Novel Water-Soluble Sedative-Hypnotic Agents: Isoindolin-1-one Derivatives

Norimasa KANAMITSU,* Takashi OSAKI, Yutaka ITSUJI, Masakazu YOSHIMURA, Hisashi TSUJIMOTO, and Manabu SOGA

Central Research Laboratory, Maruishi Pharmaceutical Co., Ltd.; 2–2–18 Imazu-naka, Tsurumi-ku, Osaka 538–0042, Japan. Received June 13, 2007; accepted October 4, 2007; published online October 5, 2007

We developed new intravenous sedative-hypnotic compounds with the isoindolin-1-one skeleton focusing on the water-soluble property and *in vivo* safety. We synthesized approximately 170 derivatives and evaluated their hypnotic effects by intravenous administration of the compounds to mice. A series of the 2-phenyl-3-[2-(4-methyl-1-piperazinyl)-2-oxoethyl]isoindolin-1-one analogs, 3(−), 5(−), 27(−), and 47(−) [JM-1232(−)], showed potent sedative-hypnotic activity with good water solubility and a wide safety margin. The hypnotic doses (HD_{50}s) of these 4 compounds when administered to mice were 2.35, 1.90, 2.17, and 3.12 mg/kg, respectively, and the lethal doses (LD_{50}s) were 88.67, 64.69, >120, and >120 mg/kg, respectively. The therapeutic indexes (LD_{50}/HD_{50}) were 37.73, 34.05, >55.30, and >38.46, respectively. Among these compound, 47(−) [JM-1232(−)] is being considered as the most potential candidate for clinical trials in humans.

Key words isoindolin-1-one derivative; sedative-hypnotic; nonbenzodiazepine; water-soluble

Currently, propofol is widely used as an intravenous anesthetic and sedative in clinical settings. Propofol is hardly soluble in water in nature and is prescribed as an emulsion formulation containing soybean oil, glycerin, and egg phospholipids.[1] As a result, the long-term use of a propofol emulsion, for example in post-operative sedation, may not only lead to overload of the fat nutrition[2] but also an increase in the risk of infection in patients by rapid microbial growth at room temperature.[3] In addition, clinical problems that have been noted with propofol include vascular pain on injection[4] and strong respiratory depression.[5] Based on the points above-mentioned, aiming at the discovery of an anesthetic and sedative as an easy-to-formulate water-soluble drug with reduced respiratory depression, we initiated a search for a compound suitable as a lead. Recently nonbenzodiazepine compounds with selective pharmacological actions on sedation and anti-anxiety have been developed.[6,7] Therefore, amongst the parent skeletons of these nonbenzodiazepine compounds, we took note of the isoindolin-1-one skeleton[6] (Fig. 1), and synthesized approximately 170 compounds and evaluated their hypnotic effects in mice after intravenous administration.[8] Herein, we report the representative preferred compounds with potent sedative-hypnotic actions.

Chemistry

The general synthetic method used to prepare isoindolin-1-one derivatives is as shown in Chart 1. Phthalic anhydride **30** was heated with an appropriate amine to afford phthalimides **31**, which were then reduced with sodium borohydride to afford the hemiacetals **32**. The hemiacetals **32** and the Wittig reagent ($Ph_3P{=}CHCO_2Et$) were heated in toluene to afford the ethyl esters **33**, which were then hydrolyzed with potassium carbonate to afford the carboxylic acids **34**. In the presence of *N*-(3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride and HOBT (1-hydroxybenzotriazole hydrate), reaction with the appropriate 1-alkylpiperazine **35**

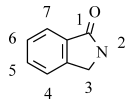


Fig. 1. Structure of Isoindolin-1-one Skeleton

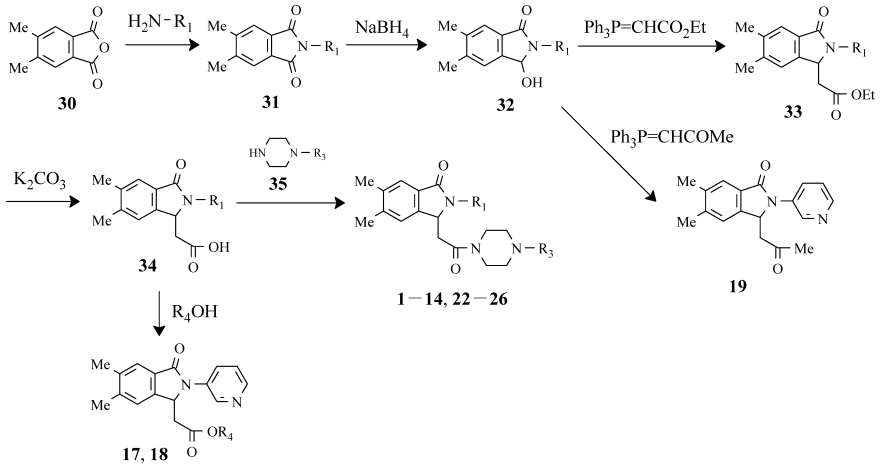


Chart 1

* To whom correspondence should be addressed. e-mail: norimasa_kanamitu@maruishi-pharm.co.jp

afforded the isoindolin-1-one derivatives **1**—**14**, **22**—**26**. In the presence of *N*-(3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride and 4-dimethylaminopyridine, carboxylic acid **34** was reacted with an appropriate alcohol to afford esters **17** and **18**. Heating of hemiacetal **32** with 1-(triphenylphosphoranylidene)-2-propanone afforded ketone **19**.

The starting material, phthalic anhydride **30** was synthesized in only a few steps and with high efficiency by the route shown in Chart 2. Specifically, the addition reaction of 2,3-dimethyl-1,3-butadiene **36** and maleic anhydride afforded acid anhydride **37**, which was then heated with bromine in acetic acid and converted into phthalic anhydride **30**.

The 1-alkylpiperazines **35** were synthesized based on the literature[9] as shown in Chart 3. Specifically, ethyl 1-piperazinecarboxylate **38** and an appropriate alkyl halide were heated in the presence of potassium carbonate to afford ethyl esters **39**, which were then heated with 47% hydrogen bromide to afford hydrobromides **40**, which were then treated with sodium hydroxide to afford the 1-alkylpiperazines **35**.

Ethers **20** and **21** were synthesized by the methods shown in Chart 4. Specifically, ester **33b** was reduced by heating with excess sodium borohydride to afford alcohol **41**, which was then reacted with methanesulfonyl chloride and transformed into methanesulfonyloxy **42**. Heating the methanesulfonyloxy **42** with sodium methoxide, prepared from methanol and sodium, or with the commercially available reagent sodium ethoxide afforded ethers **20** and **21**, respectively.

The 5,6-position cyclized compounds **27**—**29** were prepared using the phthalic anhydrides **43a**—**c** shown in Chart 5, and using the same method as shown in Chart 1. The phthalic anhydrides **43a**—**c** were synthesized as described below. First, 1,6-heptadiyne **44a**, 1,7-octadiyne **44b**, and propargyl ether **44c** were heated with diethyl acetylenedicarboxylate in the presence of dicarbonylcyclopentadienyl cobalt(I) to afford diesters **45a**—**c**, which were then treated with hydrochloric acid or potassium carbonate to afford dicarboxylic acids **46a**—**c**. Finally, heating them in acetic anhydride gave the desired phthalic anhydrides **43a**—**c**.

Synthesis of the enantiomers of **5** was achieved by the method described below. First, the racemic carboxylic acid **34** shown in Chart 1 was reacted with (*S*)-(−)-1-phenylethylamine to form the salt, which was then repeatedly recrystallized from methanol or ethanol. The obtained crystals were treated with diluted hydrochloric acid to obtain the (−) form of the carboxylic acid. This carboxylic acid **34**(−) was reacted, as in Chart 1, with 1-methylpiperazine to obtain the (−) form of **5**. The resolving agent (*R*)-(+)-1-phenylethylamine was used in the same way to obtain the (+) form of **5**. The (−) forms of **3**, **27**, and **47** (**JM-1232**) were prepared in a similar way.

Results and Discussion

We first synthesized isoindolin-1-one derivatives in racemic form. We introduced a phenyl group to the 2-position and chloro, methyl, and nitro groups to the 5-position of the isoindolin-1-one skeleton, and in order to increase the water solubility of the compounds as hydrochloride salts, we introduced a substituent at the 3-position in the form of a 2-(4-methyl-1-piperazinyl)-2-oxoethyl group (Fig. 2). When these compounds were intravenously administered to mice, loss of righting reflex was not obtained and sedation-like action (inhibition of locomotor activity) was observed. Additionally, since even when the chloro group at the 5-position

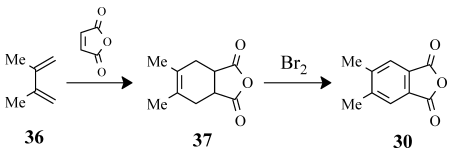


Chart 2

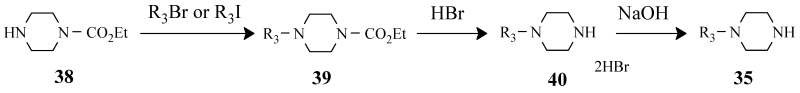
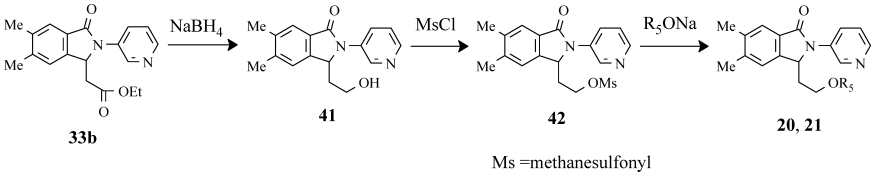


Chart 3



Ms =methanesulfonyl

Chart 4

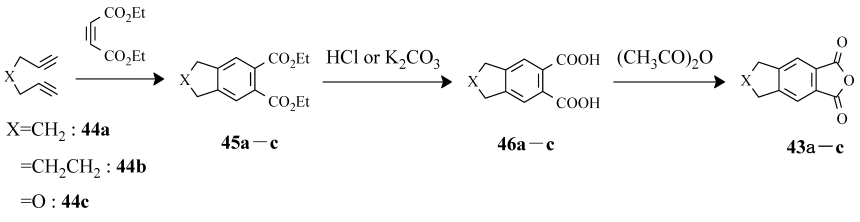


Chart 5

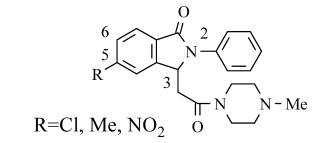


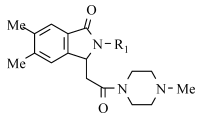
Fig. 2. Structure of 2-Phenyl-3-[2-(4-methyl-1-piperazinyl)-2-oxoethyl]-5-Chloro, Methyl and Nitroisoindolin-1-one

was changed to the 6-position, the same sedation-like action was observed, it was considered that the 5- and 6-position substituents exert some effect on the sedative-hypnotic activity. When chloro groups were introduced to both the 5,6-positions, while the activity was weaker, loss of righting reflex in mice was noted. To further increase the activity of this dichloro compound, the dichloro group was changed to the dimethyl, diethyl, and dimethoxy group, and the dimethyl compound **3** showed potent sedative-hypnotic activity (HD_{50}=9.37 mg/kg) (Table 1).

In order to increase the activity of **3** further, we next performed a series of synthetic modifications (Table 1). Introduction and fixation of a methyl group to the 5,6-positions, and a 2-(4-methyl-1-piperazinyl)-2-oxoethyl group to the 3-position, and introduction of a naphthyl or cyclohexyl to the 2-position in place of the phenyl group, did not show activity at 20 or 40 mg/kg. Thus, the 2-position was fixed as a phenyl group and various substituents were introduced to this phenyl. Initially, a fluorine atom was introduced to the *o*-, *m*-, and *p*-positions, separately, and the *m*-position substituted compound **5** (HD_{50}=6.92 mg/kg) had greater activity than non-substituted compound **3**. Since modification of the *m*-position showed a favorable tendency, other halogen atoms were introduced to the *m*-position, but activity was lower than for **3**. Furthermore, various electron-releasing and electron-withdrawing groups were introduced to this *m*-position, but these derivatives also had weaker activity. Moreover, the difluoro derivative **14** was synthesized by introduction of a fluorine atom to both *m*-positions, but activity was weaker than for the mono-substituted *m*-position derivative **5**. Therefore, it was judged that the most effective 2-position modification was an unsubstituted phenyl or a *m*-fluoro phenyl group.

Next, we attempted to optimize the 3-position substituent of the isoindolin-1-one skeleton. The combination of a 2-position hydrophobic group and a 3-position hydrophilic group was inverted. In other words, derivatives with hydrophilicity at the 2-position and hydrophobicity at the 3-position were synthesized (Table 2). In this case, a pyridine ring was selected for the 2-position hydrophilic group, and compounds **33b** and **16** were synthesized, and we examined which was the optimal 2-position substituent, a 3-pyridyl or a 4-pyridyl group. As a result, since the 3-pyridyl analog showed a hypnotic effect, the 2-position substituent was fixed as 3-pyridyl and various ester, ketone, and ether groups were introduced as the 3-position hydrophobic group (compounds **17**—**21**). However, no compounds with better activity than **5** and **3** were found. Therefore, since the combination of a 2-position hydrophobic group and a 3-position hydrophilic group gave the most favorable results, modification of the terminal methyl group of the 3-position substituent piperazine of **5** and **3** into a bulky substituent was next examined (Table 3). However, no compounds with stronger activity than **5** and **3**

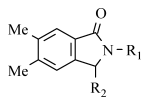
Table 1. Hypnotic Doses of Compounds **1**—**14**



Compd No.	R_1	HD_{50} (mg/kg)
1		N.D.[a]
2		N.D.[b]
3		9.37
4		N.D.[c]
5		6.92
6		14.72
7		15.72
8		16.17
9		17.04
10		15.31
11		28.82
12		31.18
13		21.47
14		10.33

a) Mean sleeping time was 0 s at 20 mg/kg. *b*) Mean sleeping time was 0 s at 40 mg/kg. *c*) Mean sleeping time was 10 s at 40 mg/kg.

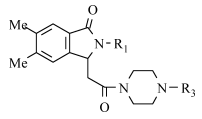
Table 2. Hypnotic Activity of Compounds **16**—**21** and **33b**



Compd No.	R_1	R_2	Hypnotic activity[a]
33b		$CH_2CO_2C_2H_5$	17 s (40 mg/kg)
16		$CH_2CO_2C_2H_5$	0 s (60 mg/kg)
17		$CH_2CO_2CH_3$	0 s (60 mg/kg)
18		$CH_2CO_2C_3H_7$	60 s (40 mg/kg)
19		$CH_2C(=O)CH_3$	0 s (60 mg/kg)
20		$CH_2CH_2OCH_3$	0 s (30 mg/kg), 50 s (50 mg/kg)
21		$CH_2CH_2OC_2H_5$	0 s (15 mg/kg), 35 s (30 mg/kg)

a) Mean sleeping time. ( ): injection dose.

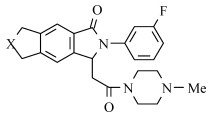
Table 3. Hypnotic Doses of Compounds **22**—**26**



Compd No.	R_1	R_3	HD_{50} (mg/kg)
22		n-C_3H_7	11.64
23		$CH(CH_3)_2$	N.D.[a]
24		$CH_2CH(C_2H_5)_2$	N.D.[b]
25			30.75
26			N.D.[c]

a) Mean sleeping times were 0 and 200 s at 15 and 20 mg/kg, respectively. *b*) Mean sleeping time was 0 s at 50 mg/kg. *c*) Mean sleeping time was 0 s at 40 mg/kg.

Table 4. Hypnotic Activity of Compounds **27**—**29**



Compd No.	X	Hypnotic activity[a]
27	CH_2	434 s
28	CH_2CH_2	0 s
29	O	88 s

a) Mean sleeping time at 7.5 mg/kg. HD_{50} was not calculated because of these compounds were racemic form.

were found.

Furthermore, we fixed the 2-position as *m*-fluoro phenyl and the 3-position as a 2-(4-methyl-1-piperazinyl)-2-oxoethyl group and modified the 5,6-position dimethyl group into a cyclized structure (Table 4). As a result, the five-membered ring compound **27** was shown to have good activity.

On the other hand, we performed optical resolution of **5**, which showed high activity in the racemic form and evaluated the activity and toxicity of the (+) and (−) isomers. As shown in Table 5, the activity of the (−) enantiomer **5**(−) was increased by more than 3-fold compared to that of the racemic form. Additionally, it showed low toxicity, with LD_{50}=64.69 mg/kg, and the therapeutic index (LD_{50}/HD_{50}) value of 34.05 was 10-times better than that for propofol (HD_{50}=11.88 mg/kg, LD_{50}=40.45 mg/kg, LD_{50}/HD_{50}=3.40).[10] Thus, since for **5**, the (−)-enantiomer showed a superior result, the (−)-enantiomer of **3** was also synthesized. As a result, since activity was more than 3 times better compared to the racemic form, the (−) forms of the 5,6-position cyclized compound **27** and the non-fluoro compound **47** (**JM-1232**) (Fig. 3) were also synthesized and their activity and toxicity were evaluated. As shown in Table 5, as well as maintenance of potent activity with **27**(−) and **47**(−) [**JM-1232**(−)], their toxicity was further reduced, with LD_{50}>120 mg/kg, compared to before cyclization, and the therapeutic indexes (LD_{50}/HD_{50}) were >55.30 and >38.46, respectively, which were good values.

Table 5. Hypnotic and Lethal Doses of Enantiomers

	HD_{50} (mg/kg)	LD_{50} (mg/kg)
5(−)	1.90	64.69
5(+)	25.13	>120
5	6.92	92.00
3(−)	2.35	88.67
3	9.37	>120
27(−)	2.17	>120
47(−)	3.12	>120

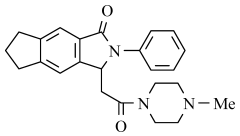


Fig. 3. Structure of **47** (**JM-1232**)

Conclusion

Synthetic studies on a series of isoindolin-1-one derivatives have led to the discovery of various compounds that showed sedative-hypnotic effects in mice after intravenous administration. Amongst them, **5** and **3** in racemic form showed strong activity (HD_{50}=6.92 mg/kg and HD_{50}=9.37 mg/kg, respectively). The hypnotic effects of **5** and **3** were enantioselective and the (−)-enantiomers **5**(−) and **3**(−) showed more potent activity (HD_{50}=1.90 mg/kg and HD_{50}=2.35 mg/kg, respectively). In addition, the 5,6-position cyclized derivatives **27**(−) and **47**(−) [**JM-1232**(−)] also showed and retained potent activity (HD_{50}=2.17 mg/kg and HD_{50}=3.12 mg/kg, respectively) and showed less toxicity in comparison to the 5,6-position dimethyl derivative. In general observation, these 4 compounds did not have the direct action on respiratory rate, but showed the tendency to decrease it in response to the depth of the sedative-hypnotic state. And as a result of the general pharmacological study done afterwards, **47**(−) [**JM-1232**(−)] was selected as a candidate for the clinical trial.

Experimental

^{1}H-NMR spectra were obtained by a Lambda 400 MHz spectrometer (JEOL Ltd., Tokyo, Japan) using Me_4Si as the internal standard. Mass spectral data were obtained on a ZQ2000 mass spectrometry (Waters, Tokyo, Japan). Silica gel 60 (Merck Ltd., Tokyo, Japan, 230—400 mesh) was used in the column chromatography.

5,6-Dimethyl-2-benzofuran-1,3-dione (30) To a solution of maleic anhydride (5.4 g, 55 mmol) in benzene (50 ml), 2,3-dimethyl-1,3-butadiene **36** (6.3 ml, 55 mmol) was added dropwise and the mixture was then stirred at 25 °C overnight. After filtration to remove insoluble material, the filtrate was concentrated under reduced pressure to afford 5,6-dimethyl-3a,4,7,7a-tetrahydro-2-benzofuran-1,3-dione **37** (9.5 g, 96%). To a solution of the obtained **37** (9.5 g, 53 mmol) in CH_3COOH (28 ml) at 115 °C, a solution of bromine (6.1 ml, 0.12 mol) in CH_3COOH (28 ml) was added dropwise over 45 min and the solution was then heated under reflux for 1 h. After allowing the reaction solution to stand overnight, the precipitate was collected by filtration, washed with diethyl ether and dried to afford **30** (3.5 g, 37%). ^{1}H-NMR (DMSO-d_6) δ: 2.43 (6H, s), 7.87 (2H, s).

6,7-Dihydro-1*H*-indeno[5,6-*c*]furan-1,3(5*H*)-dione (43a) To a solution of 1,6-heptadiyne **44a** (0.72 ml, 6.3 mmol) in xylene (5 ml), diethyl acetylenedicarboxylate (1.0 ml, 6.3 mmol) and dicarbonylcyclopentadienyl cobalt(I) (0.1 ml, 0.62 mmol) were added and the mixture was then stirred for 5 d at 80 °C. Dilute hydrochloric acid was added to the reaction mixture

and it was then extracted with EtOAc and the organic layer was washed with saturated NaCl solution, dried with anhydrous Na_2SO_4, and concentrated under reduced pressure. The resulting residue was chromatographed on a silica gel column (elution with $CHCl_3$ followed by hexane/EtOAc=10/1) to give diethyl 5,6-indanedicarboxylate **45a** (0.36 g, 22%). To a solution of the obtained **45a** (0.36 g, 1.4 mmol) in CH_3COOH (0.8 ml), concentrated hydrochloric acid (0.4 ml) was added and the mixture was stirred at 80 °C overnight. Ice-water was added to the reaction mixture and the resulting precipitate was collected by filtration, washed with water, and dried to afford 5,6-indanedicarboxylic acid **46a** (0.28 g, 97%). The obtained **46a** (0.28 g, 1.4 mmol) in acetic anhydride (6.7 ml) was heated under reflux overnight. The reaction mixture was poured into ice-water and the precipitate was collected by filtration, washed with water, and dried to afford **43a** (0.25 g, 95%). ^{1}H-NMR ($CDCl_3$) δ: 2.23 (2H, quin, J=7.5 Hz), 3.08 (4H, t, J=7.5 Hz), 7.79 (2H, s).

5,6,7,8-Tetrahydronaphtho[2,3-*c*]furan-1,3-dione (43b) Using 1,7-octadiyne **44b**, it was obtained in a similar manner as that for **43a**. ^{1}H-NMR ($CDCl_3$) δ: 1.86—1.89 (4H, m), 2.94—2.97 (4H, m), 7.68 (2H, s).

5,7-Dihydro-1*H*,3*H*-furo[3,4-*f*][2]benzofuran-1,3-dione (43c) Using propargyl ether **44c**, it was obtained in a similar manner as that for **43a**. ^{1}H-NMR ($CDCl_3$) δ: 5.25 (4H, s), 7.85 (2H, s).

In Chart 3, when R_3 is 2-ethylbutyl, the 1-alkylpiperazine **35** is **35a**.

1-(2-Ethylbutyl)piperazine (35a) A mixture of ethyl 1-piperazinecarboxylate **38** (1.50 g, 9.48 mmol), 1-bromo-2-ethylbutane (1.59 g, 9.48 mmol), and K_2CO_3 (1.57 g, 11.38 mmol) in CH_3CN (30 ml) was stirred for 24 h at 90 °C under an argon atmosphere. The reaction mixture was then concentrated under reduced pressure. Water was added to the residue, followed by extraction with $CHCl_3$, drying with anhydrous Na_2SO_4, and concentration under reduced pressure. The resulting residue was chromatographed on a silica gel column ($CHCl_3$/MeOH=50/1) to give ethyl 4-(2-ethylbutyl)-1-piperazinecarboxylate **39a** (1.90 g, 83%). The obtained **39a** (1.90 g, 7.84 mmol) in 47% HBr (51 ml) was stirred for 3 h at 110 °C. The reaction mixture was then concentrated under reduced pressure and the obtained crystals were then washed with diethyl ether, collected by filtration, and dried to afford 1-(2-ethylbutyl)piperazine dihydrobromide **40a** (1.87 g, 72%). The obtained **40a** (1.87 g, 4.61 mmol) was dissolved in H_2O (6 ml) and 1N-NaOH was added to adjust to pH 8. MeOH (50 ml) was added to it and insoluble material was removed by filtration before the filtrate was concentrated under reduced pressure. The residue was dissolved in $CHCl_3$ and insoluble material was removed by filtration before the filtrate was concentrated under reduced pressure and dried to afford **35a** (1.05 g). ^{1}H-NMR ($CDCl_3$) δ: 0.84 (6H, t, J=7.3 Hz), 1.23—1.40 (5H, m), 2.24 (2H, d, J=6.6 Hz), 2.74—2.76 (4H, m), 3.24—3.26 (4H, m).

1-Cycloheptylpiperazine Using ethyl 1-piperazinecarboxylate **38** and 1-bromocycloheptane, it was obtained in a similar manner as that for **35a**. ^{1}H-NMR ($CDCl_3$) δ: 1.38—1.57 (10H, m), 1.67—1.89 (2H, m), 2.71 (1H, br s), 2.91—2.93 (4H, m), 3.22—3.27 (4H, m).

In Chart 1, when R_1 is 3-fluorophenyl, the carboxylic acid **34** is **34a**.

2-[2-(3-Fluorophenyl)-5,6-dimethyl-3-oxo-2,3-dihydro-1*H*-isoindol-1-yl]acetic Acid (34a) A mixture of 5,6-dimethyl-2-benzofuran-1,3-dione **30** (4.0 g, 22.7 mmol) and 3-fluoroaniline (2.5 g, 22.7 mmol) in CH_3COOH (70 ml) was heated under reflux for 2 h and allowed to cool. The precipitate was then collected by filtration, washed with petroleum ether, and dried to afford 2-(3-fluorophenyl)-5,6-dimethyl-1*H*-isoindole-1,3(2*H*)-dione **31a** (4.55 g, 74%). The obtained **31a** (4.55 g, 16.9 mmol) was suspended in MeOH (70 ml) and tetrahydrofuran (70 ml) and under ice-cooling, $NaBH_4$ (1.28 g, 33.8 mmol) was added portionwise and stirring continued for 20 min at the same temperature. After water was added to the reaction mixture, the precipitate was collected by filtration, washed with water, and dried to afford 2-(3-fluorophenyl)-3-hydroxy-5,6-dimethylisoindolin-1-one **32a** (3.25 g, 71%). The obtained **32a** (3.25 g, 12 mmol) and ethyl 2-(triphenylphosphoranylidene)acetate (4.88 g, 14 mmol) in toluene (80 ml) was heated under reflux for 3.5 h under an argon atmosphere and the reaction mixture was then concentrated under reduced pressure. After the resulting residue was dissolved in $CHCl_3$ and passed through silica gel, the unreacted ethyl 2-(triphenylphosphoranylidene) acetate was removed by absorption to the silica gel and the filtrate was concentrated under reduced pressure. A solution of the crude residue in MeOH (40 ml) and 15% aqueous K_2CO_3 (11 ml) was stirred at 80 °C for 4 h and concentrated under reduced pressure. H_2O was added to the residue, followed by extraction with diethyl ether. The aqueous layer was acidified with concentrated hydrochloric acid and the precipitate was collected by filtration, washed with water, and dried to afford **34a** (2.36 g, 63%).

2-(3-Fluorophenyl)-5,6-dimethyl-3-[2-(4-methyl-1-piperazinyl)-2-oxoethyl]isoindolin-1-one (5) **34a** (0.5 g, 1.6 mmol), 1-methylpiperazine (0.16 g, 1.6 mmol), *N*-(3-dimethylaminopropyl)-*N'*-ethylcarbodiimide hydrochloride (0.31 g, 1.6 mmol) and 1-hydroxybenzotriazole hydrate (0.25 g, 1.6 mmol) in tetrahydrofuran (40 ml) were stirred at 25 °C for 16 h, and the reaction mixture then concentrated under reduced pressure. The resulting residue was chromatographed on a silica gel column ($CHCl_3$/MeOH=20/1) to give **5** (0.56 g, 89%). mp 141—141.5 °C (hydrochloride salt); ESI-MS m/z: 396.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.21—2.23 (2H, m), 2.27 (3H, s), 2.36 (3H, s), 2.37 (3H, s), 2.39—2.44 (3H, m), 2.90 (1H, dd, J=3.2, 16.0 Hz), 3.20—3.31 (2H, m), 3.64—3.77 (2H, m), 5.75—5.79 (1H, m), 6.88—6.93 (1H, m), 7.37—7.42 (3H, m), 7.58—7.61 (1H, m), 7.68 (1H, s).

Using 5,6-dimethyl-2-benzofuran-1,3-dione **30** and an appropriate amine, carboxylic acid **34** was obtained in a similar manner as that for **34a**, and then using the appropriate 1-alkylpiperazine, compound **1**—**4**, **6**—**14**, **22**—**26** were obtained in a similar manner as that for **5**.

1: mp 197—201 °C (hydrochloride salt); ESI-MS m/z: 428.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.06—2.31 (7H, m), 2.39 (3H, s), 2.40 (3H, s), 2.50—2.81 (2H, m), 2.90—3.60 (4H, m), 5.61—5.81 (1H, m), 7.41—7.93 (9H, m).

2: mp 147—150 °C (hydrochloride salt); ESI-MS m/z: 384.4 $[M+H]^+$; ^{1}H-NMR (CD_3OD) δ: 1.14—1.43 (4H, m), 1.67—1.97 (6H, m), 2.08—2.48 (4H, m), 2.29 (3H, s), 2.32 (3H, s), 2.33 (3H, s), 2.63 (1H, dd, J=8.6, 16.0 Hz), 3.10 (1H, dd, J=4.4, 16.0 Hz), 3.43—3.75 (5H, m), 5.07—5.10 (1H, m), 7.22 (1H, s), 7.45 (1H, s).

3: mp 124—132 °C (hydrochloride salt); ESI-MS m/z: 378.4 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.18—2.21 (2H, m), 2.26 (3H, s), 2.36 (3H, s), 2.37 (3H, s), 2.38—2.43 (3H, m), 2.88 (1H, dd, J=3.7, 16.0 Hz), 3.17—3.29 (2H, m), 3.60—3.76 (2H, m), 5.78—5.82 (1H, m), 7.19—7.23 (1H, m), 7.37 (1H, s), 7.41—7.46 (2H, m), 7.64—7.68 (3H, m).

4: mp 151—153 °C (hydrochloride salt); ESI-MS m/z: 396.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.27 (13H, m), 2.49 (1H, dd, J=8.5, 15.6 Hz), 2.70 (1H, dd, J=4.6, 15.6 Hz), 3.27 (2H, br s), 3.49—3.73 (2H, m), 5.66—5.70 (1H, m), 7.18—7.34 (3H, m), 7.37 (1H, s), 7.42—7.46 (1H, m), 7.70 (1H, s).

6: mp 146—151 °C (hydrochloride salt); ESI-MS m/z: 396.1 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.19—2.22 (2H, m), 2.26 (3H, s), 2.34—2.43 (3H, m), 2.36 (3H, s), 2.37 (3H, s), 2.81 (1H, dd, J=3.7, 16.0 Hz), 3.21—3.27 (2H, m), 3.61—3.73 (2H, m), 5.73—5.76 (1H, m), 7.10—7.15 (2H, m), 7.36 (1H, s), 7.56—7.61 (2H, m), 7.67 (1H, s).

7: mp 138—144.5 °C (hydrochloride salt); ESI-MS m/z: 412.2, 414.1 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.22 (2H, t, J=5.1 Hz), 2.27 (3H, s), 2.36—2.44 (9H, m), 2.87 (1H, dd, J=3.6, 15.8 Hz), 3.20—3.30 (2H, m), 3.66—3.74 (2H, m), 5.75—5.78 (1H, m), 7.17—7.20 (1H, m), 7.35 (1H, d, J=8.0 Hz), 7.38 (1H, s), 7.48—7.52 (1H, m), 7.67 (1H, s), 7.78 (1H, t, J=1.9 Hz).

8: mp 105—105.5 °C (hydrochloride salt); ESI-MS m/z: 456.1, 458.1 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.22 (2H, t, J=5.1 Hz), 2.27 (3H, s), 2.36—2.44 (9H, m), 2.86 (1H, dd, J=3.7, 15.9 Hz), 3.21—3.31 (2H, m), 3.65—3.75 (2H, m), 5.74—5.78 (1H, m), 7.28—7.35 (2H, m), 7.38 (1H, s), 7.52—7.55 (1H, m), 7.66 (1H, s), 7.93 (1H, t, J=2.0 Hz).

9: mp 136—141.5 °C (hydrochloride salt); ESI-MS m/z: 392.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.16—2.23 (2H, m), 2.26 (3H, s), 2.34—2.39 (3H, m), 2.36 (3H, s), 2.37 (3H, s), 2.40 (3H, s), 2.87 (1H, dd, J=3.6, 16.0 Hz), 3.16—3.31 (2H, m), 3.59—3.79 (2H, m), 5.74—5.78 (1H, m), 7.03 (1H, d, J=7.6 Hz), 7.29—7.39 (3H, m), 7.50 (1H, s), 7.67 (1H, s).

10: mp 140—146 °C (hydrochloride salt); ESI-MS m/z: 408.3 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.19—2.22 (2H, m), 2.26 (3H, s), 2.35—2.42 (3H, m), 2.36 (3H, s), 2.37 (3H, s), 2.92 (1H, dd, J=3.4, 16.0 Hz), 3.18—3.30 (2H, m), 3.63—3.75 (2H, m), 3.85 (3H, s), 5.75—5.78 (1H, m), 6.77 (1H, dd, J=2.4, 8.4 Hz), 7.13 (1H, d, J=8.0 Hz), 7.35 (1H, dd, J=8.0, 8.0 Hz), 7.38—7.39 (2H, m), 7.67 (1H, s).

11: mp 158.5—162.5 °C (hydrochloride salt); ESI-MS m/z: 421.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.18—2.21 (2H, m), 2.26 (3H, s), 2.34—2.41 (3H, m), 2.36 (3H, s), 2.37 (3H, s), 2.94 (1H, dd, J=3.4, 16.0 Hz), 2.99 (6H, s), 3.17—3.31 (2H, m), 3.61—3.76 (2H, m), 5.74—5.77 (1H, m), 6.59 (1H, dd, J=2.4, 8.0 Hz), 6.82 (1H, d, J=7.6 Hz), 7.17 (1H, dd, J=2.2, 2.2 Hz), 7.25—7.29 (1H, m), 7.37 (1H, s), 7.67 (1H, s).

12: mp 164.5—166.5 °C (hydrochloride salt); ESI-MS m/z: 446.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.15—2.22 (2H, m), 2.26 (3H, s), 2.32—2.47 (3H, m), 2.37 (3H, s), 2.38 (3H, s), 2.85 (1H, dd, J=3.6, 16.0 Hz), 3.19—3.30 (2H, m), 3.64—3.72 (2H, m), 5.82—5.85 (1H, m), 7.39 (1H, s), 7.46 (1H, d, J=7.8 Hz), 7.56 (1H, dd, J=7.8, 7.8 Hz), 7.68 (1H, s), 7.78 (1H, d, J=8.3 Hz), 8.06 (1H, s).

13: mp 172—175.5 °C (hydrochloride salt); ESI-MS m/z: 423.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.20—2.25 (2H, m), 2.27 (3H, s), 2.36—2.41 (2H, m),

2.38 (3H, s), 2.39 (3H, s), 2.47 (1H, dd, J=9.3, 16.0 Hz), 2.85 (1H, dd, J=3.4, 16.0 Hz), 3.21—3.32 (2H, m), 3.62—3.75 (2H, m), 5.84—5.88 (1H, m), 7.40 (1H, s), 7.61 (1H, dd, J=8.2, 8.2 Hz), 7.69 (1H, s), 7.99 (1H, dd, J=8.2, 8.2 Hz), 7.97—8.07 (2H, m), 8.65 (1H, dd, J=2.1, 2.1 Hz).

14: mp 145—146 °C (hydrochloride salt); ESI-MS m/z: 414.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.23—2.26 (2H, m), 2.28 (3H, s), 2.36 (3H, s), 2.37 (3H, s), 2.40—2.44 (3H, m), 2.93 (1H, dd, J=2.6, 16.0 Hz), 3.22—3.34 (2H, m), 3.66—3.79 (2H, m), 5.70—5.73 (1H, m), 6.62—6.67 (1H, m), 7.31—7.36 (2H, m), 7.38 (1H, s), 7.67 (1H, s).

22: mp 137—138 °C (hydrochloride salt); ESI-MS m/z: 406.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 0.89 (3H, t, J=7.3 Hz), 1.49 (2H, sext, J=7.3 Hz), 2.22—2.46 (7H, m), 2.36 (3H, s), 2.37 (3H, s), 2.87 (1H, dd, J=3.4, 16.0 Hz), 3.17—3.34 (2H, m), 3.60—3.78 (2H, m), 5.78—5.82 (1H, m), 7.19—7.23 (1H, m), 7.37 (1H, s), 7.42—7.46 (2H, m), 7.64—7.68 (3H, m).

23: mp 79—84 °C; ESI-MS m/z: 406.3 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 1.00 (6H, d, J=6.5 Hz), 2.29—2.32 (2H, m), 2.36 (3H, s), 2.37 (3H, s), 2.38—2.43 (1H, m), 2.46—2.49 (2H, m), 2.67 (1H, sept, J=6.5 Hz), 2.87 (1H, dd, J=3.6, 16.0 Hz), 3.17—3.30 (2H, m), 3.61—3.73 (2H, m), 5.79—5.82 (1H, m), 7.19—7.25 (1H, m), 7.38 (1H, s), 7.42—7.46 (2H, m), 7.64—7.68 (3H, m).

24: mp 112.5—119 °C (hydrochloride salt); ESI-MS m/z: 466.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 0.83 (6H, t, J=7.3 Hz), 1.22—1.41 (5H, m), 2.11—2.21 (4H, m), 2.36—2.42 (9H, m), 2.90 (1H, dd, J=3.3, 16.0 Hz), 3.19—3.28 (2H, m), 3.62—3.74 (2H, m), 5.76—5.79 (1H, m), 6.88—6.93 (1H, m), 7.37—7.42 (3H, m), 7.59—7.63 (1H, m), 7.67 (1H, s).

25: mp 142.5—144 °C (hydrochloride salt); ESI-MS m/z: 464.3 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 1.01—1.30 (5H, m), 1.60—1.63 (1H, m), 1.77—1.80 (4H, m), 2.22—2.27 (1H, m), 2.36 (6H, s), 2.36—2.43 (3H, m), 2.49—2.58 (2H, m), 2.89 (1H, dd, J=3.7, 16.0 Hz), 3.17—3.29 (2H, m), 3.62—3.73 (2H, m), 5.75—5.79 (1H, m), 6.87—6.94 (1H, m), 7.37—7.42 (3H, m), 7.58—7.63 (1H, m), 7.67 (1H, s).

26: mp 135—142 °C (hydrochloride salt); ESI-MS m/z: 478.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 1.33—1.88 (12H, m), 2.25—2.60 (6H, m), 2.36 (3H, s), 2.37 (3H, s), 2.90 (1H, dd, J=3.2, 16.0 Hz), 3.23 (2H, br s), 3.67 (2H, br s), 5.75—5.78 (1H, m), 6.88—6.93 (1H, m), 7.35—7.42 (3H, m), 7.60 (1H, d, J=10.8 Hz), 7.67 (1H, s).

Using **43a**, **43b**, and **43c**, compounds **27**, **28**, and **29**, respectively, were obtained in a similar manner as that for **5**.

27: mp 182—184 °C; ESI-MS m/z: 408.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.14 (2H, quin, J=7.5 Hz), 2.21—2.26 (5H, m), 2.37—2.45 (3H, m), 2.96 (1H, dd, J=3.3, 16.0 Hz), 2.98 (4H, t, J=7.5 Hz), 3.21—3.31 (2H, m), 3.64—3.74 (2H, m), 5.76—5.79 (1H, m), 6.87—6.92 (1H, m), 7.35—7.41 (2H, m), 7.44 (1H, s), 7.59 (1H, dt, J=10.9, 2.0 Hz), 7.71 (1H, s).

28: mp 172—175 °C; ESI-MS m/z: 422.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 1.82—1.84 (4H, m), 2.19—2.27 (5H, m), 2.34—2.45 (3H, m), 2.87—2.93 (5H, m), 3.20—3.31 (2H, m), 3.63—3.76 (2H, m), 5.75—5.78 (1H, m), 6.87—6.92 (1H, m), 7.29 (1H, s), 7.35—7.41 (2H, m), 7.58—7.61 (1H, m), 7.59 (1H, s).

29: mp 185—187 °C; ESI-MS m/z: 410.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.24—2.28 (5H, m), 2.39—2.45 (3H, m), 2.95 (1H, dd, J=3.1, 16.2 Hz), 3.22—3.31 (2H, m), 3.69—3.71 (2H, m), 5.17 (4H, s), 5.81—5.84 (1H, m), 6.93 (1H, td, J=7.8, 2.4 Hz), 7.35—7.44 (2H, m), 7.52 (1H, s), 7.58 (1H, dt, J=10.6, 2.3 Hz), 7.74 (1H, s).

In Chart 1, when R_1 is 3-pyridinyl, the hemiacetal **32** is **32b**.

3-Hydroxy-5,6-dimethyl-2-(3-pyridinyl)isoindolin-1-one (32b) 5,6-Dimethyl-2-benzofuran-1,3-dione **30** (2.0 g, 11 mmol) and 3-aminopyridine (1.0 g, 11 mmol) in CH_3COOH (30 ml) were heated under reflux for 1.5 h. After cooling, H_2O was added and the precipitate was collected by filtration, washed with water, and dried to afford 5,6-dimethyl-2-(3-pyridinyl)-1*H*-isoindole-1,3(2*H*)-dione **31b** (2.3 g, 83%). The obtained **31b** (0.5 g, 2.0 mmol) was suspended in MeOH (10 ml) and tetrahydrofuran (10 ml), and under ice-cooling $NaBH_4$ (75 mg, 2.0 mmol) was added portionwise then stirred at the same temperature for 30 min. To the reaction mixture, H_2O was added then the precipitate was collected by filtration, washed with water, and dried to afford **32b** (0.4 g, 79%). ^{1}H-NMR ($CDCl_3$) δ: 2.39 (3H, s), 2.42 (3H, s), 6.36 (1H, s), 7.43 (2H, dd, J=4.6, 8.2 Hz), 7.45 (1H, s), 7.62 (1H, s), 8.28—8.31 (1H, m), 8.36 (1H, d, J=3.6 Hz), 8.97 (1H, d, J=2.0 Hz).

In Chart 1, when R_1 is 3-pyridinyl, the ethyl ester **33** is **33b**.

Ethyl 2-[5,6-Dimethyl-3-oxo-2-(3-pyridinyl)-2,3-dihydro-1*H*-isoindol-1-yl]acetate (33b) A solution of **32b** (0.4 g, 1.6 mmol) and ethyl 2-(triphenylphosphoranylidene)acetate (0.66 g, 1.9 mmol) in toluene (10 ml) was heated under reflux for 4 h under an argon atmosphere and the reaction mixture was then concentrated under reduced pressure. The resulting residue was chromatographed on a silica gel column ($CHCl_3$/acetone=5/1) to give **33b** (0.37 g, 71%). mp 125.5—126.5 °C; ESI-MS m/z: 325.4 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 1.19 (3H, t, J=7.3 Hz), 2.37 (3H, s), 2.39 (3H, s), 2.54 (1H, dd, J=8.1, 16.1 Hz), 2.91 (1H, dd, J=3.9, 16.1 Hz), 4.06—4.12 (2H, m), 5.56—5.59 (1H, m), 7.31 (1H, s), 7.40 (1H, dd, J=4.9, 8.3 Hz), 7.69 (1H, s), 8.08—8.11 (1H, m), 8.48 (1H, dd, J=1.4, 4.6 Hz), 8.78 (1H, d, J=2.7 Hz).

Ethyl 2-[5,6-Dimethyl-3-oxo-2-(4-pyridinyl)-2,3-dihydro-1*H*-isoindol-1-yl]acetate (16) Using 5,6-dimethyl-2-benzofuran-1,3-dione **30** and 4-aminopyridine, it was obtained in a similar manner as that for **33b**. mp 140—144.5 °C; ESI-MS m/z: 325.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 1.24 (3H, t, J=7.1 Hz), 2.37 (3H, s), 2.39 (3H, s), 2.51 (1H, dd, J=9.0, 16.0 Hz), 3.05 (1H, dd, J=3.2, 16.0 Hz), 4.11—4.24 (2H, m), 5.54—5.57 (1H, m), 7.29 (1H, s), 7.68—7.69 (3H, m), 8.63 (2H, d, J=6.1 Hz).

In Chart 1, when R_1 is 3-pyridinyl, the carboxylic acid **34** is **34b**.

2-[5,6-Dimethyl-3-oxo-2-(3-pyridinyl)-2,3-dihydro-1*H*-isoindol-1-yl]acetic Acid (34b) A solution of **33b** (0.20 g, 0.59 mmol) in MeOH (1.5 ml) and 15% aqueous K_2CO_3 (0.46 ml) was stirred at 75 °C for 4 h. The reaction mixture was concentrated under reduced pressure and H_2O was added to the residue before the mixture was extracted with diethyl ether. The aqueous layer was acidified with concentrated hydrochloric acid and the resulting precipitate was collected by filtration, washed with water, and dried to afford **34b** (0.12 g, 69%). ^{1}H-NMR (DMSO-d_6) δ: 2.34 (3H, s), 2.36 (3H, s), 2.61 (1H, dd, J=6.8, 16.4 Hz), 2.86 (1H, dd, J=4.2, 16.4 Hz), 5.67—5.70 (1H, m), 7.49—7.63 (3H, m), 8.00—8.03 (1H, m), 8.44 (1H, dd, J=1.5, 4.6 Hz), 8.84 (1H, d, J=2.2 Hz), 12.34 (1H, br s).

Propyl 2-[5,6-Dimethyl-3-oxo-2-(3-pyridinyl)-2,3-dihydro-1*H*-isoindol-1-yl]acetate (18) **34b** (74 mg, 0.25 mmol), 1-propanol (16 mg, 0.27 mmol), and 4-dimethylaminopyridine (3 mg, 0.025 mmol) were dissolved in dichloromethane, and at 5 °C, *N*-(3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (53 mg, 0.27 mmol) was added then the solution was warmed to 25 °C over 1.5 h. The reaction mixture was concentrated under reduced pressure and H_2O was added to the residue, which was then extracted with EtOAc. The organic layer was washed with saturated aqueous $NaHCO_3$ and water, then dried over anhydrous Na_2SO_4 and concentrated under reduced pressure to afford **18** (34 mg, 40%). mp 123—127 °C; ESI-MS m/z: 339.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 0.88 (3H, t, J=7.6 Hz), 1.53—1.62 (2H, m), 2.37 (3H, s), 2.39 (3H, s), 2.55 (1H, dd, J=8.2, 16.0 Hz), 2.93 (1H, dd, J=4.1, 16.1 Hz), 3.96—4.04 (2H, m), 5.57—5.60 (1H, m), 7.30 (1H, s), 7.39 (1H, dd, J=4.9, 8.3 Hz), 7.69 (1H, s), 8.09—8.12 (1H, m), 8.48 (1H, dd, J=1.3, 4.9 Hz), 8.79 (1H, d, J=2.4 Hz).

Methyl 2-[5,6-Dimethyl-3-oxo-2-(3-pyridinyl)-2,3-dihydro-1*H*-isoindol-1-yl]acetate (17) Using **34b** and methanol, it was obtained in a similar manner as that for **18**. mp 162.5—169.5 °C (hydrochloride salt); ESI-MS m/z: 311.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.37 (3H, s), 2.39 (3H, s), 2.55 (1H, dd, J=8.3, 16.4 Hz), 2.92 (1H, dd, J=4.2, 16.4 Hz), 3.65 (3H, s), 5.56—5.59 (1H, m), 7.29 (1H, s), 7.40 (1H, dd, J=4.6, 8.3 Hz), 7.69 (1H, s), 8.06—8.11 (1H, m), 8.48 (1H, dd, J=1.3, 4.6 Hz), 8.79 (1H, d, J=2.7 Hz).

5,6-Dimethyl-3-(2-oxopropyl)-2-(3-pyridinyl)isoindolin-1-one (19) **32b** (150 mg, 0.59 mmol) and acetylmethylene triphenylphosphorane (188 mg, 0.59 mmol) in toluene (12 ml) were heated under reflux for 24 h under an argon atmosphere and the reaction mixture was then concentrated under reduced pressure. The resulting residue was chromatographed on a silica gel column ($CHCl_3$/MeOH=25/1) to give **19** (20 mg, 12%). mp 141.5—144 °C; ESI-MS m/z: 295.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.14 (3H, s), 2.37 (6H, s), 2.64 (1H, dd, J=9.5, 16.0 Hz), 3.04 (1H, dd, J=2.9, 16.0 Hz), 5.68—5.71 (1H, m), 7.24 (1H, s), 7.39 (1H, dd, J=4.6, 8.3 Hz), 7.68 (1H, s), 8.10—8.13 (1H, m), 8.47 (1H, d, J=4.1 Hz), 8.77 (1H, d, J=1.7 Hz).

2-[5,6-Dimethyl-3-oxo-2-(3-pyridinyl)-2,3-dihydro-1*H*-isoindol-1-yl]ethyl Methanesulfonate (42) To a solution of **33b** (8.4 g, 26 mmol) in MeOH (250 ml), $NaBH_4$ (11 g, 0.52 mol) was added portionwise, followed by stirring at 80 °C for 3 h. Ice-water was added to the reaction mixture and the resulting precipitate was collected by filtration, washed with water, and dried to afford 3-(2-hydroxyethyl)-5,6-dimethyl-2-(3-pyridinyl)isoindolin-1-one **41** (6.0 g, 82%). After the obtained **41** (5.5 g, 20 mmol) was dissolved in dichloromethane (140 ml), triethylamine (5.4 ml, 29 mmol) and methanesulfonyl chloride (2.4 ml, 21 mmol) were added to it and the mixture was stirred at 25 °C for 2 h then concentrated under reduced pressure. The resulting residue was chromatographed on a silica gel column ($CHCl_3$/MeOH=20/1) to give **42** (5.5 g, 76%). ^{1}H-NMR ($CDCl_3$) δ: 2.31—2.50 (2H, m), 2.38 (3H, s), 2.42 (3H, s), 2.80 (3H, s), 3.88—3.94 (1H, m), 4.02—4.08 (1H, m), 5.41—5.43 (1H, m), 7.33 (1H, s), 7.42 (1H, dd, J=4.6, 8.3 Hz), 7.71 (1H, s), 8.14 (1H, d, J=8.3 Hz), 8.49 (1H, d, J=4.8 Hz), 8.80 (1H, s).

3-(2-Ethoxyethyl)-5,6-dimethyl-2-(3-pyridinyl)isoindolin-1-one (21) NaOEt (38 mg, 0.55 mmol) and **42** (100 mg, 0.28 mmol) in EtOH (15 ml)

were stirred at 75 °C for 3 h. After adding H_2O to the reaction mixture, it was extracted with $CHCl_3$ and the organic layer was concentrated under reduced pressure. The resulting residue was chromatographed on a silica gel column ($CHCl_3$/EtOAc=1/1) to give **21** (42 mg, 48%). mp 131.5—135 °C; ESI-MS *m/z*: 311.2 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, t, *J*=7.0 Hz), 2.03—2.12 (1H, m), 2.17—2.25 (1H, m), 2.37 (3H, s), 2.40 (3H, s), 3.17—3.32 (4H, m), 5.37—5.39 (1H, m), 7.31 (1H, s), 7.38 (1H, dd, *J*=4.6, 8.2 Hz), 7.68 (1H, s), 8.12 (1H, d, *J*=8.2 Hz), 8.46 (1H, d, *J*=4.0 Hz), 8.83 (1H, s).

3-(2-Methoxyethyl)-5,6-dimethyl-2-(3-pyridinyl)isoindolin-1-one (20) mp 200—202 °C (hydrochloride salt); ESI-MS *m/z*: 297.4 $[M+H]^+$; ^{1}H-NMR ($CDCl_3$) δ: 2.01—2.09 (1H, m), 2.18—2.26 (1H, m), 2.38 (3H, s), 2.41 (3H, s), 3.13—3.28 (2H, m), 3.18 (3H, s), 5.36—5.39 (1H, m), 7.30 (1H, s), 7.38 (1H, dd, *J*=4.6, 8.2 Hz), 7.69 (1H, s), 8.12 (1H, d, *J*=8.3 Hz), 8.46 (1H, d, *J*=4.4 Hz).

(−)-2-[2-(3-Fluorophenyl)-5,6-dimethyl-3-oxo-2,3-dihydro-1*H*-isoindol-1-yl]acetic Acid (34a(−)) **34a** (10.77 g, 34 mmol) was dissolved in a mixture of $CHCl_3$ and MeOH (1 : 1) and to this solution, a solution of (*S*)-(−)-1-phenylethylamine (4.12 g, 34 mmol) in CH_3OH was added and the mixture was stirred at ambient temperature for 30 min then concentrated under reduced pressure. The resulting crystals were repeatedly recrystallized from MeOH and then dissolved in H_2O and acidified with 1 N-HCl. The precipitate was collected by filtration, washed with water, and dried to afford **34a**(−) (385 mg, 4%). $[\alpha]_D^{29}$ −61.6° (*c*=1.0, $CHCl_3$: MeOH=1 : 1).

(+)-2-[2-(3-Fluorophenyl)-5,6-dimethyl-3-oxo-2,3-dihydro-1*H*-isoindol-1-yl]acetic Acid (34a(+)) Using **34a** and (*R*)-(+)-1-phenylethylamine, it was obtained in a similar manner as that for **34a**(−). $[\alpha]_D^{28}$ +57.6° (*c*=1.0, $CHCl_3$: MeOH=1 : 1).

(−)-2-(3-Fluorophenyl)-5,6-dimethyl-3-[2-(4-methyl-1-piperazinyl)-2-oxoethyl]isoindolin-1-one (5(−)) A mixture of **34a**(−) (185 mg, 0.59 mmol), 1-methylpiperazine (59 mg, 0.59 mmol), *N*-(3-dimethylaminopropyl)-*N'*-ethylcarbodiimide hydrochloride (113 mg, 0.59 mmol), and 1-hydroxybenzotriazole hydrate (90 mg, 0.59 mmol) in tetrahydrofuran (15 ml) was stirred at 25 °C for 16 h, and the reaction mixture was then concentrated under reduced pressure. The resulting residue was chromatographed on a silica gel column ($CHCl_3$/MeOH=15/1) to give **5**(−) (203 mg, 87%). $[\alpha]_D^{27}$ −46.1° (*c*=1.0, MeOH).

(+)-2-(3-Fluorophenyl)-5,6-dimethyl-3-[2-(4-methyl-1-piperazinyl)-2-oxoethyl]isoindolin-1-one (5(+)) Using **34a**(+), it was obtained in a similar manner as that for **5**(−). $[\alpha]_D^{27}$ +47.4° (*c*=1.0, MeOH).

Compounds **3**(−), **27**(−) and **47**(−) [**JM-1232**(−)] were obtained in a similar manner as that for **34a**(−), **5**(−).

3(−): $[\alpha]_D^{29}$ −93.7° (*c*=1.0, $CHCl_3$).

27(−): $[\alpha]_D^{26}$ −49.6° (*c*=1.0, $CHCl_3$: MeOH=1 : 1).

47(−) [JM-1232(−)]: mp 187—189 °C; ESI-MS *m/z*: 390.2 $[M+H]^+$; positive HR-ESI-MS *m/z*: 390.2200 $[M+H]^+$ (Calcd for $C_{24}H_{28}N_3O_2$: 390.2183); ^{1}H-NMR ($CDCl_3$) δ: 2.15 (2H, quin, *J*=7.3 Hz), 2.20—2.22 (2H, m), 2.26 (3H, s), 2.35—2.44 (3H, m), 2.88 (1H, dd, *J*=3.4, 16.0 Hz), 2.98 (4H, t, *J*=7.5 Hz), 3.18—3.30 (2H, m), 3.61—3.74 (2H, m), 5.79—5.82 (1H, m), 7.20—7.23 (1H, m), 7.42—7.46 (3H, m), 7.64—7.67 (2H, m), 7.73 (1H, s). $[\alpha]_D^{32}$ −37.1° (*c*=1.0, MeOH).

Pharmacology. Determinations of HD_{50}, LD_{50} and Therapeutic Index The hypnotic dose (HD_{50}) and lethal dose (LD_{50}) were obtained using male ICR mice (4 weeks old) that had access to food and water *ad libitum* under a 12-h light/dark cycle. Six mice per dose were used. A selected dose of each compound was injected intravenously. Each compound was prepared as the hydrochloride salt and was dissolved in saline. The rate and volume of injection through the lateral tail vein was 0.1 ml/10 s and 0.1 ml/10 g body weight, respectively. After each injection, mice were placed on their backs to test for loss of righting reflex (LRR) as an index of hypnotic activity. From the percentage of mice in each group showing LRR for 30 s or longer, a probit analysis was used to calculate 50% hypnotic dose (HD_{50}). The duration of sleep was measured as the interval between the loss and regain of righting reflex. When HD_{50} was not calculated because of less activity, the mean sleeping time of 2 or 3 mice was calculated. The LD_{50} was determined from probit analysis from the number of mice dying at each dose. The therapeutic index was calculated as the LD_{50}/HD_{50} ratio.

All animal procedures were performed in accordance with the Guiding Principles for the Care and Use of Laboratory Animals, approved by the Committee of Animal Experimentation, Maruishi Central Research Laboratories.

Acknowledgements We thank Central Glass Co., Ltd. for cooperation in this research.

References and Notes

1) Sebel P. S., Lowdon J. D., *Anesthesiology*, **71**, 260—277 (1989).
2) Lindholm M., *Minerva Anesthesiol.*, **58**, 875—879 (1992).
3) Bennett S. N., Mcneil M. M., Bland L. A., Arduino M. A., Villarino M. E., Perrotta D. M., Burwen D. R., Welbel S. F., Pegues D. A., Stroud L., Zeitz P. S., Jarvis W. R., *N. Engl. J. Med.*, **333**, 147—154 (1995).
4) Hynynen M., Korttila K., Tammisto T., *Acta Anaesthesiologica Scandinavica*, **29**, 651—652 (1985).
5) Muir W. W., Gadawski J. E., *Am. J. Vet. Res.*, **59**, 157—161 (1998).
6) Wada T., Nakajima R., Kurihara E., Narumi S., Masuoka Y., Goto G., Saji Y., Fukuda N., *Jpn. J. Pharmacol.*, **49**, 337—349 (1989).
7) Dubinsky B., Vaidya A. H., Rosenthal D. I., Hochman C., Crooke J. J., Deluca S., Devine A., Cheo-Isaacs C. T., Carter A. R., Jordan A. D., Reitz A. B., Shank R. P., *J. Pharmacol. Exp. Ther.*, **303**, 777—790 (2002).
8) Japanese Patent Application Laid Open, No. 2004–189733.
9) Tapia I., Alonso-Cires L., López-Tudanca P. L., Mosquera R., Labeaga L., Innerárity A., Orjales A., *J. Med. Chem.*, **42**, 2870—2880 (1999).
10) Glen J. B., *Br. J. Anaesth.*, **52**, 731—742 (1980).