7.48 (3H, m), 7.46-7.40 (4H, m), 7.38-7.31 (2H, m), 7.29-7.26 (1H, m), 7.23 (1H, d, *J* = 8.3 Hz), 7.05 (1H, d, *J* = 1.7 Hz), 4.43 (1H, d, *J* = 17.1 Hz), 4.36 (1H, d, *J* = 17.1 Hz), 3.64 (1H, d, *J* = 12.9 Hz), 3.28 (3H, s), 3.18 (1H, d, *J* = 12.9 Hz), 2.27 (6H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.9, 154.9, 142.2, 141.2, 141.0, 137.9, 136.8, 132.2, 132.0, 129.5, 129.1, 128.8, 128.7, 127.4, 127.3, 121.7, 110.0, 107.8, 60.2, 45.7, 44.0, 37.0; IR (ATR) 1778, 1662, 1484, 756, 694 cm^{-1}; HRMS (ESI) m/z calcd for C$_{25}$H$_{26}$N$_3$O$_3$ [M+H]$^+$ 416.1969; found 416.1960; Anal. Calcd for C$_{25}$H$_{25}$N$_3$O$_3$: C, 72.27; H, 6.06; N, 10.11. Found: C, 72.26; H, 6.10; N, 10.20.

N-{3-[(Dimethylamino)methyl]phenyl}-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl) acetamide (48n)

To a suspension of **49b** (200 mg, 0.515 mmol) and carbon tetrabromide (427 mg, 1.29 mmol) in CH$_3$CN (7.0 mL) was added triphenylphosphine (338 mg, 1.29 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 1 h. The solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (2:3, v/v) as eluent to give *N*-[3-(Bromomethyl)phenyl]-*N*-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2*H*)-yl)acetamide (**50b**) (209 mg, 90%) as a white solid. Compound **50b** (150 mg, 0.332 mmol) was dissolved in DMF (3.0 mL), then 50% methylamine solution (150 mg, 1.66 mmol) was added to the solution with cooling in an ice bath, and the mixture was stirred at room temperature for 1 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H$_2$O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using CHCl$_3$/MeOH (50:1, v/v) as eluent to give **48n** (63.7 mg, 46%) as a colorless oil.

^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.54 (2H, d, *J* = 7.1 Hz), 7.48-7.42 (3H, m), 7.40-7.20 (6H, m), 7.06 (1H, s), 4.37 (2H, s), 3.48 (2H, s), 3.32 (3H, s), 2.27 (6H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.7, 142.2, 142.1, 142.0, 140.8, 137.9, 131.7, 131.1, 130.2, 129.3, 128.8, 127.4, 127.3, 125.7, 121.8, 110.1, 107.6, 63.7, 45.4, 43.9, 37.8; IR (ATR) 1772, 1670, 1481, 756, 698 cm^{-1}; HRMS (ESI) m/z calcd for C$_{25}$H$_{26}$N$_3$O$_3$ [M+H]$^+$ 416.1969; found 416.1960; Anal. Calcd for C$_{25}$H$_{25}$N$_3$O$_3$·0.25H$_2$O: C, 71.49; H, 6.12; N, 10.01. Found: C, 71.30; H, 6.04; N, 10.01.

N-{4-[(Dimethylamino)methyl]phenyl}-N-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl) acetamide (48o)

To a suspension of **49c** (150 mg, 0.386 mmol) and carbon tetrabromide (320 mg, 0.965 mmol) in CH₃CN (5.0 mL) was added triphenylphosphine (253 mg, 0.965 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 1 h. The solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (2:3, v/v) as eluent to give N-[4-(Bromomethyl)phenyl]-N-methyl-2-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl)acetamide (**50c**) (151 mg, 87%) as a white solid. Compound **50c** (100 mg, 0.222 mmol) was dissolved in DMF (2.0 mL), then 50% methylamine solution (99.9 mg, 1.11 mmol) was added to the solution with cooling in an ice bath, and the mixture was stirred at room temperature for 1 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using CHCl₃/MeOH (50:1, v/v) as eluent to give **48o** (90.2 mg, 98%) as a colorless oil.

^{1}H-NMR (400 MHz, CDCl₃) δ 7.53 (2H, d, J = 7.1 Hz), 7.45 (4H, m), 7.36 (1H, t, J = 6.7 Hz), 7.32-7.26 (3H, m), 7.23 (1H, d, J = 8.3 Hz), 7.03 (1H, s), 4.36 (2H, s), 3.47 (2H, s), 3.31 (3H, s), 2.27 (6H, s); ^{13}C-NMR (100 MHz, CDCl₃) δ: 165.1, 154.7, 142.2, 140.8, 140.7, 140.2, 137.9, 131.7, 130.8, 128.8, 127.4, 127.3, 127.0, 121.8, 110.1, 107.5, 63.6, 45.5, 43.9, 37.8; IR (ATR) 1772, 1670, 1481, 1383, 1250 cm⁻¹; HRMS (ESI) m/z calcd for $C_{25}H_{26}N_3O_3$ [M+H]⁺ 416.1969; found 416.1960; Anal. Calcd for $C_{25}H_{25}N_3O_3 \cdot 0.25H_2O$: C, 71.49; H, 6.12; N, 10.01. Found: C, 71.84; H, 6.12; N, 10.12.

N-Methyl-3-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl)-N-phenylpropanamide (51)

To a suspension of **45** (106 mg, 0.500 mmol) and K_2CO_3 (117 mg, 0.850 mmol) in DMF (1.0 mL) was added a solution of 3-bromo-N-methyl-N-phenylpropanamide (72.6 mg, 0.300 mmol) in DMF (1.0 mL) with cooling in an ice bath. The reaction mixture was stirred at 80 °C for 6 h and cooled to room temperature. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (3:1, v/v) as eluent to give **51** (116 mg, 62%) as a yellow solid.

Mp 96-97 °C; ^{1}H-NMR (400 MHz, CDCl₃) δ 7.56 (2H, d, J = 7.6 Hz), 7.46 (2H, dd, J = 7.6, 7.6 Hz), 7.41-7.34 (3H, m), 7.34-7.28 (3H, m), 7.22 (1H, d, J = 8.3 Hz), 7.06 (2H, d, J = 7.6 Hz), 4.15 (2H, t, J = 7.0 Hz), 3.23 (3H, s), 2.58 (2H, t, J = 7.0 Hz); ^{13}C-NMR (100 MHz, CDCl₃) δ: 169.9, 154.5, 143.2, 142.1, 140.5, 137.8, 131.5, 130.1, 128.9, 128.2, 127.5, 127.2, 127.1, 121.4, 110.0, 107.7, 38.8, 37.3, 32.2; IR (ATR) 1759, 1655, 1481, 758, 698 cm⁻¹; HRMS (ESI) m/z calcd for $C_{23}H_{21}N_2O_3$ [M+H]⁺ 373.1547; found

373.1538; Anal. Calcd for $C_{23}H_{20}N_2O_3 \cdot 0.25H_2O$: C, 73.29; H, 5.48; N, 7.43. Found: C, 73.64; H, 5.39; N, 7.58.

N-Methyl-4-(2-oxo-5-phenyl-1,3-benzoxazol-3(2H)-yl)-N-phenylbutanamide (52)

Compound **52** was prepared from **45** (31.7 mg, 0.150 mmol) and 3-bromo-N-methyl-N-phenyl butanamide (46.1 mg, 0.180 mmol) in a manner similar to that described for compound **51** as a white solid (13.5 mg, 23%).

Mp 153-155 ^{o}C; ^{1}H-NMR (400MHz, CDCl$_3$) δ 7.57 (2H, d, J = 7.3 Hz), 7.46 (2H, dd, J = 7.3, 7.3 Hz), 7.40-7.20 (7H, m), 7.06-7.01 (2H, m), 3.90 (2H, t, J = 6.6 Hz), 3.21 (3H, s), 2.17-2.03 (4H, m); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 171.5, 154.7, 143.6, 142.1, 140.6, 137.8, 131.8, 129.8, 128.9, 128.0, 127.5, 127.2, 127.2, 121.3, 110.0, 107.4, 41.5, 37.2, 30.7, 23.2; IR (ATR) 1774, 1651, 1485, 1259, 702 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{23}N_2O_3$ [M+H]$^+$ 387.1703; found 387.1701; Anal. Calcd for $C_{24}H_{22}N_2O_3 \cdot 0.50H_2O$: C, 72.89; H, 5.86; N, 7.08. Found: C, 73.06; H, 5.77; N, 7.08.

3-(2-Oxo-2-phenylethyl)-5-phenyl-1,3-benzoxazol-2(3H)-one (53)

To a suspension of **45** (500 mg, 2.37 mmol) and K$_2$CO$_3$ (491 mg, 3.55 mmol) in DMF (5.0 mL) was added 2-bromoacetophenone (518 mg, 2.60 mmol) at room temperature. The reaction mixture was stirred at room temperature for 15 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H$_2$O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (7:3, v/v) as eluent to give **53** (524 mg, 67%) as a brown solid.

Mp 54-56 ^{o}C; ^{1}H-NMR (400MHz, CDCl$_3$) δ 8.06-8.02 (2H, m), 7.67 (1H, t, J = 7.4 Hz), 7.54 (2H, dd, J = 7.4, 7.4 Hz), 7.51-7.47 (2H, m), 7.43-7.37 (2H, m), 7.36-7.26 (3H, m), 6.99 (1H, d, J = 1.2 Hz), 5.28 (2H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 190.6, 154.9, 142.3, 140.5, 138.0, 134.4, 134.1, 131.6, 129.1, 128.8, 128.2, 127.5, 127.3, 121.9, 110.3, 107.5, 48.1; IR (ATR) 1772, 1695, 1481, 1344, 1227 cm^{-1}; HRMS (ESI) m/z calcd for $C_{21}H_{16}NO_3$ [M+H]$^+$ 330.1125; found 330.1126; Anal. Calcd for $C_{21}H_{15}NO_3 \cdot 0.10H_2O$: C, 76.17; H, 4.63; N, 4.23. Found: C, 76.11; H, 4.70; N, 4.25.

3-{2-[Methyl(phenyl)amino]ethyl}-5-phenyl-1,3-benzoxazol-2(3H)-one (54)

To a solution of **48a** (100 mg, 0.279 mmol) in THF (2.0 mL) was added dropwise 1.0 M boran·THF complex in THF (0.977 mL, 0.977 mmol) with cooling in an ice bath. The reaction mixture was stirred at reflux for 1.5 h and cooled to room temperature. The reaction was then quenched by dropwise addition of MeOH (1.0 mL), and the solvent was removed in vacuo. The residue was dissolve in MeOH (2.0 mL). To a solution thus obtained was added 4 N HCl in 1,4-dioxane (0.279 mM, 1.17 mmol) at room temperature,

and heated at reflux for 1 h. The reaction mixture was cooled and a solvent was removed in vacuo. The residue was diluted with toluene, and 1 M NaOH solution was added to the solution. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (5:1, v/v) as eluent to give **54** (19.2 mg, 20%) as a yellow solid.

Mp 105-106 ^{o}C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.43-7.32 (5H, m), 7.31-7.19 (4H, m), 6.87 (1H, d, J = 1.7 Hz), 6.76 (1H, t, J = 7.3 Hz), 6.71 (2H, d, J = 8.0 Hz), 4.07 (2H, t, J = 6.2 Hz), 3.81 (2H, t, J = 6.2 Hz), 2.87 (3H, s) ; ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 154.7, 148.0, 142.1, 140.3, 137.7, 131.7, 129.6, 128.8, 127.5, 127.1, 121.4, 117.0, 112.0, 110.1, 107.1, 50.3, 39.7, 39.2; IR (ATR) 1772, 1763, 1541, 1506, 754 cm^{-1}; HRMS (ESI) m/z calcd for C$_{22}$H$_{21}$N$_2$O$_2$ [M+H]$^{+}$ 345.1598; found 345.1597; Anal. Calcd for C$_{22}$H$_{20}$N$_2$O$_2$: C, 76.72; H, 5.85; N, 8.13. Found: C, 76.56; H, 5.89; N, 8.15.

2-Nitrobiphenyl-3-ol (56), 4-nitrobiphenyl-3-ol (57) and 6-nitrobiphenyl-3-ol (58)

To a solution of 3-phenylphenol (6.00 g, 35.3 mmol) in AcOH (35 mL) was added dropwise 6 N HNO$_3$ solution (6.00 mL, 36.0 mmol) with cooling in an ice bath, and stirred at room temperature for 30 min. Water was then added, and the mixture was extracted with CH$_2$Cl$_2$. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (10:1 to 1:1, v/v) as eluent to give **57** (1.96 g, 26%) as a yellow solid and a mixture of **56** and **58** as a brown oil. A mixture of **56** and **58** was purified by silica gel column chromatography using CHCl$_3$/MeOH (50:1, v/v) as eluent to give **56** (1.08 g, 14%) as a brown solid and **58** (3.11 g, 41%) as a brown oil.

56: mp 81-83 ^{o}C [lit.[41a]: 85-86 ^{o}C (benzene)]; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 9.54 (1H, s), 7.49 (1H, t, J = 7.9 Hz), 7.42-7.40 (3H, m), 7.27-7.25 (2H, m), 7.15 (1H, dd, J = 7.4, 1.3 Hz), 6.90 (1H, dd, J = 7.4, 1.3 Hz); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 153.8, 139.4, 138.5, 134.7, 134.6, 128.6, 127.9, 127.5, 123.9, 118.5; IR (ATR) 1593, 1525, 1350, 1296, 1211 cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_8$NO$_3$ [M-H]$^{-}$ 214.0510; found 214.0505.

57: mp 103-105 ^{o}C (EtOH) [lit.[41a]: 104-105 ^{o}C (EtOH)]; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 10.71 (1H, s), 8.17 (1H, d, J = 8.8 Hz), 7.63-7.62 (2H, m), 7.53-7.42 (3H, m), 7.38-7.35 (1H, m), 7.22 (1H, dd, J = 8.8, 1.7 Hz) ; ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 155.4, 150.6, 138.2, 132.6, 129.3, 129.1, 127.3, 125.6, 119.2, 117.7; IR (ATR) 2362, 616, 1574, 1277, 1169 cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_8$NO$_3$ [M-H]$^{-}$ 214.0510; found 214.0505.

58: ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.92 (1H, d, J = 8.8 Hz), 7.40-7.39 (3H, m), 7.28-7.26 (2H, m), 6.86-6.83 (1H, m), 6.79 (1H, d, J = 2.7 Hz), 6.02 (1H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 159.1, 142.0, 139.8, 137.8, 128.5, 128.1, 127.6, 127.3, 118.6, 114.6; IR (ATR) 3365, 1574, 1508, 1308, 1201cm^{-1}; HRMS (ESI) m/z calcd for C$_{12}$H$_8$NO$_3$ [M-H]$^{-}$ 214.0510; found 214.0505.

2-Aminobiphenyl-3-ol (59)

To a solution of **56** (861 mg, 4.00 mmol) in MeOH (20 mL) was added 10% Pd/C (50% wet, 200 mg), and stirred at room temperature for 6 h under hydrogen atmosphere. The reaction mixture was filtered through Celite, and the filtrate was concentrated to give **59** (707 mg, 95%) as a yellow solid.

Mp 119-120 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 9.52 (1H, s), 7.46-7.42 (4H, m), 7.35-7.31 (1H, m), 6.77-6.73 (1H, m), 6.63-6.55 (2H, m), 4.95 (2H, br s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 145.1, 139.4, 131.3, 128.6, 128.6, 127.6, 126.8, 120.8, 118.0, 113.5; IR (ATR) 2922, 1558, 1471, 1296, 1186 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{12}NO$ [M+H]$^+$ 186.0913; found 186.0908.

4-Aminobiphenyl-3-ol (60)

Compound **60** was prepared from **57** (1.45 g, 6.74 mmol) in a manner similar to that described for compound **59** as a brown solid (1.22, 98%).

Mp 179-181 °C (EtOH) [lit.[43]: 182-184 °C (EtOH)]; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 9.14 (1H, s), 7.47 (2H, d, J = 7.3 Hz), 7.36 (2H, dd, J = 7.3, 7.3 Hz), 7.20 (1H, t, J = 7.3 Hz), 6.96 (1H, d, J = 2.0 Hz), 6.89 (1H, dd, J = 8.0, 2.0 Hz), 6.66 (1H, d, J = 8.0 Hz), 4.67 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 144.2, 140.9, 136.5, 128.7, 128.4, 125.7, 125.4, 117.9, 114.6, 112.5; IR (ATR) 3356, 3282, 1601, 1489, 1431 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{12}NO$ [M+H]$^+$ 186.0913; found 186.0909.

4-Phenyl-1,3-benzoxazol-2(3*H*)-one (61)

Compound **61** was prepared from **59** (604 mg, 3.26 mmol) in a manner similar to that described for compound **45** as a brown solid (412 mg, 60%).

Mp 198-199 °C (CHCl$_3$); ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 11.72 (1H, s), 7.57-7.56 (2H, m), 7.50 (2H, dd, J = 7.6, 7.6 Hz), 7.42 (1H, t, J = 7.6 Hz), 7.29 (1H, dd, J = 7.6, 1.5 Hz), 7.24-7.16 (2H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 154.7, 143.7, 136.1, 128.9, 128.1, 127.9, 127.8, 124.1, 123.8, 122.2, 108.5; IR (ATR) 3178, 1765, 1429, 1259, 1157 cm^{-1}; HRMS (ESI) m/z calcd for $C_{13}H_8NO_2$ [M-H]$^-$ 210.0561; found 210.0555.

6-Phenyl-1,3-benzoxazol-2(3*H*)-one (62)

Compound **62** was prepared from **60** (1.02 g, 5.51 mmol) in a manner similar to that described for compound **45** as a brown solid (0.969 g, 83%).

Mp 250-251 °C (CHCl$_3$); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 11.72 (1H, s), 7.66-7.61 (3H, m), 7.47-7.43 (3H, m), 7.35 (1H, t, J = 7.3 Hz), 7.17 (1H, d, J = 8.0 Hz); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 154.5, 144.1, 139.7, 134.5, 129.8, 128.9, 127.2, 126.6, 122.3, 109.9, 107.9; IR (ATR) 3213, 1772, 1724, 1477, 1259 cm^{-1}; HRMS (ESI) m/z calcd for $C_{13}H_8NO_2$ [M-H]$^-$ 210.0561; found 210.0556.

tert-Butyl (2-oxo-4-phenyl-1,3-benzoxazol-3(2_H_)-yl)acetate (63)

To a suspension of **61** (250 mg, 1.18 mmol) and K_2CO_3 (245 mg, 1.78 mmol) in DMF (5.0 mL) was added _tert_-butyl bromoacetate (0.192 mL, 1.30 mmol) with cooling in an ice bath. The reaction mixture was stirred at 60 °C for 2 h and cooled to room temperature. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was recrystallized from hexane/EtOAc to give **63** (269 mg, 70%) as a white solid.

Mp 159-160 °C; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.47-7.40 (3H, m), 7.35-7.30 (2H, m), 7.28-7.21 (1H, m), 7.14 (1H, t, J = 7.9 Hz), 7.02 (1H, d, J = 7.9 Hz), 4.10 (2H, s), 1.32 (9H, s); ^{13}C-NMR (100 MHz, $CDCl_3$) δ: 165.9, 155.3, 142.9, 136.4, 129.4, 128.4, 128.2, 127.8, 126.3, 125.6, 122.1, 109.2, 82.6, 45.1, 27.8; IR (ATR) 1770, 1734, 1458, 1238, 1153 cm^{-1}; HRMS (ESI) m/z calcd for $C_{19}H_{18}NO_4$ [M-H]$^-$ 324.1236; found 324.1233.

tert-Butyl (2-oxo-6-phenyl-1,3-benzoxazol-3(2_H_)-yl)acetate (64)

Compound **64** was prepared from **61** (700 mg, 3.31 mmol) in a manner similar to that described for compound **63** as a yellow solid (937 mg, 87%).

Mp 181-183 °C (EtOAc-hexane); ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.54 (2H, d, J = 7.8 Hz), 7.48-7.32 (5H, m), 6.94 (1H, d, J = 8.0 Hz), 4.49 (2H, s), 1.48 (9H, s); ^{13}C-NMR (100 MHz, $CDCl_3$) δ: 165.6, 154.6, 143.1, 140.3, 136.9, 130.0, 128.9, 127.4, 127.1, 122.9, 109.0, 108.5, 83.5, 43.9, 28.0; IR (ATR) 1770, 1732, 1485, 1356, 1236 cm^{-1}; HRMS (ESI) m/z calcd for $C_{19}H_{18}NO_4$ [M-H]$^-$ 324.1236; found 324.1233.

(2-Oxo-4-phenyl-1,3-benzoxazol-3(2_H_)-yl)acetic acid (65)

Compound **65** was prepared from **63** (1.24 g, 3.82 mmol) according to the general procedure B as a pale brown solid (1.01 g, 98%).

Mp 218-219 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 12.99 (1H, s), 7.46-7.45 (4H, m), 7.36-7.34 (2H, m), 7.23 (1H, t, J = 7.9 Hz), 7.06 (1H, d, J = 7.9 Hz), 4.07 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.4, 154.4, 142.2, 135.7, 129.2, 128.4, 128.3, 127.5, 126.2, 125.3, 122.3, 109.1, 44.4; IR (ATR) 3167, 1747, 1732, 1454, 1190 cm^{-1}; HRMS (ESI) m/z calcd for $C_{15}H_{10}NO_4$ [M-H]$^-$ 268.0615; found 268.0610.

(2-Oxo-6-phenyl-1,3-benzoxazol-3(2_H_)-yl)acetic acid (66)

Compound **66** was prepared from **64** (1.65 g, 5.07 mmol) according to the general procedure B as a white solid (1.34 g, 98%).

Mp 224-226 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 13.42 (1H, s), 7.71-7.67 (3H, m), 7.54 (1H, dd, J = 8.2, 1.6 Hz), 7.46 (2H, dd, J = 7.7, 7.7 Hz), 7.41-7.34 (2H, m), 4.70 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6)

δ: 168.7, 154.0, 142.6, 139.6, 135.2, 130.5, 128.9, 127.3, 126.7, 122.6, 109.8, 108.2, 43.0; IR (ATR) 1770, 1732, 1485, 1358, 1236 cm^{-1}; HRMS (ESI) m/z calcd for $C_{15}H_{10}NO_4$ [M-H]$^-$ 268.0615; found 268.0610.

N-Methyl-2-(2-oxo-4-phenyl-1,3-benzoxazol-3(2H)-yl)-N-phenylacetamide (67)

Compound **67** was prepared from **65** (108mg, 0.400 mmol) and N-methylaniline (52.0 μL, 0.480 mmol) according to the general procedure C as a white solid (92.7 mg, 65%).

Mp 182-184 ^{o}C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.54 (1H, t, J = 7.1 Hz), 7.47 (2H, dd, J = 7.1, 7.1 Hz), 7.38-7.28 (5H, m), 7.22 (1H, d, J = 8.0 Hz), 7.10 (1H, dd, J = 8.0, 8.0 Hz), 6.95 (1H, d, J = 8.0 Hz), 6.74-6.67 (2H, m), 3.95 (2H, s), 3.10 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 155.6, 143.0, 141.4, 137.1, 129.9, 129.7, 128.4, 128.3, 128.2, 128.0, 127.3, 126.3, 125.3, 121.9, 109.2, 45.6, 37.4; IR (ATR) 1772, 1676, 1458, 1363, 1255 cm^{-1}; HRMS (ESI) m/z calcd for $C_{22}H_{19}N_2O_3$ [M+H]$^+$ 359.1390; found 359.1382; Anal. Calcd for $C_{22}H_{18}N_2O_3$: C, 73.73; H, 5.06; N, 7.82. Found: C, 73.33; H, 5.00; N, 7.84.

N-Methyl-2-(2-oxo-6-phenyl-1,3-benzoxazol-3(2H)-yl)-N-phenylacetamide (68)

Compound **68** was prepared from **66** (108mg, 0.400 mmol) and N-methylaniline (52.0 μL, 0.480 mmol) according to the general procedure C as a white solid (111 mg, 77%).

Mp 201-203 ^{o}C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.56-7.49 (4H, m), 7.48-7.40 (4H, m), 7.40-7.32 (4H, m), 6.94 (1H, d, J = 8.0 Hz), 4.35 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.7, 144.6, 143.2, 142.0, 140.5, 136.7, 130.5, 130.5, 128.9, 127.3, 127.2, 127.1, 122.9, 109.0, 108.7, 43.9, 37.8; IR (ATR) 1774, 1672, 1485, 1387, 1367 cm^{-1}; HRMS (ESI) m/z calcd for $C_{22}H_{19}N_2O_3$ [M+H]$^+$ 359.1390; found 359.1382; Anal. Calcd for $C_{22}H_{18}N_2O_3$·0.50H$_2$O: C, 71.92; H, 5.21; N, 7.62. Found: C, 72.15; H, 4.94; N, 7.69.

2-Amino-6-bromophenol (70)

To a solution of reduced iron (7.17 g, 128 mmol) in AcOH (30 mL) was added dropwise a solution of 2-bromo-6-nitrophenol (4.00 g, 18.3 mmol) in AcOH (20 mL) at 90 ^{o}C. The reaction mixture was stirred at 90 ^{o}C for 30 min and cooled to room temperature. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was diluted with EtOAc and aqueous saturated NaHCO$_3$. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (5:1, v/v) as eluent to give **70** (2.84 g, 83%) as a brown solid.

Mp 84-86 ^{o}C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 6.87-6.83 (1H, m), 6.65-6.63 (2H, m), 5.41 (1H, br s), 3.85 (2H, br s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 140.0, 135.5, 121.8, 121.0, 114.9, 110.0; IR (ATR) 3032, 1578, 1473, 1456, 1227 cm^{-1}; HRMS (ESI) m/z calcd for C_6H_7BrNO [M+H]$^+$ 187.9706; found 187.9704.

7-Bromo-1,3-benzoxazol-2(3*H*)-one (71)

Compound **71** was prepared from **70** (2.74 g, 14.6 mmol) in a manner similar to that described for compound **45** as an orange solid (3.04 g, 97%).

Mp 244-245 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 11.96 (1H, s), 7.28 (1H, dd, J = 7.0, 2.8 Hz), 7.11-7.09 (2H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 153.3, 141.2, 131.4, 125.2, 124.5, 109.2, 100.8; IR (ATR) 3101, 1716, 1616, 1448, 1398 cm^{-1}; HRMS (ESI) m/z calcd for C$_7$H$_3$BrNO$_2$ [M-H]$^-$ 211.9353; found 211.9350.

tert-Butyl (7-bromo-2-oxo-1,3-benzoxazol-3(2*H*)-yl)acetate (72)

Compound **72** was prepared from **71** (1.00 g, 4.67 mmol) in a manner similar to that described for compound **63** as a white solid (1.38 g, 90%).

Mp 99-100 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.27 (1H, d, J = 8.3 Hz), 7.07 (1H, dd, J = 8.3, 8.3 Hz), 6.82 (1H, d, J = 8.3 Hz), 4.45 (2H, s), 1.47 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.3, 153.4, 140.7, 131.6, 126.1, 125.0, 107.3, 102.7, 83.7, 44.1, 27.9; IR (ATR) 1772, 1718, 1616, 1468, 1369 cm^{-1}; HRMS (ESI) m/z calcd for C$_{13}$H$_{13}$BrNO$_4$ [M-H]$^-$ 326.0033; found 326.0032.

(7-Bromo-2-oxo-1,3-benzoxazol-3(2*H*)-yl)acetic acid (73)

Compound **73** was prepared from **72** (1.32 g, 4.02 mmol) according to the general procedure B as a white solid (1.07 g, 98%).

Mp 235-237 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 13.41 (1H, s), 7.39-7.33 (2H, m), 7.19 (1H, t, J = 8.0 Hz), 4.67 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.5, 153.0, 139.8, 132.1, 125.5, 125.2, 109.0, 101.1, 43.3; IR (ATR) 1770, 1730, 1616, 1467, 1252 cm^{-1}; HRMS (ESI) m/z calcd for C$_9$H$_5$BrNO$_4$ [M-H]$^-$ 269.9407; found 269.9402.

2-(7-Bromo-2-oxo-1,3-benzoxazol-3(2*H*)-yl)-*N*-methyl-*N*-phenylacetamide (74)

Compound **74** was prepared from **73** (20.0 mg, 73.5 μmol) and *N*-methylaniline (7.96 μL, 73.5 μmol) according to the general procedure C as a white solid (9.40 mg, 35%).

Mp 170-172 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.52 (2H, dd, J = 7.6, 7.6 Hz), 7.45 (1H, t, J = 7.6 Hz), 7.32 (2H, d, J = 7.6 Hz), 7.26-7.23 (1H, m), 7.04 (1H, dd, J = 8.0, 8.0 Hz), 6.82 (1H, d, J = 8.0 Hz), 4.31 (2H, s), 3.32 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.8, 153.6, 141.8, 140.7, 132.0, 130.5, 129.0, 127.2, 125.8, 124.9, 107.6, 102.6, 44.1, 37.8; IR (ATR) 1782, 1670, 1466, 1254, 1016 cm^{-1}; HRMS (ESI) m/z calcd for C$_{16}$H$_{14}$BrN$_2$O$_3$ [M+H]$^+$ 361.0182; found 361.0182.

N-Methyl-2-(2-oxo-7-phenyl-1,3-benzoxazol-3(2H)-yl)-N-phenylacetamide (75)

Compound **75** was prepared from **74** (50 mg, 138 μmol) and phenylboronic acid (20.3 mg, 166 μmol) according to the general procedure A as a brown solid (45.3 mg, 92%).

Mp 126-128 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.77-7.72 (2H, m), 7.55-7.42 (5H, m), 7.41-7.28 (4H, m), 7.23 (1H, t, J = 7.8 Hz), 6.84 (1H, d, J = 6.6 Hz), 4.36 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.6, 142.0, 140.0, 134.6, 131.7, 130.5, 128.9, 128.7, 128.4, 128.2, 127.2, 124.4, 124.2, 122.4, 107.4, 44.0, 37.8; IR (ATR) 1774, 1664, 1470, 1435, 748 cm^{-1}; HRMS (ESI) m/z calcd for C$_{22}$H$_{19}$N$_2$O$_3$ [M+H]$^+$ 359.1390; found 359.1385; Anal. Calcd for C$_{22}$H$_{18}$N$_2$O$_3$: C, 73.73; H, 5.06; N, 7.82. Found: C, 73.38; H, 5.08; N, 7.84.

2-Amino-4-bromophenol (77)

To a solution of 4-bromo-2-nitrophenol (50.7 g, 233 mmol) in THF (500 mL) was added 5% Rh/C (5.00 g), and stirred at room temperature for 11 h under hydrogen atmosphere. The reaction mixture was filtered through Celite, and the filtrate was concentrated to give **77** (43.3 g, 99%) as a brown solid.

Mp 133-135 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 9.29 (1H, s), 6.72 (1H, d, J = 2.4 Hz), 6.56 (1H, d, J = 8.3 Hz), 6.50 (1H, dd, J = 8.3, 2.4 Hz), 4.91 (2H, br s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 143.3, 138.6 118.2, 116.1, 115.6, 110.6; IR (ATR) 3062, 1497, 1444, 1437, 1279 cm^{-1}; HRMS (ESI) m/z calcd for C$_6$H$_7$BrNO [M+H]$^+$ 187.9706; found 187.9704.

5-Bromo-1,3-benzoxazol-2(3H)-one (78)

Compound **78** was prepared from **77** (49.0 g, 261 mmol) in a manner similar to that described for compound **45** as a brown solid (53.2 g, 95%).

Mp 206-208 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 11.99 (1H, s), 7.27-7.26 (3H, m); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 154.0, 142.5, 132.0, 124.3, 115.3, 112.4, 111.3; IR (ATR) 2359, 1751, 1622, 1473, 1254cm^{-1}; HRMS (ESI) m/z calcd for C$_7$H$_3$BrNO$_2$ [M-H]$^-$ 211.9353; found 211.9350.

tert-Butyl (5-bromo-2-oxo-1,3-benzoxazol-3(2H)-yl)acetate (79)

Compound **79** was prepared from **78** (53.0 g, 248 mmol) in a manner similar to that described for compound **63** as a beige solid (75.2 g, 92%).

Mp 144-145 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.26 (1H, dd, J=8.5, 1.9 Hz), 7.10 (1H, d, J = 8.5 Hz), 7.03 (1H, d, J = 1.9 Hz), 4.43 (2H, s), 1.48 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.3, 154.0, 141.6, 132.1, 125.6, 116.5, 111.7, 111.5, 83.8, 43.9, 28.0; IR (ATR) 1781, 1736, 1608, 1485, 1387 cm^{-1}; HRMS (ESI) m/z calcd for C$_{13}$H$_{15}$BrNO$_4$ [M+H]$^+$ 328.0179; found 328.0181.

(5-Bromo-2-oxo-1,3-benzoxazol-3(2*H*)-yl)acetic acid (80)

Compound **80** was prepared from **79** (170 mg, 0.518 mmol) according to the general procedure B as a white solid (132 mg, 94%).

Mp 204-206 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 13.42 (1H, br s), 7.68 (1H, d, *J* = 1.7 Hz), 7.37-7.33 (2H, m), 4.66 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.6, 153.6, 141.1, 132.7, 125.0, 115.8, 112.6, 111.6, 43.1; IR (ATR) 2953, 1736, 2701, 1483, 1227 cm^{-1}; HRMS (ESI) m/z calcd for $C_9H_5BrNO_4$ [M-H]$^-$ 269.9407; found 269.9406.

2-(5-Bromo-2-oxo-1,3-benzoxazol-3(2*H*)-yl)-*N*-methyl-*N*-phenylacetamide (81)

Compound **81** was prepared from **80** (10.0 g, 36.8 mmol) and *N*-methylaniline (4.78 mL, 44.1 mmol) according to the general procedure C as a white solid (7.16 g, 54%).

Mp 122-124 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.52 (2H, dd, *J* = 7.6, 7.6 Hz), 7.45 (1H, t, *J* = 7.6 Hz), 7.34 (2H, d, *J* = 7.6 Hz), 7.23 (1H, dd, *J* = 8.3, 1.7 Hz), 7.06 (1H, d, *J* = 8.3 Hz), 7.01 (1H, d, *J* = 1.7 Hz), 4.28 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.2, 154.6, 142.7, 142.0, 131.2, 130.5, 128.9, 127.2, 123.8, 122.6, 110.1, 108.6, 43.9, 37.8; IR (ATR) 1772, 1666, 1483, 1377, 1244 cm^{-1}; HRMS (ESI) m/z calcd for $C_{16}H_{14}BrN_2O_3$ [M+H]$^+$ 361.0182; found 361.0177; Anal. Calcd for $C_{16}H_{13}BrN_2O_3$: C, 53.21; H, 3.63; N, 7.76; Br, 22.12. Found: C, 53.15; H, 3.68; N, 7.84; Br, 21.90.

2-[5-(3-Methoxyphenyl)-2-oxo-1,3-benzoxazol-3(2*H*)-yl]-*N*-methyl-*N*-phenylacetamide (82)

Compound **68** was prepared from **67** (90.3 mg, 0.250 mmol) and 3-methoxyphenylboronic acid (49.4 mg, 0.325 mmol) according to the general procedure A as a yellow solid (51.9 mg, 53%).

Mp 182-184 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.52 (2H, dd, *J* = 7.6, 7.6 Hz), 7.44 (1H, t, *J* = 7.6 Hz), 7.39-7.32 (3H, m), 7.29 (1H, dd, *J* = 8.2, 1.6 Hz), 7.23 (1H, d, *J* = 8.2 Hz), 7.12 (1H, d, *J* = 7.6 Hz), 7.06 (1H, s), 7.02 (1H, s), 6.91 (1H, dd, *J* = 8.4, 2.8 Hz), 4.35 (2H, s), 3.88 (3H, s), 3.32 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 159.9, 154.8, 142.3, 142.2, 142.0, 137.7, 131.7, 130.5, 129.8, 128.9, 127.3, 121.8, 119.9, 113.5, 112.5, 110.1, 107.6, 55.4, 43.9, 37.8; IR (ATR) 1778, 1670, 1483, 1379, 1242 cm^{-1}; HRMS (ESI) m/z calcd for $C_{23}H_{21}N_2O_4$ [M+H]$^+$ 389.1496; found 389.1487; Anal. Calcd for $C_{23}H_{20}N_2O_4$: C, 71.12; H, 5.19; N, 7.21. Found: C, 70.77; H, 5.17; N, 7.25.

2-[5-(4-Methoxyphenyl)-2-oxo-1,3-benzoxazol-3(2*H*)-yl]-*N*-methyl-*N*-phenylacetamide (83)

Compound **83** was prepared from **81** (90.3 mg, 0.250 mmol) and 4-methoxyphenylboronic acid (49.4 mg, 0.325 mmol) according to the general procedure A as a white solid (77.7 mg, 80%).

Mp 168-170 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.51 (2H, dd, *J* = 7.8, 7.8 Hz), 7.48-7.43 (3H, m), 7.34 (2H, d, *J* = 8.0 Hz), 7.27-7.19 (2H, m), 7.00-6.95 (3H, m), 4.35 (2H, s), 3.86 (3H, s), 3.32 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.2, 159.2, 154.8, 142.0, 141.8, 137.6, 133.4, 131.7, 130.5, 128.9, 128.4,

127.2, 121.4, 114.2, 110.1, 107.2, 55.4, 43.9, 37.8; IR (ATR) 1772, 1670, 1489, 1387, 1248 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{21}$N$_2$O$_4$ [M+H]$^+$ 389.1496; found 389.1487; Anal. Calcd for C$_{23}$H$_{20}$N$_2$O$_4$: C, 71.12; H, 5.19; N, 7.21. Found: C, 70.82; H, 5.32; N, 7.13.

N-Methyl-2-{2-oxo-5-[3-(trifluoromethyl)phenyl]-1,3-benzoxazol-3(2*H*)-yl}-*N*-phenylacetamide (84)

Compound **84** was prepared from **81** (90.3 mg, 0.250 mmol) and 3-(trifluoromethyl)phenylboronic acid (61.7 mg, 0.325 mmol) according to the general procedure A as a yellow solid (77.7 mg, 73%).

Mp 193-195 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.77 (1H, s), 7.71 (1H, d, *J* = 7.6 Hz), 7.62 (1H, d, *J* = 7.6 Hz), 7.59-7.49 (3H, m), 7.45 (1H, t, *J* = 7.3 Hz), 7.36 (2H, d, *J* = 7.3 Hz), 7.33-7.27 (2H, m), 7.04 (1H, s), 4.37 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.6, 142.7, 142.0, 141.6, 136.4, 132.0, 131.4 (m), 131.1 (m), 130.7, 130.5, 129.3, 129.0, 127.3, 125.5 (m), 124.1 (m), 121.9, 110.4, 107.6, 43.9, 37.8; IR (ATR) 1788, 1651, 1489, 1381, 1329 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{18}$F$_3$N$_2$O$_3$ [M+H]$^+$ 427.1264; found 427.1252; Anal. Calcd for C$_{23}$H$_{17}$F$_3$N$_2$O$_3$·0.50H$_2$O: C, 63.45; H, 4.17; N, 6.43; F, 13.09. Found: C, 63.47; H, 4.04; N, 6.29; F, 13.21.

N-Methyl-2-{2-oxo-5-[4-(trifluoromethyl)phenyl]-1,3-benzoxazol-3(2*H*)-yl}-*N*-phenylacetamide (85a)

Compound **85** was prepared from **81** (500 mg, 1.38 mmol) and 4-(trifluoromethyl)phenylboronic acid (316 mg, 1.66 mmol) according to the general procedure A as a white solid (342 mg, 58%).

Mp 218-220 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.70 (2H, d, *J* = 8.3 Hz), 7.64 (2H, d, *J* = 8.3 Hz), 7.53 (2H, dd, *J* = 7.4, 7.4 Hz), 7.45 (1H, t, *J* = 7.4 Hz), 7.35 (2H, d, *J* = 7.4 Hz), 7.33-7.27 (2H, m), 7.05 (1H, d, *J* = 1.5 Hz), 4.37 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.6, 144.3, 142.8, 141.9, 136.4, 132.0, 130.5, 129.3 (m), 129.0, 127.6, 127.2, 125.8 (m), 125.7 (m), 122.0, 110.4, 107.7, 43.9, 37.8; IR (ATR) 1784, 1772, 1684, 1676, 1489 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{18}$F$_3$N$_2$O$_3$ [M+H]$^+$ 427.1264; found 427.1251; Anal. Calcd for C$_{23}$H$_{17}$F$_3$N$_2$O$_3$·0.75H$_2$O: C, 62.80; H, 4.24; N, 6.37; F, 12.96. Found: C, 62.47; H, 4.02; N, 6.35; F, 12.60.

N-Methyl-2-{2-oxo-5-[4-(trifluoromethoxy)phenyl]-1,3-benzoxazol-3(2*H*)-yl}-*N*-phenylacetamide (86a)

Compound **86** was prepared from **81** (50.0 mg, 0.138 mmol) and 4-(trifluoromethoxy)phenylboronic acid (34.2 mg, 0.166 mmol) according to the general procedure A as an orange solid (40.0 mg, 66%).

Mp 164-166 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.56-7.49 (4H, m), 7.45 (1H, t, *J* = 7.3 Hz), 7.35 (2H, d, *J* = 7.6 Hz), 7.29 (2H, d, *J* = 8.5 Hz), 7.26-7.24 (2H, m), 7.01 (1H, s), 4.35 (2H, s), 3.32 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.6, 148.7, 142.4, 141.9, 139.6, 136.5, 131.9, 130.5, 128.9, 128.7, 127.2, 121.8, 121.3, 119.2, 110.3, 107.5, 43.9, 37.8; IR (ATR) 1786, 1774, 1662, 1489, 1385 cm^{-1}; HRMS

(ESI) m/z calcd for $C_{23}H_{18}F_3N_2O_4$ [M+H]$^+$ 443.1213; found 443.1199; Anal. Calcd for $C_{23}H_{17}F_3N_2O_4$: C, 62.44; H, 3.87; N, 6.33; F, 12.88. Found: C, 62.32; H, 3.93; N, 6.45; F, 12.84.

N-Methyl-2-[2-oxo-5-(pyridin-2-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-phenylacetamide (87)

To a solution of **81** (181 mg, 500 μmol) and 2-(tributylstannyl)pyridine (0.192 mL, 600 μmol) in toluene (3.0 mL) was added Pd(PPh$_3$)$_4$ (28.9 mg, 25.0 μmol) in room temperature. The reaction mixture was stirred at reflux for 7 h and cooled to room temperature. Water was then added, and the mixture was extracted with EtOAc. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (1:3, v/v) as eluent to give **87** (131 mg, 73%) as a beige solid.

Mp 166-168 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.68 (1H, d, *J* = 4.9 Hz), 7.79-7.73 (1H, m), 7.70 (1H, d, *J* = 7.8 Hz), 7.68-7.62 (2H, m), 7.53 (2H, dd, *J* = 7.3, 7.3 Hz), 7.45 (1H, t, *J* = 7.3 Hz), 7.37 (2H, d, *J* = 7.3 Hz), 7.29-7.23 (2H, m), 4.39 (2H, s), 3.31 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 156.7, 154.7, 149.5, 143.4, 142.0, 136.9, 135.9, 132.0, 130.4, 128.9, 127.3, 122.2, 121.4, 120.6, 110.0, 107.4, 43.9, 37.8; IR (ATR) 1786, 1660, 1587, 1471, 1464 cm^{-1}; HRMS (ESI) m/z calcd for $C_{21}H_{18}N_3O_3$ [M+H]$^+$ 360.1343; found 360.1341; Anal. Calcd for $C_{21}H_{17}N_3O_3$: C, 70.18; H, 4.77; N, 11.69. Found: C, 70.05; H, 4.83; N, 11.67.

N-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-phenylacetamide (88a)

Compound **88a** was prepared from **81** (1.81 g, 5.00 mmol) and 3-pyridineboronic acid (0.738 g, 6.00 mmol) according to the general procedure A as a pale brown solid (1.44 g, 80%).

Mp 163-165 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.80 (1H, d, *J* = 2.0 Hz), 8.61 (1H, dd, *J* = 5.0, 2.0 Hz), 7.86-7.82 (1H, m), 7.53 (2H, dd, *J* = 7.6, 7.6 Hz), 7.45 (1H, t, *J* = 7.6 Hz), 7.40-7.33 (3H, m), 7.30-7.26 (2H, m), 7.04 (1H, s), 4.37 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.0, 154.6, 148.7, 148.4, 142.7, 141.9, 136.3, 134.6, 134.3, 132.1, 130.5, 129.0, 127.3, 123.6, 121.9, 110.5, 107.5, 43.9, 37.8; IR (ATR) 1780, 1657, 1483, 1425, 1385 cm^{-1}; HRMS (ESI) m/z calcd for $C_{21}H_{18}N_3O_3$ [M+H]$^+$ 360.1343; found 360.1341; Anal. Calcd for $C_{21}H_{17}N_3O_3 \cdot 0.25H_2O$: C, 69.32; H, 4.85; N, 11.55. Found: C, 68.97; H, 4.75; N, 11.18.

N-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-phenylacetamide hydrochloride (88a·HCl)

To a solution of **88a** (1.20 g, 3.34 mmol) in THF (100 mL) was added 4 N HCl in 1,4-dioxane (2.50 mL) at room temperature. The reaction mixture was stirred at room temperature for 0.25 h. The resulting solid was filtered, and washed by hexane to give hydrochloride of **88a** (1.17 g, 88%) as a white solid.

Mp 268-270 $^{\circ}$C (*i*PrOH); ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 9.21 (1H, s), 8.86 (1H, d, J = 5.4 Hz), 8.74 (1H, d, J = 8.0 Hz), 8.07-8.02 (1H, m), 7.85 (1H, s), 7.85 (1H, s), 7.65 (1H, d, J = 8.3 Hz), 7.63-7.53 (4H, m), 7.51-7.44 (1H, m), 4.46 (2H, s), 3.22 (3H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 164.8, 153.9, 142.8, 142.1, 141.8, 141.5, 141.1, 137.6, 132.6, 130.5, 130.1, 128.4, 127.4, 126.5, 121.7, 110.4, 108.6, 43.7, 37.2; IR (ATR) 2322, 1770, 1659, 1599, 1498 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{21}H_{18}N_3O_3$ [M+H]$^+$ 360.1343; Found 360.1337; Anal. Calcd for $C_{21}H_{17}N_3O_3 \cdot HCl \cdot 0.50H_2O$: C, 62.30; H, 4.73; N, 10.38; Cl, 8.76. Found: C, 62.35; H, 4.59; N, 10.51; Cl, 8.56.

N-Methyl-2-[2-oxo-5-(pyridin-4-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-phenylacetamide (89a)

Compound **89a** was prepared from **81** (2.00 g, 5.54 mmol) and 4-pyridineboronic acid (0.817 g, 6.64 mmol) in a manner similar to that described for compound **89a** as a grey solid (1.35 g, 68%).

Mp 213-214 $^{\circ}$C (MeOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 8.67 (2H, d, J = 4.9 Hz), 7.53 (2H, dd, J = 7.4, 7.4 Hz), 7.49-7.43 (3H, m), 7.39-7.34 (3H, m), 7.29 (1H, d, J = 8.3 Hz), 7.10 (1H, d, J = 1.5 Hz), 4.38 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.0, 154.5, 150.3, 147.9, 143.3, 141.9, 134.7, 132.2, 130.5, 129.0, 127.3, 121.8, 110.6, 107.4, 43.9, 37.8; IR (ATR) 1780, 1770, 1668, 1597, 1485 cm^{-1}; HRMS (ESI) m/z calcd for $C_{21}H_{18}N_3O_3$ [M+H]$^+$ 360.1343; found 360.1340; Anal. Calcd for $C_{21}H_{17}N_3O_3 \cdot 0.25H_2O$: C, 69.32; H, 4.85; N, 11.55. Found: C, 69.58; H, 4.74; N, 11.69.

N-Methyl-2-[2-oxo-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-1,3-benzoxazol-3(2*H*)-yl]-*N*-phenylacetamide (91)

To a suspension of **81** (6.18 g, 17.1 mmol) in 1,4-dioxane (90 mL) were added bis(pinacolato)diboron (4.75 g, 18.7 mmol), Pd$_2$(dba)$_3$ (0.781 g, 0.853 mmol), tricyclohexylphosphine (PCy$_3$) (1.15 g, 4.10 mmol) and KOAc (2.51 g, 25.6 mmol) in room temperature. The reaction mixture was stirred at reflux for 28 h and cooled to room temperature. Water was then added, and the mixture was extracted with EtOAc. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (2:1, v/v) as eluent to give **91** (5.81 g, 83%) as a white solid.

Mp 163-164 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.60 (1H, dd, J = 7.8, 1.0 Hz), 7.52 (2H, dd, J = 7.6, 7.6 Hz), 7.44 (1H, dd, J = 7.6, 7.6 Hz), 7.37 (2H, d, J = 7.6 Hz), 7.28 (1H, br s), 7.19 (1H, d, J = 7.8 Hz), 4.32 (2H, s), 3.32 (3H, s), 1.36 (12H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.5, 145.1, 141.9, 131.0, 130.4, 129.9, 128.8, 127.3, 114.0, 109.5, 84.0, 43.7, 37.7, 24.9; IR (ATR) 1788, 1668, 1458, 1379, 1342 cm^{-1}; HRMS (ESI) m/z calcd for $C_{22}H_{24}BN_2O_5$ [M-H]$^-$ 407.1784; found 407.1775; Anal. Calcd for $C_{22}H_{25}BN_2O_5$: C, 64.72; H, 6.17; N, 6.86. Found: C, 64.57; H, 6.28; N, 6.72.

General procedure F for the Suzuki-Miyaura Coupling Reaction

To a suspension of 7 and arylhalide (1 equiv.) in 1 M K_2CO_3 solution (3 equiv.) and 1,4-dioxane (0.15 M) was added $Pd(PPh_3)_4$ (3 mol%) in room temperature. The reaction mixture was stirred at reflux for 2 h and cooled to room temperature. Water was then added, and the mixture was extracted with EtOAc. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography to afford the benzoxazolone derivative.

N-Methyl-2-[2-oxo-5-(pyridin-2-yl)-1,3-benzoxazol-3(2H)-yl]-N-phenylacetamide (87)

Compound **87** was prepared from **91** (150 mg, 367 μmol) and 2-bromopyridine (35.0 μL, 367 μmol) according to general procedure F as a beige solid (92.5 mg, 70%).

2-[5-{2-[(Dimethylamino)methyl]phenyl}-2-oxo-1,3-benzoxazol-3(2H)-yl]-N-methyl-N-phenyl acetamide (92)

Compound **92** was prepared from **91** (150 mg, 367 μmol) and 2-bromo-N,N-dimethylbenzylamine (78.7 mg, 367 μmol) according to general procedure F as a brown solid (66.0 mg, 43%).

Mp 106-107 ^{o}C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.52-7.46 (3H, m), 7.43 (1H, dd, J = 6.8, 6.8 Hz), 7.38-7.29 (4H, m), 7.28-7.24 (1H, m), 7.21 (1H, d, J = 8.0 Hz), 7.11 (1H, dd, J = 8.0, 1.7 Hz), 7.09-7.06 (1H, m), 4.32 (2H, s), 3.29 (3H, s), 3.27 (2H, s), 2.18 (6H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.0, 154.9, 142.0, 141.8, 141.8, 137.5, 136.4, 130.9, 130.4, 130.4, 130.3, 128.8, 127.4, 127.3, 126.9, 124.0, 110.1, 109.4, 61.2, 45.3, 43.8, 37.8; IR (ATR) 1782, 1697, 1684, 653, 1558 cm^{-1}; HRMS (ESI) m/z calcd for $C_{25}H_{26}N_3O_3$ [M+H]$^+$ 416.1969; found 416.1958; Anal. Calcd for $C_{25}H_{25}N_3O_3 \cdot 0.25H_2O$: C, 71.49; H, 6.12; N, 10.01. Found: C, 71.64; H, 6.18; N, 10.26.

2-[5-{3-[(Dimethylamino)methyl]phenyl}-2-oxo-1,3-benzoxazol-3(2H)-yl]-N-methyl-N-phenyl acetamide (93)

Compound **93** was prepared from **91** (150 mg, 367 μmol) and 3-bromo-N,N-dimethylbenzylamine (78.7 mg, 367 μmol) according to general procedure F as a white solid (136 mg, 89%).

Mp 146-148 ^{o}C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.59 (1H, s), 7.52 (2H, dd, J = 7.5, 7.5 Hz), 7.45 (2H, dd, J = 7.5. 7.5 Hz), 7.43-7.37 (3H, m), 7.33-7.28 (2H, m), 7.23 (1H, d, J = 8.3 Hz), 7.15 (1H, s), 4.37 (2H, s), 3.59 (2H, s), 3.32 (3H, s), 2.36 (6H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.2, 154.8, 142.2, 142.0, 141.0, 137.6, 131.8, 130.4, 128.8, 128.3, 128.2, 127.3, 126.4, 121.8, 110.0, 107.7, 64.1, 45.1, 43.9, 37.8; IR (ATR) 1778, 1662, 1595, 1487, 1383 cm^{-1}; HRMS (ESI) m/z calcd for $C_{25}H_{26}N_3O_3$ [M+H]$^+$ 416.1969; found 416.1959; Anal. Calcd for $C_{25}H_{25}N_3O_3 \cdot 0.50H_2O$: C, 70.74; H, 6.17; N, 9.90. Found: C, 70.74; H, 6.04; N, 10.11.

2-[5-{4-[(Dimethylamino)methyl]phenyl}-2-oxo-1,3-benzoxazol-3(2H)-yl]-N-methyl-N-phenyl acetamide (94)

Compound **94** was prepared from **91** (150 mg, 367 μmol) and 4-bromo-N,N-dimethylbenzylamine (78.7 mg, 367 μmol) according to general procedure F as a brown solid (107 mg, 70%).

Mp 64-66 °C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.55-7.41 (5H, m), 7.40-7.32 (4H, m), 7.32-7.26 (1H, m), 7.23 (1H, d, J = 8.3 Hz), 7.04 (1H, s), 4.35 (2H, s), 3.48 (2H, s), 3.32 (3H, s), 2.28 (6H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.8, 142.1, 142.0, 139.6, 138.1, 137.7, 131.7, 130.5, 129.6, 128.9, 127.2, 127.2, 121.7, 110.1, 107.4, 64.0, 45.4, 43.9, 37.8; IR (ATR) 1774, 1668, 1595, 1489, 1385 cm^{-1}; HRMS (ESI) m/z calcd for C$_{25}$H$_{26}$N$_3$O$_3$ [M+H]$^+$ 416.1969; found 416.1961; Anal. Calcd for C$_{25}$H$_{25}$N$_3$O$_3$·0.75H$_2$O: C, 69.99; H, 6.23; N, 9.79. Found: C, 70.27; H, 6.16; N, 9.48.

2-[5-(6-Aminopyridin-2-yl)-2-oxo-1,3-benzoxazol-3(2H)-yl]-N-methyl-N-phenylacetamide (95)

Compound **95** was prepared from **91** (100 mg, 245 μmol) and 2-amino-6-bromopyridine (42.4 mg, 245 μmol) according to general procedure F as a white solid (59.1 mg, 64%): mp 214-216 °C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.63 (1H, dd, J = 8.3, 1.7 Hz), 7.55-7.48 (4H, m), 7.44 (1H, dd, J = 7.3, 7.3 Hz), 7.36 (2H, d, J = 7.3 Hz), 7.22 (1H, d, J = 8.3 Hz), 7.05 (1H, d, J = 7.3 Hz), 6.48 (1H, d, J = 7.3 Hz), 4.55 (2H, s), 4.38 (2H, s), 3.32 (3H, s); ^{13}C-NMR (100MHz, CDCl$_3$) δ: 165.2, 158.2, 155.3, 154.8, 143.1, 142.0, 138.5, 136.2, 131.6, 130.4, 128.8, 127.3, 121.4, 110.9, 109.8, 107.3, 107.2, 43.9, 37.8; IR (ATR) 1782, 1662, 1593, 1462, 1441 cm^{-1}; HRMS (ESI) m/z calcd for C$_{21}$H$_{19}$N$_4$O$_3$ [M+H]$^+$ 375.1452; found 375.1450; Anal. Calcd for C$_{21}$H$_{18}$N$_4$O$_3$·0.25H$_2$O: C, 66.57; H, 4.92; N, 14.79. Found: C, 66.69; H, 4.74; N, 14.55.

2-[5-(6-Methoxypyridin-2-yl)-2-oxo-1,3-benzoxazol-3(2H)-yl]-N-methyl-N-phenylacetamide (96)

Compound **96** was prepared from **91** (55.0 mg, 135 μmol) and 2-bromo-6-methoxypyridine (25.3 mg, 135 μmol) according to general procedure F as a white solid (35.6 mg, 68%).

Mp 179-181 °C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.77 (1H, dd, J = 8.5, 1.7 Hz), 7.64 (1H, m), 7.58 (1H, d, J = 1.7 Hz), 7.52 (2H, dd, J = 7.6, 7.6 Hz), 7.44 (1H, dd, J = 7.6, 7.6 Hz), 7.36-7.29 (3H, m), 7.24 (1H, d, J = 8.5 Hz), 6.71 (1H, d, J = 8.5 Hz), 4.39 (2H, s), 4.05 (3H, s), 3.33 (3H, s); ^{13}C-NMR (100MHz, CDCl$_3$) δ: 165.1, 163.7, 154.8, 153.9, 143.3, 142.1, 139.3, 135.6, 131.6, 130.4, 128.9, 127.2, 121.4, 112.9, 110.0, 109.2, 107.0, 53.3, 43.9, 37.8; IR (ATR) 1778, 1668, 1597, 1576, 1497 cm^{-1}; HRMS (ESI) m/z calcd for C$_{22}$H$_{20}$N$_3$O$_4$ [M+H]$^+$ 390.1448; found 390.1445; Anal. Calcd for C$_{22}$H$_{19}$N$_3$O$_4$: C, 67.86; H, 4.92; N, 10.79. Found: C, 67.49; H, 4.83; N, 10.60.

2-[5-(5-Acetylpyridin-3-yl)-2-oxo-1,3-benzoxazol-3(2*H*)-yl]-*N*-methyl-*N*-phenylacetamide (97)

Compound **97** was prepared from **91** (100 mg, 245 μmol) and 3-acetyl-5-bromopyridine (49.0 mg, 245 μmol) according to general procedure F as a white solid (68.2 mg, 69%).

Mp 220-222 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 9.15 (1H, d, *J* = 2.0 Hz), 8.98 (1H, d, *J* = 2.0 Hz), 8.39 (1H, dd, *J* = 2.0, 2.0 Hz), 7.54 (2H, dd, *J* = 7.6, 7.6 Hz), 7.47 (1H, dd, *J* = 7.6, 7.6 Hz), 7.42-7.29 (4H, m), 7.09 (1H, s), 4.39 (2H, s), 3.33 (3H, s), 2.72 (3H, s); ^{13}C-NMR (100MHz, CDCl$_3$) δ: 196.7, 165.0, 154.5, 152.0, 148.6, 143.1, 141.8, 136.4, 133.6, 133.1, 132.3, 132.2, 130.5, 129.0, 127.3, 121.9, 110.7, 107.6, 43.9, 37.8, 27.0; IR (ATR) 1786, 1682, 1639, 1595, 1500 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{20}$N$_3$O$_4$ [M+H]$^+$ 402.1448; found 402.1446; Anal. Calcd for C$_{23}$H$_{19}$N$_3$O$_4$·0.50H$_2$O: C, 67.31; H, 4.91; N, 10.24. Found: C, 67.47; H, 4.54; N, 10.23.

N-Methyl-2-(2-oxo-5-phenoxy-1,3-benzoxazol-3(2*H*)-yl)-*N*-phenylacetamide (98)

A mixture of **81** (722 mg, 2.00 mmol), phenol (753 mg, 8.00 mmol), CuO (796 mg, 10.0 mmol) and K$_2$CO$_3$ (1.66 g, 12.0 mmol) in pyridine (10 mL) was heated at reflux for 18 h and cooled to room temperature. The reaction mixture was filtered through Celite, and the filtrate was diluted with CHCl$_3$ and 2M HCl solution. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (3:1, v/v) as eluent. The solvent was removed in vacuo, and the resulting solid was triturated with Et$_2$O to give **98** (269 mg, 36%) as a white solid.

Mp 133-135 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ 7.48 (2H, dd, *J* = 7.3, 7.3 Hz), 7.42 (1H, t, *J* = 7.3 Hz), 7.35 (2H, dd, *J* = 7.8, 7.8 Hz), 7.28 (2H, d, *J* = 7.3 Hz), 7.14-7.10 (2H, m), 6.99 (2H, d, *J* = 7.8 Hz), 6.72 (1H, dd, *J* = 8.5, 2.2 Hz), 6.58 (1H, d, *J* = 2.2 Hz), 4.25 (2H, s), 3.29 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.9, 157.6, 155.0, 153.6, 141.9, 138.5, 132.2, 130.4, 129.8, 128.9, 127.2, 123.2, 118.4, 113.0, 110.6, 100.8, 43.8, 37.7; IR (ATR) 1778, 1664, 1487, 1387, 1217 cm^{-1}; HRMS (ESI) m/z calcd for C$_{22}$H$_{19}$N$_2$O$_4$ [M+H]$^+$ 375.1339; found 375.1337; Anal. Calcd for C$_{22}$H$_{18}$N$_2$O$_4$: C, 70.58; H, 4.85; N, 7.48. Found: C, 70.32; H, 4.98; N, 7.44.

N-Methyl-2-{5-[methyl(phenyl)amino]-2-oxo-1,3-benzoxazol-3(2*H*)-yl}-*N*-phenylacetamide (99)

A mixture of **81** (181 mg, 0.500 mmol), *N*-methylaniline (81.3 μL, 0.750 mmol), Pd$_2$(dba)$_3$ (22.9 mg, 0.0250 mmol), Xantphos (43.4 mg, 0.0750 mmol) and Cs$_2$CO$_3$ (228 mg, 0.700 mmol) in toluene (4.0 mL) was heated at reflux for 7 h and cooled to room temperature. The reaction was quenched by adding aqueous saturated NaHCO$_3$, and then the mixture was extracted with EtOAc. The organic layer was washed with brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (3:1, v/v) as eluent to give **99** (60.6 mg, 31%) as a white solid.

Mp 123-125 $^{\circ}$C (*i*PrOH); ^{1}H-NMR (400MHz, CDCl$_3$) δ 7.47 (2H, dd, *J* = 7.3, 7.3 Hz), 7.41 (1H, t, *J* = 7.3 Hz), 7.30-7.22 (4H, m), 7.11 (1H, d, *J* = 8.5 Hz), 6.95-6.88 (3H, m), 6.80 (1H, dd, *J* = 8.5, 2.0 Hz), 6.58 (1H, d, *J* = 2.0 Hz), 4.23 (2H, s), 3.30 (3H, s), 3.28 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 155.1, 149.3, 145.8, 142.0, 138.4, 132.0, 130.4, 129.2, 128.8, 127.2, 120.2, 118.3, 117.0, 110.6, 103.8, 43.7, 40.8, 37.8; IR (ATR) 1768, 1655, 1495, 1489, 1392 cm^{-1}; HRMS (ESI) m/z calcd for C$_{23}$H$_{22}$N$_3$O$_3$ [M+H]$^+$ 388.1656; found 388.1652; Anal. Calcd for C$_{23}$H$_{21}$N$_3$O$_3$·0.50H$_2$O: C, 69.68; H, 5.59; N, 10.60. Found: C, 69.84; H, 5.41; N, 10.54.

N-Methyl-2-(2-oxo-1,3-benzoxazol-3(2*H*)-yl)-*N*-phenylacetamide (100)

To a solution of **81** (542 mg, 1.50 mmol) in MeOH (50 mL) was added 10% Pd/C (50% wet, 271 mg), and stirred at room temperature for 2.5 h under hydrogen atmosphere. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was purified by silica gel column chromatography using CHCl$_3$ as eluent to give **100** (407 mg, 96%) as a white solid.

Mp 141-143 $^{\circ}$C (*i*PrOH); ^{1}H-NMR (400MHz, CDCl$_3$) δ 7.51 (2H, dd, *J* = 7.6, 7.6 Hz), 7.44 (1H, t, *J* = 7.6 Hz), 7.33 (2H, d, *J* = 7.6 Hz), 7.20-7.06 (3H, m), 6.88 (1H, d, *J* = 7.1 Hz), 4.32 (2H, s), 3.32 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.2, 154.6, 142.7, 142.0, 131.2, 130.5, 128.9, 127.2, 123.8, 122.6, 110.1, 108.6, 43.9, 37.8; IR (ATR) 1767, 1670, 1489, 1369, 1240 cm^{-1}; HRMS (ESI) m/z calcd for C$_{16}$H$_{15}$N$_2$O$_3$ [M+H]$^+$ 283.1077; found 283.1071; Anal. Calcd for C$_{16}$H$_{14}$N$_2$O$_3$: C, 68.07; H, 5.00; N, 9.92. Found: C, 68.13; H, 4.99; N, 10.04.

2-(5-Cyano-2-oxo-1,3-benzoxazol-3(2*H*)-yl)-*N*-methyl-*N*-phenylacetamide (102)

Compound **102** was prepared from (5-cyano-2-oxo-1,3-benzoxazol-3(2H)-yl)acetic acid (218 mg, 1.00 mmol) and *N*-methylaniline (0.108 mL, 1.00 mmol) according to general procedure C as a white solid (209 mg, 68%).

Mp 166-167 $^{\circ}$C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.55 (2H, t, *J* = 7.6 Hz), 7.50-7.44 (2H, m), 7.38-7.34 (2H, m), 7.28 (1H, d, *J* = 8.0 Hz), 7.16 (1H, d, *J* = 1.2 Hz), 4.33 (2H, s), 3.34 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.4, 153.7, 145.4, 141.7, 132.1, 130.6, 129.1, 127.8, 127.2, 118.3, 112.2, 110.9, 107.8, 44.0, 37.8; IR (ATR) 1776, 1668, 1595, 1493, 1387 cm^{-1}; HRMS (ESI) m/z calcd for C$_{17}$H$_{14}$N$_3$O$_3$ [M+H]$^+$ 308.1030; found 308.1028; Anal. Calcd for C$_{17}$H$_{13}$N$_3$O$_3$: C, 66.44; H, 4.26; N, 13.67. Found: C, 66.50; H, 4.27; N, 13.78.

tert-Butyl {2-oxo-5-[4-(trifluoromethyl)phenyl]-1,3-benzoxazol-3(2*H*)-yl}acetate (103)

Compound **103** was prepared from **79** (7.20 g, 21.9 mmol) and 4-(trifluoromethyl)phenylboronic acid (5.00 g, 26.3 mmol) according to general procedure A as a pale yellow solid (7.36 g, 85%).

Mp 155-156 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.70 (2H, d, J = 8.3 Hz), 7.63 (2H, d, J = 8.3 Hz), 7.37-7.29 (2H, m), 7.06 (1H, d, J = 1.2 Hz), 4.52 (2H, s), 1.48 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.6, 154.5, 144.0, 142.7, 136.4, 131.5, 130.5 (m), 127.5, 125.9 (m), 125.8 (m), 122.2, 110.5, 107.3, 83.6, 43.9, 28.0; IR (ATR) 1772, 1757, 1738, 1325, 1240 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{20}$H$_{17}$F$_3$NO$_4$ [M-H]$^-$ 392.1110; Found 392.1111.

tert-Butyl {2-oxo-5-[4-(trifluoromethoxy)phenyl]-1,3-benzoxazol-3(2*H*)-yl}acetate (104)

Compound **104** was prepared from **79** (4.82   g, 14.7 mmol) and 4-(trifluoromethoxy)phenylboronic acid (3.18 g, 15.4 mmol) according to general procedure A as a pale yellow solid (4.62 g, 77%).

Mp 123-125 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 7.53 (2H, dd, J = 6.5, 2.1 Hz), 7.30-7.29 (4H, m), 7.01 (1H, s), 4.50 (2H, s), 1.48 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.6, 154.5, 148.8, 142.4, 139.3, 136.6, 131.4, 128.6, 122.0, 121.7, 121.3, 119.2, 110.4, 107.1, 83.6, 43.9; IR (ATR) 1763, 1749, 1716, 1489, 1234 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{20}$H$_{17}$F$_3$NO$_5$ [M-H]$^-$ 408.1059; Found 408.1062.

tert-Butyl [2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]acetate (105)

Compound **105** was prepared from **79** (3.28   g, 10.0 mmol) and 3-pyridineboronic acid (1.48 g, 12.0 mmol) according to general procedure A as a brown solid (3.26 g, quant.).

Mp 106-107 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.80 (1H, d, J = 1.5 Hz), 8.61 (1H, dd, J = 4.9, 1.5 Hz), 7.85-7.80 (1H, m), 7.38 (1H, dd, J = 7.4, 4.9 Hz), 7.33-7.32 (2H, m), 7.05-7.04 (1H, m), 4.52 (2H, s), 1.48 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.5, 154.4, 148.7, 148.3, 142.7, 136.1, 134.5, 131.6, 123.6, 122.1, 110.6, 107.1, 83.7, 43.9, 28.0; IR (ATR) 1761, 1740, 1477, 1369, 1240 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{18}$H$_{19}$N$_2$O$_4$ [M+H]$^+$ 327.1339; Found 327.1339.

tert-Butyl [2-oxo-5-(pyridin-4-yl)-1,3-benzoxazol-3(2*H*)-yl]acetate (106)

Compound **106** was prepared from **79** (3.28   g, 10.0 mmol) and 4-pyridineboronic acid (1.48 g, 12.0 mmol) according to general procedure A as a brown solid (3.26 g, quant.). mp 124-125 $^{\circ}$C; ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.67 (2H, d, J = 6.1 Hz), 7.47-7.44 (2H, m), 7.41 (1H, dd, J = 8.3, 1.7 Hz), 7.33 (1H, d, J = 8.3 Hz), 7.10 (1H, d, J = 1.7 Hz), 4.52 (2H, s), 1.49 (9H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.5, 154.4, 150.4, 147.7, 143.2, 134.8, 131.7, 122.0, 121.7, 110.7, 107.0, 83.7, 43.9, 28.0; IR (ATR) 1784, 1772, 1726, 1597, 1242 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{18}$H$_{19}$N$_2$O$_4$ [M+H]$^+$ 327.1339; Found 327.1335.

{2-Oxo-5-[4-(trifluoromethyl)phenyl]-1,3-benzoxazol-3(2*H*)-yl}acetic acid (107)

Compound **107** was prepared from **103** (7.20 g, 18.3 mmol) according to general procedure B as a pale yellow solid (6.01 g, 97%).

Mp 190-192 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 7.92 (2H, d, J = 8.0 Hz), 7.84-7.83 (3H, m), 7.55-7.50 (2H, m), 4.74 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.7, 154.0, 143.5, 142.2, 134.9, 132.0, 127.5, 128.0 (m), 125.8 (m), 125.7 (m), 121.4, 110.2, 108.3, 43.0; IR (ATR) 3197, 1782, 1757, 1734, 1318 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{16}H_9F_3NO_4$ [M-H][-] 336.0489; Found 336.0479.

{2-Oxo-5-[4-(trifluoromethoxy)phenyl]-1,3-benzoxazol-3(2*H*)-yl}acetic acid (108)

Compound **108** was prepared from **104** (4.62 g, 11.3 mmol) according to general procedure B as a white solid (3.84 g, 96%).

Mp 147-148 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 13.39 (1H, br s), 7.83-7.73 (3H, m), 7.50-7.44 (4H, m), 4.72 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.7, 154.0, 147.8, 141.8, 138.9, 135.1, 131.8, 128.7, 121.5, 121.1, 118.8, 110.1, 108.2, 43.0; IR (ATR) 3080, 1747, 1716, 1489, 1254 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{16}H_9F_3NO_5$ [M-H][-] 352.0438; Found 352.0428.

[2-Oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2*H*)-yl]acetic acid hydrochloride (109)

Compound **109** was prepared from **105** (3.86 g, 11.8 mmol) according to general procedure B as a beige solid (3.06 g, quant.).

Mp 267-269 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 9.24 (1H, s), 8.91-8.81 (2H, m), 8.14-8.07 (1H, m), 8.06 (1H, s), 7.72 (1H, dd, J = 8.4, 1.8 Hz), 7.59 (1H, d, J = 8.4 Hz), 4.74 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.5, 153.8, 142.9, 142.1, 141.1, 140.3, 137.9, 132.2, 130.1, 126.9, 121.9, 110.5, 108.5, 43.1; IR (ATR) 2818, 1772, 1734, 1213, 1028 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{14}H_{11}N_2O_4$ [M+H][+] 271.0713; Found 271.0710.

[2-Oxo-5-(pyridin-4-yl)-1,3-benzoxazol-3(2*H*)-yl]acetic acid hydrochloride (110)

Compound **110** was prepared from **106** (3.82 g, 11.7 mmol) according to general procedure B as a beige solid (2.71 g, 93%).

Mp 290-292 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 8.97 (2H, d, J = 6.6 Hz), 8.39 (2H, d, J = 6.6 Hz), 8.20 (1H, s), 7.90 (1H, d, J = 8.5 Hz), 7.63 (1H, d, J = 8.5 Hz), 4.77 (2H, s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 168.5, 154.2, 153.7, 144.3, 142.6, 132.4, 130.4, 123.4, 123.0, 110.7, 109.0, 43.2; IR (ATR) 3055, 1778, 1707, 1635, 1489 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{14}H_{11}N_2O_4$ [M+H][+] 271.0713; Found 271.0711.

N-Methyl-2-{2-oxo-5-[4-(trifluoromethyl)phenyl]-1,3-benzoxazol-3(2*H*)-yl}-*N*-(pyridin-3-yl) acetamide (85b)

Compound **85b** was prepared from **107** (438 mg, 1.30 mmol) and *N*-methy-3-pyridinamine (169 mg, 1.56 mmol) according to general procedure D as a white solid (251 mg, 45%).

Mp 209-210 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.72 (1H, d, *J* = 3.4 Hz), 8.67 (1H, s), 7.75 (1H, d, *J* = 8.5 Hz), 7.70 (2H, d, *J* = 8.0 Hz), 7.64 (2H, d, *J* = 8.0 Hz), 7.54-7.47 (1H, m), 7.35-7.24 (2H, m), 7.09 (1H, s), 4.36 (2H, s), 3.35 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.5, 150.2, 148.6, 144.2, 142.7, 138.6, 136.5, 134.9, 131.8, 129.8 (m), 129.5 (m), 127.7, 125.8 (m), 124.9 (m), 122.2, 110.5, 107.7, 43.9, 38.1; IR (ATR) 1784, 1678, 1327, 1111, 1068 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{17}$F$_3$N$_3$O$_3$ [M+H]$^+$ 428.1217; Found 428.1209; Anal. Calcd for C$_{22}$H$_{16}$F$_3$N$_3$O$_3$: C, 61.83; H, 3.77; N, 9.83; F, 13.34. Found: C, 61.82; H, 3.85; N, 10.05; F, 13.13.

N-Methyl-2-{2-oxo-5-[4-(trifluoromethoxy)phenyl]-1,3-benzoxazol-3(2*H*)-yl}-*N*-(pyridin-3-yl) acetamide (86b)

Compound **86b** was prepared from **108** (353 mg, 1.00 mmol) and *N*-methy-3-pyridinamine (270 mg, 2.50 mmol) according to general procedure C as a white solid (241 mg, 54%).

Mp 132-133 ^{o}C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.71 (1H, d, *J* = 4.1 Hz), 8.67 (1H, s), 7.75 (1H, d, *J* = 8.0 Hz), 7.59-7.46 (3H, m), 7.32-7.24 (4H, m), 7.04 (1H, s), 4.34 (2H, s), 3.35 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.1, 154.5, 150.1, 148.8, 148.6, 142.4, 139.4, 138.6, 136.6, 134.9, 131.6, 128.7, 124.8, 122.0, 121.3, 119.2, 110.4, 107.5, 43.9, 38.1; IR (ATR) 1782, 1763, 1666, 1489, 1248 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{17}$F$_3$N$_3$O$_4$ [M+H]$^+$ 444.1166; Found 444.1157; Anal. Calcd for C$_{22}$H$_{16}$F$_3$N$_3$O$_4$·0.25H$_2$O: C, 59.00; H, 3.71; N, 9.38; F, 12.73. Found: C, 58.64; H, 3.74; N, 9.53; F, 12.56.

N-Methyl-2-{2-oxo-5-[4-(trifluoromethoxy)phenyl]-1,3-benzoxazol-3(2*H*)-yl}-*N*-(pyridin-3-yl) acetamide hydrochloride (86b·HCl)

To a solution of **86b** (232 mg, 523 μmol) in THF (5.0 mL) was added 4 N HCl in 1,4-dioxane (392 μL) at room temperature. The reaction mixture was stirred at room temperature for 0.5 h. The solvent was removed in vacuo, and the residue was triturated with hexane. The resulting solid was filtered, and washed by hexane to give hydrochloride of **86b** (222 mg, 88%) as a white solid.

Mp 206-208 ^{o}C (MeOH); ^{1}H-NMR (DMSO-*d$_6$*) δ: 9.10-8.98 (1H, br m), 8.81-8.68 (1H, br m), 8.49-8.43 (1H, m), 7.97-7.87 (1H, br m), 7.83-7.77 (2H, m), 7.76-7.67 (1H, br m), 7.51-7.44 (4H, m), 5.35-4.46 (2H, br m), 3.79-3.16 (3H, br m); ^{13}C-NMR (100 MHz, DMSO-*d$_6$*) δ: 166.0, 154.1, 147.8, 145.8, 141.8, 139.5, 139.1, 135.1, 132.1, 128.8, 126.3, 121.5, 121.4, 121.2, 118.8, 116.3, 110.0, 108.5, 44.0, 36.7; IR (ATR) 2359, 1780, 1668, 1558, 1489 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{17}$F$_3$N$_3$O$_4$ [M+H]$^+$ 444.1166; Found 444.1154; Anal. Calcd for C$_{22}$H$_{16}$F$_3$N$_3$O$_4$·0.90HCl·0.50H$_2$O: C, 54.46; H, 3.72; N, 8.66; Cl, 6.58; F, 11.75. Found: C, 54.38; H, 3.72; N, 8.76; Cl, 6.72; F, 11.41.

N-Ethyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2H)-yl]-N-phenylacetamide (88b)

Compound **88b** was prepared from **109** (50.0 mg, 163 μmol) and *N*-ethylaniline (24.6 μL, 196 μmol) according to general procedure C as a white solid (17.1 mg, 28%).

Mp 134-136 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.82-8.77 (1H, m), 8.64-8.59 (1H, m), 7.87-7.80 (1H, m), 7.53 (2H, dd, *J* = 7.6, 7.6 Hz), 7.46 (1H, t, *J* = 7.6, 7.6 Hz), 7.38 (1H, dd, *J* = 7.7, 5.0 Hz), 7.35-7.23 (4H, m), 7.04 (1H, s), 4.32 (2H, s), 3.79 (2H, q, *J* = 7.2 Hz), 1.15 (3H, t, *J* = 7.2 Hz); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.5, 154.6, 148.6, 148.4, 142.7, 140.2, 136.3, 134.6, 134.3, 132.1, 130.4, 129.0, 128.3, 123.6, 121.9, 110.5, 107.6, 44.8, 44.1, 12.9; IR (ATR) 1786, 1653, 1479, 1387, 1255 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{20}$N$_3$O$_3$ [M+H]$^+$ 374.1499; Found 374.1495; Anal. Calcd for C$_{22}$H$_{19}$N$_3$O$_3$: C, 70.76; H, 5.13; N, 11.25. Found: C, 70.49; H, 5.17; N, 11.52.

N-(4-Methoxyphenyl)-N-methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2H)-yl]acetamide (88c)

Compound **88c** was prepared from **109** (50.0 mg, 163 μmol) and *N*-methyl-*p*-anisidine (26.8 mg, 196 μmol) according to general procedure C as a purple solid (51.7 mg, 81%).

Mp 127-128 °C (*i*PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.80 (1H, d, *J* = 1.5 Hz), 8.61 (1H, dd, *J* = 4.8, 1.6 Hz), 7.86-7.81 (1H, m), 7.38 (1H, dd, *J* = 7.4, 4.8 Hz), 7.30-7.25 (4H, m), 7.04-6.98 (3H, m), 4.36 (2H, s), 3.86 (3H, s), 3.29 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.4, 159.7, 154.7, 148.6, 148.4, 142.7, 136.3, 134.6, 134.5, 134.3, 132.1, 128.4, 123.6, 121.9, 115.6, 110.5, 107.5, 55.6, 43.9, 37.9; IR (ATR) 1772, 1655, 1510, 1477, 1248 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{20}$N$_3$O$_4$ [M+H]$^+$ 390.1448; Found 390.1443; Anal. Calcd for C$_{22}$H$_{19}$N$_3$O$_4$·0.50H$_2$O: C, 66.32; H, 5.06; N, 10.55. Found: C, 66.31; H, 4.97; N, 10.82.

N-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2H)-yl]-N-[4-(trifluoromethyl)phenyl] acetamide (88d)

Compound **88d** was prepared from **109** (56.0 g, 183 mmol) and *N*-methyl-4-(trifluoromethyl)aniline (41.6 g, 237 mmol) according to general procedure D as a pale yellow solid (52.5 g, 67%).

Mp 152-154 °C (EtOAc-Et$_2$O); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.79 (1H, d, *J* = 2.4 Hz), 8.60 (1H, dd, *J* = 4.9, 2.4 Hz), 7.85-7.75 (3H, m), 7.52 (2H, d, *J* = 7.8 Hz), 7.37 (1H, dd, *J* = 7.8, 4.9 Hz), 7.30 (2H, d, *J* = 0.7 Hz), 7.06 (1H, s), 4.38 (2H, s), 3.35 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.8, 154.5, 148.7, 148.3, 145.0, 142.7, 136.1, 134.5, 134.4, 131.9, 131.3 (m), 127.8 (m), 127.7 (m), 124.8, 123.5, 122.0, 110.6, 107.5, 43.9, 37.8; IR (ATR) 1768, 1674, 1612, 1323, 1120 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{17}$F$_3$N$_3$O$_3$ [M+H]$^+$ 428.1217; Found 428.1208; Anal. Calcd for C$_{22}$H$_{16}$F$_3$N$_3$O$_3$: C, 61.83; H, 3.77; N, 9.83; F, 13.34. Found: C, 61.74; H, 3.79; N, 9.98; F, 13.08.

N-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2H)-yl]-N-[4-(trifluoromethyl)phenyl] acetamide hydrochloride (88d·HCl)

To a solution of **88d** (90.0 g, 211 mmol) in EtOH (300 mL) was added 36% HCl solution (263 mL) at room temperature. The reaction mixture was stirred at room temperature for 0.5 h. The solvent was removed in vacuo, and the resulting solid was recrystallized from EtOH to give hydrochloride of **88d** (95.6 g, 98%) as a white solid.

Mp 250-252 $^{\circ}$C; ^{1}H-NMR (400 MHz, DMSO-d_6) δ: 9.22 (1H, d, J = 1.7 Hz), 8.86 (1H, d, J = 4.6 Hz), 8.75 (1H, d, J = 8.3 Hz), 8.09-8.02 (1H, m), 8.02-7.69 (5H, br m), 7.67 (1H, dd, J = 8.3, 1.8 Hz), 7.56 (1H, d, J = 8.3 Hz), 4.57 (2H, br s), 3.31 (3H, br s); ^{13}C-NMR (100 MHz, DMSO-d_6) δ: 164.9, 153.9, 145.8, 142.8, 142.1, 141.4, 141.2, 137.7, 132.5, 130.4 (m), 128.2 (m), 127.1 (m), 126.5, 125.3, 122.6, 121.8, 110.4, 108.7, 44.1, 37.0; IR (ATR) 2362, 1770, 1662, 1317, 1159 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{22}H_{17}F_3N_3O_3$ [M+H]$^+$ 428.1217; Found 428.1204; Anal. Calcd for $C_{22}H_{17}ClF_3N_3O_3$: C, 56.97; H, 3.69; N, 9.06; Cl, 7.64; F, 12.29. Found: C, 56.97; H, 3.73; N, 9.11; Cl, 7.64; F, 12.22.

N-Methyl-2-[2-oxo-5-(pyridin-3-yl)-1,3-benzoxazol-3(2H)-yl]-N-[4-(trifluoromethoxy)phenyl] acetamide (88e)

Compound **88e** was prepared from **109** (399 mg, 1.30 mmol) and N-methyl-4-(trifluoromethoxy)aniline (298 mg, 1.56 mmol) according to general procedure D as a white solid (236 mg, 41%).

Mp 125-127 $^{\circ}$C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.80 (1H, d, J = 1.7 Hz), 8.61 (1H, dd, J = 5.1, 1.7 Hz), 7.86-7.82 (1H, m), 7.44-7.35 (5H, m), 7.31-7.30 (2H, m), 7.05 (1H, s), 4.37 (2H, s), 3.32 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.0, 154.6, 149.2, 148.7, 148.4, 142.7, 140.3, 136.3, 134.6, 134.5, 132.0, 129.0, 123.6, 122.9, 122.0, 121.6, 110.6, 107.5, 43.9, 38.0; IR (ATR) 1759, 1655, 1481, 1254, 1215 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{22}H_{17}F_3N_3O_4$ [M+H]$^+$ 444.1166; Found 444.1155; Anal. Calcd for $C_{22}H_{16}F_3N_3O_4$·0.50H$_2$O: C, 58.41; H, 3.79; N, 9.29; F, 12.60. Found: C, 58.44; H, 3.77; N, 9.49; F, 12.31.

N-Ethyl-2-[2-oxo-5-(pyridin-4-yl)-1,3-benzoxazol-3(2H)-yl]-N-phenylacetamide (89b)

Compound **89b** was prepared from **110** (307 mg, 1.00 mmol) and N-ethylaniline (151 μL, 1.20 mmol) according to general procedure C as a white solid (244 mg, 65%).

Mp 134-136 $^{\circ}$C (iPrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.67 (2H, dd, J = 4.5, 1.6 Hz), 7.53 (2H, dd, J = 7.4, 7.4 Hz), 7.50-7.43 (3H, m), 7.38-7.26 (4H, m), 7.10 (1H, d, J = 1.7 Hz), 4.32 (2H, s), 3.80 (2H, q, J = 7.2 Hz), 1.16 (3H, t, J = 7.2 Hz); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.4, 154.5, 150.3, 147.9, 143.3, 140.2, 134.7, 132.2, 130.4, 129.0, 128.3, 121.8, 110.5, 107.4, 44.8, 44.1, 12.9; IR (ATR) 1768, 1664, 1595, 1481, 1274 cm^{-1}; HRMS (ESI) m/z Calcd for $C_{22}H_{20}N_3O_3$ [M+H]$^+$ 374.1499; Found 374.1494; Anal. Calcd for $C_{22}H_{19}N_3O_3$: C, 70.76; H, 5.13; N, 11.25. Found: C, 70.83; H, 5.16; N, 11.35.

_N_-(4-methoxyphenyl)-_N_-methyl-2-[2-oxo-5-(pyridin-4-yl)-1,3-benzoxazol-3(2_H_)-yl]acetamide (89c)

Compound **89c** was prepared from **110** (307 mg, 1.00 mmol) and _N_-methyl-_p_-anisidine (165 mg, 1.20 mmol) according to general procedure C as a white solid (291 mg, 75%).

Mp 234-235 °C (_i_PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.66 (2H, dd, J = 4.4, 1.7 Hz), 7.46 (2H, dd, J = 4.4, 1.7 Hz), 7.36 (1H, dd, J = 8.3, 1.7 Hz), 7.31-7.25 (3H, m), 7.10 (1H, d, J = 1.7 Hz), 7.04-6.98 (2H, m), 4.36 (2H, s), 3.86 (3H, s), 3.29 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.3, 159.7, 154.5, 150.3, 147.9, 143.3, 134.7, 134.5, 132.2, 128.4, 121.8, 115.6, 110.5, 107.4, 55.6, 43.8, 37.9; IR (ATR) 1780, 1668, 1599, 1512, 1248 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{20}$N$_3$O$_4$ [M+H]$^+$ 390.1448; Found 390.1443; Anal. Calcd for C$_{22}$H$_{19}$N$_3$O$_4$: C, 67.86; H, 4.92; N, 10.79. Found: C, 67.92; H, 4.94; N, 10.90.

_N_-Methyl-2-[2-oxo-5-(pyridin-4-yl)-1,3-benzoxazol-3(2_H_)-yl]-_N_-[4-(trifluoromethyl)phenyl] acetamide (89d)

Compound **89d** was prepared from **110** (375 mg, 1.30 mmol) and _N_-methyl-4-(trifluoromethyl)aniline (273 mg, 1.56 mmol) according to general procedure D as a white solid (241 mg, 43%).

Mp 193-194 °C (_i_PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.66 (2H, d, J = 6.1 Hz), 7.80 (2H, d, J = 7.8 Hz), 7.52 (2H, d, J = 7.8 Hz), 7.48-7.43 (2H, m), 7.38 (1H, dd, J = 8.0, 2.0 Hz), 7.31 (1H, d, J = 8.0 Hz), 7.13 (1H, s), 4.38 (2H, s), 3.36 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 164.8, 154.4, 150.3, 147.8, 145.1, 145.1, 143.3, 134.8, 132.0 (m), 127.8, 127.7 (m), 127.6 (m), 122.0, 121.8, 110.6, 107.4, 43.9, 37.8; IR (ATR) 1780, 1659, 1595, 1489, 1331 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{17}$F$_3$N$_3$O$_3$ [M+H]$^+$ 428.1217; Found 428.1209; Anal. Calcd for C$_{22}$H$_{16}$F$_3$N$_3$O$_3$: C, 61.83; H, 3.77; N, 9.83; F, 13.34. Found: C, 61.51; H, 3.86; N, 10.01; F, 13.26.

_N_-Methyl-2-[2-oxo-5-(pyridin-4-yl)-1,3-benzoxazol-3(2_H_)-yl]-_N_-[4-(trifluoromethoxy)phenyl] acetamide (89e)

Compound **89e** was prepared from **110** (50.0 mg, 163 μmol) and _N_-methyl-4-(trifluoromethoxy)aniline (37.4 mg, 196 μmol) according to general procedure C as a white solid (34.8 mg, 48%).

Mp 196-197 °C (_i_PrOH); ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 8.67 (2H, dd, J = 4.4, 1.7 Hz), 7.47-7.36 (7H, m), 7.30 (1H, d, J = 8.5 Hz), 7.12 (1H, s), 4.37 (2H, s), 3.33 (3H, s); ^{13}C-NMR (100 MHz, CDCl$_3$) δ: 165.0, 154.4, 150.3, 149.2, 147.9, 143.3, 140.3, 134.8, 132.1, 129.0, 122.9, 122.0, 121.8, 116.0, 110.7, 107.4, 43.9, 38.0; IR (ATR) 1782, 1674, 1597, 1483, 1161 cm^{-1}; HRMS (ESI) m/z Calcd for C$_{22}$H$_{17}$F$_3$N$_3$O$_4$ [M+H]$^+$ 444.1166; Found 444.1155; Anal. Calcd for C$_{22}$H$_{16}$F$_3$N$_3$O$_4$: C, 59.60; H, 3.64; N, 9.48; F, 12.85. Found: C, 59.70; H, 3.64; N, 9.66; F, 12.73.

6-2 Pharmacology

Rat TSPO-binding assay

Male SD rats (Japan Charles River) were decapitated, and the kidney was dissected. The kidney was homogenized in 5 volumes of ice-cold 50 mM Tris-HCl buffer (pH 7.6) with a microhomogenizer (Physcotron, Niti-on Inc.). The homogenate was centrifuged at 20,000 g and 4 ^{o}C for 10 min, and the resulting pellet was resuspended in the same volume of fresh buffer and recentrifuged. Resuspension and centrifugation were repeated once more, and the obtained pellet was resuspended in 50 mM Tris-HCl buffer (pH 7.6) at a protein concentration of 2.63 mg/mL and stored frozen at -80 ^{o}C until use. The mitochondrial membrane suspension (0.895 mL) was incubated with [^{3}H]-PK11195 (final concentration 1.0 nM) and various concentrations of the test compounds in a total volume of 1.0 mL for 1 h at 4 ^{o}C. The reaction was terminated by rapid filtration through a GF/B glass filter presoaked with 0.3% polyethyleneimine. The filters were immediately washed with ice-cold 50 mM Tris-HCl buffer (pH 7.6), and the filter-bound radioactivity was quantified using a liquid scintillation analyzer (Tri Carb 2700TR, Packard). Nonspecific binding was determined in the presence of 10 μM PK11195. All assays were carried out in duplicate except for total binding and nonspecific binding, which were in quadruplicate. Specific binding was determined by subtracting nonspecific from total binding. The IC_{50} values for each test compound were determined according to a nonlinear least-square curve-fitting method using the SAS$^®$ system (SAS Institute Inc.). In the assay with rat kidney TSPOs, K_i values were calculated according to the following formula: $K_i = IC_{50}/(1+[L]/K_D)$ where [L] and K_D are the concentration of [^{3}H]-PK11195 and the dissociation constant of PK11195 calculated by Scatchard analysis, respectively.

Human TSPO-binding assay

Human U-87 MG glioma cells (HTB-14, American Type Culture Collection, Rockville, MD, U.S.A.) were cultivated in Dulbecco's modified Eagle's medium supplemented with 10% heat-inactivated fetal bovine serum, 10,000 U/mL penicillin and 10 mg/mL streptomycin, until confluence. The cells were washed with 15 mL of 50 mM Tris-HCl buffer (pH 7.6) and harvested from the tissue culture flask with a cell scraper. The collected cell pellet was centrifuged at 12,900 g and 4 ^{o}C for 10 min, and the resulting pellet was homogenized in the 15 mL of 50 mM Tris-HCl buffer (pH 7.6) with a microhomogenizer (Physcotron, Niti-on Inc). Centrifugation and homogenization were repeated once more, and the obtained pellet was suspended in 50 mM Tris-HCl buffer (pH 7.6) at a protein concentration of 4.3 mg/mL and stored frozen -80 ^{o}C until use. The mitochondrial membrane suspension (149 μL) was incubated with [^{3}H]-PK11195 (final concentration 1.0 nM) and various concentration of the test compounds in a total volume of 200 μL for 1 h at 4 ^{o}C. The reaction was terminated by rapid fitration through a GF/B glass filter presoaked with 0.3% polyethyleneimine. The filters were immediately washed with ice-cold 50 mM

Tris-HCl buffer (pH 7.6), and the filter-bound radioactivity was quantified using a liquid scintillation analyzer (Tri Carb 2700TR, Packard). Nonspecific binding was determined in the presence of 10 μM PK11195. All assays were carried out in duplicate except for total binding and nonspecific binding, which were in quadruplicate. Specific binding was determined by subtracting nonspecific from total binding. The IC_{50} values for each test compound were determined according to a nonlinear least-square curve-fitting method using the SAS® system (SAS Institute Inc.). K_i values were calculated according to the same procedure as that described above in the assay with rat TSPOs.

Rat CBR-binding assay

Male SD rats (Japan Charles River) were decapitated, and the cerebral cortex was dissected. The cerebral cortex suspension was homogenized in 10 volumes of ice-cold potassium phosphate buffer (200 mM KCl, 20 mM KOH, 20 mM KH_2PO_4, pH 7.4) with a microhomogenizer (Physcotron, Niti-on Inc.). The homogenate was centrifuged at 32,500 g and 4 °C for 15 min, and the resulting pellet was resuspended in the same volume of fresh buffer and recentrifuged. Resuspension and centrifugation were repeated once more, and the obtained pellet was resuspended in potassium phosphate buffer (200 mM KCl, 20 mM KOH, 20 mM KH_2PO_4, pH 7.4) at a protein concentration of 2.63 mg/mL and stored frozen at -80 °C until use. The cerebral cortex membrane suspension (0.895 mL) was incubated with [³H]-flumazenil (final concentration 1.0 nM) and 10 μM of the test compounds in a total volume of 1.0 mL for 1 h at 25 °C. The reaction was terminated by rapid fitration through a GF/B glass filter presoaked with 0.3% polyethyleneimine. The filters were immediately washed with ice-cold potassium phosphate buffer (200 mM KCl, 20 mM KOH, 20 mM KH_2PO_4, pH 7.4), and the filter-bound radioactivity was quantified using a liquid scintillation analyzer (Tri Carb 2700TR, Packard). Nonspecific binding was determined in the presence of 10 μM [³*H*]- flumazenil. All assays were done in duplicate. Specific binding was determined by subtracting nonspecific from total binding.

Vogel-type conflict test in rats

For this test, the method of Vogel and colleagues[32] with a minor modification was used in male SD rats. For the test operant, behavior boxes (26 x 25 x 16.5 cm^3) with a stainless steel grid floor (Ohara Co., Ltd.) were used. A water bottle with a metal drinking tube was fitted from the outside to the box so that only the drinking tube extended into the box. Electric shocks (0.30 mA or 0.50 mA, 0.5 sec) were administered to each rat by automatically switching the connections to the drinking tube and the grid floor from the drinko-meter to an electric stimulator. After 48 h of water deprivation, the rats were individually placed in the test chamber. The number of shocks that each rat received after every 20 licks was recorded for 3 min. Test compounds were orally administered 1 h before the test session.

Rota-rod test

Male ddY mice were placed on a rota-rod (a 3 cm in diameter rod supported horizontally and rotated at 6 rpm) with their head against the direction of rotation before the experiment. Mice that could stay on the rod for 2 min or more were selected. One hour after oral administration of test compounds or vehicle each mouse was placed on the rota-rod again and the time spent on the rod was measured.

Step-through passive avoidance test

Male Wistar rats were obtained from Charles River Japan. The apparatus consisted of a light compartment (20 x 25 x 20 cm^3) and a dark compartment (20 x 15 x 20 cm^3) with a grid floor (PA-3001A, Ohara Co., Ltd.). These two compartments were separated by a sliding door. In the training session, the animals were placed in the light compartment and allowed to explore for 10 s. The sliding door was then opened, and the step-through latency for animals to enter the dark compartment was measured. As soon as the animals entered the dark compartment, the door was closed. Three seconds later, an inescapable foot-shock (0.5 mA, 3 s) was delivered through the grid floor with a constant current shock generator (PA-2030A, Ohara Co., Ltd.). All of the animal examined in this study entered the dark compartment within 300 s as cut-off latency in the training session and received a foot-shock. The test session was performed 24 h after the training session using the same paradigm, but without the foot-shock, and the step-through latency for animals to enter the dark compartment was measured. When an animal did not enter the dark room within 300 s, the step-through latency was recorded as 300 s.

For investigation for effects of test compounds on the passive avoidance response, test compounds were administered orally 1h before training session.

For examination of the effects of test compounds on MK-801-induced deficits in the passive avoidance acquisition, MK-801 (0.05 mg/kg, s.c.) was administered twice 30 min before the traning and test session. Test compounds were orally administered 1h before the training session. Trilostane (10 mg/kg, i.p.) was administered 1.5 h before the training session.

Potentiation of hexobarbital-induced anesthesia

Male ddY mice were given orally test compounds or vehicle. One hour later, hexabarbital (70 mg/kg, i.p.) was administered, and the time of loss of righting reflexes was measured.

Locomotor activity test

Male ddY mice were individually isolated in clear plastic cages (26.5 x 42.5 x 20 cm^3). Locomotor activity was measured by Supermex (Muromachi Kikai Co., Ltd.) for 30 min. This test was conducted 1 h after oral administration of test compounds.

引用文献

1. Taliani, S.; Da Settimo, F.; Da Pozzo, E.;Chelli, B.; Martini, C. *Curr. Med. Chem.* **2009**, *16*, 3359.

2. (a) Bandelow, B.; Zohar, J.; Hollander, E.; Kasper, S,; Möller, H.-J. *World J. Biol. Psychiatry* **2008**, *9*, 248.

   (b) Griebel, G.; Holmes, A. *Nat. Rev. Drug Discovery* **2013**, *12*, 667.

3. (a) O'Brien, C. P. *J. Clin. Psychiatry* **2005**, *66*, 28.

   (b) Rudolph, U.; Knoflach, F. *Nat. Rev. Drug Discovery* **2011**, *10*, 685.

4. (a) Squires, R. F.; Braestrup, C. *Nature*, **1977**, *266*, 732.

   (b) Braestrup, C.; Albrechtsen, R.; Squires, R. F. *Nature*, **1977**, *269*, 702.

   (c) Möhler, H.; Okada, T. *Science*, **1977**, *198*, 849.

5. (a) Costa, E.; Guidotti, A. *Annu. Rev. Pharmacol. Toxicol.* **1979**, *19*, 531.

   (b) Möhler, H; Fritschy, J. M.; Rudolph, U. *J. Pharmacol. Exp. Ther.* **2002**, *300*, 2.

6. Braestrup, C.; Squires, R. F. *Proc. Natl. Acad. Sci. U.S.A.* **1977**, *74*, 3805.

7. (a)Gavish, M.; Bachman, I.; Shoukrun, R.; Katz, Y.; Veenman, L.; Weisinger, G.; Weizman, A. *Pharmacol. Rev.* **1999**, *51*, 629.

   (b) Scarf, A. M.; Ittner, L. M.; Kassiou, M. *J. Med. Chem.* **2009**, *52*, 581.

   (c) Rupprecht, R.; Papadopoulos, V.; Rammes, G.; Baghai, T. C.; Fan, J.; Akula, N.; Groyer, G.; Adams, D.; Schumacher, M. *Nat. Rev. Drug Discovery* **2010**, *9*, 971.

8. Anholt, R. R. H.; Pedersen, P. L.; De Souza, E. B.; Snyder, S. H. *J. Biol. Chem.* **1986**, *261*, 576.

9. Papadopolous, V.; Baraldi, M.; Guilarte, T. R.; Knudsen, T. B.; Lacapère, J.-J.; Lindemann, P.; Norenberg, M. D.; Nutt, D.; Weizman, A.; Zhang, M.-R.; Gavish, M. *Trends Pharmacol. Sci.* **2006**, *27*, 402.

10. Schoemaker, H.; Bliss, M.; Yamamura, H. I. *Eur. J. Pharmacol.* **1981**, *71*, 173.

11. Mcenery, M. W.; Snowman, A. M.; Trifiletti, R. R.; Snyder, S. H. *Proc. Natl. Acad. Sci. U.S.A.* **1992**, *89*, 3170.

12. (a) Lacapère, J.-J.; Papadopoulos. V. *Steroids* **2003**, *68*, 569.

    (b) Da Pozzo, E.; Costa, B.; Martini, C. *Curr. Mol. Med.* **2012**, *12*, 426.

13. Chelli, B.; Falleni, A.; Salvetti, F.; Gremigni, V.; Lucacchini, A.; Martini, C. *Biochem. Pharmacol.* **2001**, *61*, 695.

14. Casellas, P.; Galiegue, S.; Basile, A. S. *Neurochem. Int.* **2002**, *40*, 475.

15. (a) Zavala, F. *Pharmacol. Ther.* **1997**, *75*, 199.

    (b) Bribes, E.; Bourrie, B.; Esclangon, M.; Galiegue, S.; Vidal, H.; Casellas, P. *Eur. J. Pharmacol.* **2002**, *452*, 111.

16. (a) Krueger, K. E.; Papadopoulos, V. *J. Biol. Chem.* **1990**, *265*, 15015.

(b) Papadopoulos, V.; Amri, H.; Boujrad, N.; Cascio, C.; Culty, M.; Garnier, M.; Hardwick, M.; Li, H.; Vidic, B.; Brown, A. S.; Reversa, J. L.; Bernassau, J. M.; Drieu, K. *Steroids* **1997**, *62*, 21.

17. Stocco, D. M. *Annu. Rev. Physiol*. **2001**, *63*, 193.

18. (a) Semeniuk, T.; Jhangri, G. S. Le Mellédo J.-M. *J. Neuropsychiatry Clin. Neurosci*. **2001**, *13*, 398.

    (b) Le Mellédo J.-M.; Baker G. B. *J. Psychiatry Neurosci*. **2002**, *27*, 161.

19. Uzunova, V.; Sheline, Y.; Davis, J. M.; Rasmusson, A.; Uzunov, D. P.; Costa, E.; Guidotti, A. *Proc. Natl. Acad. Sci. U.S.A*. **1998**, *95*, 3239.

20. (a) Wolkowitz, O. M.; Reus, V. I.; Keebler, A.; Nelson, N.; Friedland, M.; Brizendine, L.; Roberts, E. *Am. J. Psychiatry* **1999**, *156*, 646.

    (b) Khisti, R. T.; Chopde, C. T.; Jain, S. P. *Pharmacol., Biochem. Behav*. **2000**, *67*, 137.

    (c) Gavioli, E. C.; Duarte, F. S.; Bressan, E.; Ferrara, P.; Farges, R. C.; De Lima, T. C. M. *Eur. J. Pharmacol*. **2003**, *471*, 21.

21. (a) Rupprecht, R.; Holsboer, F. *Trends Neurosci*. **1999**, *22*, 410.

    (b) Rupprecht, R.; di Michele, F.; Hermann, B.; Ströhle, A.; Lancel, M.; Romeo, E.; Holsboer, F. *Brain Res. Rev*. **2001**, *37*, 59.

    (c) Mellon, S. H.; Griffin, L. D. *Trends Endocrin. Met*. **2002**, *13*, 35

22. Belelli, d.; Lambert, J. J. *Nature Rev. Neurosci*. **2005**, *6*, 565.

23. Carter, R. B.; Wood, P. L.; Wieland, S.; Hawkinson, J. E.; Belelli, D.; Lambert, J. J.; White, H. S.; Wolf, H. H.; Mirsadeghi, S.; Tahir, S. H.; Bolger, M. B.; Lan, N. C.; Gee, K. W. *J. Pharmacol. Exp. Ther*. **1997**, *280*, 1284.

24. (a) Weizman, R.; Tanne, Z.; Granek, K.; Karp. L.; Golomb, M.; Tyano, S.; Gavish, M. *Eur. J. Pharmacol*. **1987**, *138,*. 289.

    (b) Gavish, M.; Laor, N.; Bidder, M.; Fisher, D.; Fonia, O.; Muller, U.; Reiss, A.; Wolmer, L.; Karp, L.; Weizman, R. *Neuropsychopharmacology* **1996**, *14*, 181.

25. (a) Corda, M. G.; Ferrari, M.; Guidotti, A.; Konkel, D.; Costa, E. *Neurosci. Lett*. **1984**, *47*, 319.

    (b) Gray, P. W.; Glaister, D.; Seeburg, P. H.; Guidotti, A.; Costa, E. *Proc. Natl. Acad. Sci. U.S.A*. **1986**, *83*, 7547.

    (c) Papadopoulos, V. *Endocr. Rev*. **1993**, *14*, 222.

26. (a) Snyder, S. H.; Verma, A.; Trifiletti, R. R. *FASEB J*. **1987**, *1*, 282.

    (b) Verma, A.; Nye, J. S.; Snyder, S. H. *Proc. Natl. Acad. Sci. U.S.A*. **1987**, *84*, 2256.

    (c) Verma, A.; Snyder, S. H. *Mol. Pharmacol*. **1988**, *34*, 800.

27. (a) Le Fur, G.; Vaucher, N.; Perrier, M. L.; Flamier, A.; Benavides, J.; Renault, C.; Dubroeucq, M. C.; Guérémy, C. Uzan, A. *Life Sci*. **1983**, *33*, 449.

    (b) Schoemaker, H.; Boles, R. G.; Horst, W. D.; Yamamura, H. I. *J. Pharmacol. Exp. Ther*. **1983**, *225*, 61.

(c) Farges, R.; Joseph-Liauzun, E.; Shire, D.; Caput, D.; Le Fur, G.; Ferrara, P. *Mol. Pharmacol*. **1994**, *46*, 1160.

28. (a) Le Fur, G.; Perrier, M. L.; Vaucher, N.; Imbault, F.; Flamier, A.; Benavides, J.; Uzan, A.; Renault, C.; Dubroeucq, M. C.; Guérémy, C. *Life Sci*. **1983**, *32*, 1839.

(b) Le Fur, G.; Guilloux, F.; Rufat, P.; Benavides, J.; Uzan, A.; Renault, C.; Dubroeucq, M. C.; Guérémy, C. *Life Sci*. **1983**, *32*, 1849.

29. Garnier, M.; Dimchev, A. B.; Boujrad, N.; Price, J. M.; Musto, N. A.; Papadopoulos, V. *Mol. Pharmacol*. **1994**, *45*, 201.

30. (a) Okuyama, S.; Chaki, S.; Yoshikawa, R.; Ogawa, S.; Suzuki, Y.; Okubo, T.; Nakazato, A.; Nagamine, M.; Tomisawa, K. *Life Sci*. **1999**, *64*, 1455

(b) Okubo, T.; Yoshikawa, R.; Chaki, S.; Okuyama, S.; Nakazato, A. *Bioorg. Med. Chem*. **2004**, *12*, 423.

31. (a) Kozikowski, A. P.; Ma, D.; Rome, E.; Auta, J.; Papadopoulos, V.; Puia, G.; Costa, E.; Guidotti, *Angew. Chem. Int. Ed.* **1992**, *31*, 1060

(b) Romeo, E.; Auta, J.; Kozikowski, A. P.; Ma, D.; Papadopoulos, V.; Puia, G.; Costa, E.; Guidotti, A. *J. Pharmacol. Exp. Ther*. **1992**, *262*, 971.

(c) Auta, J.; Romeo, E.; Kozikowski, A.; Ma, D.; Costa, E.; Guidotti, A. *J. Pharmacol. Exp. Ther*. **1993**, *265,* 649.

(d) Romeo, E.; Cavallaro, S.; Korneyev, A.; Kozikowski, A. P.; Ma, D.; Polo, A.; Costa, E.; Guidotti, A. *J. Pharmacol. Exp. Ther*. **1993**, *267*, 462.

32. (a) Kita, A.; Kohayakawa, H.; Kinoshita, T.; Ochi, Y.; Nakamichi, K.; Kurumiya, S.; Furukawa, K.; Oka, M. *Br. J. Pharmacol.* **2004**, *142*, 1059.

(b) Kita, A.; Furukawa, K. *Pharmacol. Biochem. Behav*. **2008**, *89*, 171.

(c) Kita, A.; Kinoshita, T.; Kohayakawa, H.; Furukawa, K.; Akaike, A. *Prog. Neuro-psychopharmacol. Biol. Psychiatry* **2009**, *33*, 1040.

(d) Rupprecht, R,; Rammes, G.; Eser, D.; Baghai, T. C.; Schüle, C.; Nothdurfter, C.; Troxler, T.; Gentsch, C.; Kalkman, H. O.; Chaperon, F.; Uzunov, V.; McAllister, K. H.; Bertaina-Anglade, V.; La Rochelle, C. D.; Tuerck, D.; Floesser, A.; Kiese, B.; Schumacher, M.; Landgraf, R.; Holsboer, F.; Kucher, K. *Science*, **2009**, *325*, 490.

33. Kodo, T.; Fukaya, T.; Koyama, K.; Masumoto, S.; Fujibayashi, N. PCT Int. Appl. WO2005/080334, 2005

34. (a) Campiani, G.; Nacci, V.; Fiorini, I., De Filippis, M. P.; Garofalo, A.; Ciani, S. M.; Greco, G.; Novellino, E.; Williams, D. C.; Zisterer, D. M.; Woods, M. J.; Mihai, C. M.; Manzoni, C.; Mennini, T. *J. Med. Chem.* **1996,** *39,* 3435

(b) Anzini, M.; Cappelli, A.; Vomero, S.; Giorgi, G.; Langer, T.; Bruni, G.; Romeo, M. R.; Basile, A. S. *J. Med. Chem.* **1996,** *39,* 4275

(c) Anzini, M.; Cappelli, A.; Vomero, S.; Seeber, M.; Menziani, M. C.; Langer, T.; Hagen, B.; Manzoni, C.; Bourguignon, J.-J. *J. Med. Chem.* **2001,** *44,* 1134.

35. Miyaura, N.; Suzuki, A. *Chem. Rev.* **1995**, *95*, 2457.

36. Stille, J. K. *Angew. Chem. Int. Ed.* **1986**, *25*, 508.

37. Harris, M. C.; Geis, O.; Buchwald, S. L. *J. Org. Chem.* **1999**, *64*, 6019.

38. (a) Smith, D. A.; Jones, B. C.; Walker, D. K. *Med. Res. Rev.* **1996**, *16*, 243

(b) Thompson, T. N. *Med. Res. Rev.* **2001**, *21*, 412

(c) van de Waterbeemd, H.; Smith, D. A.; Beaumont, K.; Walker, D. K. *J. Med Chem.* **2001**, *44*, 1313.

39. Primofiore, G.; Da Settimo, F.; Taliani, S.; Simorini, F.; Patrizi, M. P.; Novellino, E.; Greco, G.; Abignente, E.; Costa, B.; Chelli, B.; Martini, C. *J. Med. Chem.* **2004**, *47*, 1852.

40. Colbert, J. C.; Meigs, W.; Jenkins, R. L. *J. Am. Chem. Soc.* **1937**, *59*, 1122.

41. (a) Colbert, J. C.; Fox, D. W.; Matuszak, C. *J. Am. Chem. Soc.* **1955**, *77*, 2447

(b) Tarzia, G.; Duranti, A.; Gatti, G.; Piersanti, G.; Tontini, A.; Rivara, S.; Lodola, A.; Plazzi, P. V.; Mor, M.; Kathuria, S.; Piomelli, D. *Chem. Med. Chem.* **2006**, *1*, 130.

42. Taneda, S.; Kamata, K.; Hayashi, H.; Toda, N.; Seki, K.; Sakushima, A.; Yoshino, S.; Yamaki, K.; Sakata, M., Mori, Y.; Suzuki, A. K. *J. Health Sci.* **2004**, *50*, 133.

43. Boyland, E.; Sims, P. *J. Chem. Soc.* **1954**, 980.

44. Akao, A.; Sato, K.; Nonoyama, N.; Mase, T.; Yasuda, N. *Tetrahedoron Lett.* **2006**, *47*, 969.

45. von Wantoch Rekowski, M.; Pyriochou, A.; Papapetropoulos, N.; Stößel, A; Papapetropoulos, A.; Giannis, A. *Bioorg. Med. Chem.* **2010**, *18*, 1288.

46. (a) Ishiyama, T.; Murata, M.; Miyaura, N. *J. Org. Chem.* **1995**, *60*, 7508.

(b) Ishiyama, T.; Itoh, Y.; Kitano, T.; Miyaura, N. *Tetrahedron Lett.* **1997**, *38*, 3447.

(c) Ishiyama, T.; Ishida, K.; Miyaura, N. *Tetrahedron* **2001**, *57*, 9813.

47. Tomita, M.; Fujitani, K.; Aoyagi, Y. *Chem. Pharm. Bull.* **1965**, *13,*1341.

48. (a) Hansch, C.; Leo, A.; Unger, S. H.; Kim, K. H.; Nikaitani, D.; Lien, E. J. *J. Med. Chem.* **1973,** *16,* 1207.

(b) Hansch, C.; Leo, A.; Taft, R. W. *Chem. Rev.* **1991**, *91*, 165.

49. Phan V.-L.; Su, T.-R.; Privat, A.; Maurice, T. *Eur. J. Neurosci.* **1999**, *11*, 2385.

50. Vogel, J. R.; Beer, B.; Clody, D. E. *Psycopharmacologia* **1971**, *21*, 1.

51. Ishiyama, T.; Tokuda, K.; Ishibashi, T.; Ito, A. Toma, S.; Ohno, Y. *Eur. J. Pharmacol.* **2007**, *572*, 160.

52. (a) Anger, T.; Madge, D. J.; Mulla, M.; Riddall, D. *J. Med. Chem.* **2001**, *44*, 115.

(b) Levin, S. I.; Khaliq, Z. M.; Aman, T. K.; Grieco, T. M.; Kearney, J. A.; Raman, I. M.; Meisler, M. H. *J. Neurophysiol*, **2006**, *96*, 785.

(c) Kalume, F.; Yu, F. H.; Westenbroek, R. E.; Scheuer, T.; Gatterall, W. A. *J. Neurosci.* **2007**, *27*, 11065.

53. Schweitzer, P. J.; Fallon, B. A.; Mann, J. J.; Kumar, J. S. D. *Drug Discovery Today* **2010,** *15*, 933.