A Novel Series of Thromboxane A_2 Synthetase Inhibitors with Free Radical Scavenging and Anti-peroxidative Activities

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A novel series of indoline derivatives with imidazole and carboxyl moieties were synthesized and evaluated for their thromboxane A_2 (TXA_2) synthetase inhibiting, radical scavenging and anti-peroxidative activities. Among the compounds synthesized, 3-{5-substituted-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl}propionic acids showed free radical scavenging activity and inhibitory effects on lipid-peroxidation of rat brain homogenate and on arachidonate-induced TXA_2-dependent aggregation of rabbit platelets. The anti-platelet and anti-peroxidative activities were related to the lipophilicity of the 5-substituent. The 5-hexyloxy derivative (13) showed about 35-fold higher inhibitory activity on TXA_2 synthesis than that of ozagrel and about 100-fold higher activity on lipid peroxidation than that of α-tocopherol. Compound 13 showed *in vivo* anti-thrombotic effect in mice and *ex vivo* anti-peroxidative activity in rats.

Key words indoline derivative; thromboxane A_2 synthetase inhibitor; radical scavenger; anti-peroxidation

Thromboxane A_2 (TXA_2) is produced from prostaglandin H_2 (PGH_2) by TXA_2 synthetase in the arachidonic acid cascade, and has potent vasoconstricting and platelet aggregating activities.[1)] Prostaglandin I_2 (PGI_2) is also produced from PGH_2 and has vasodilatory and antiaggregatory activities. Thus, excessive production of TXA_2 and/or imbalance of TXA_2 and PGI_2 play an important role in pathogenesis of ischemic cerebral and heart disease, thromboembolic disorders and atherosclerosis.[1—3)] Selective TXA_2 synthetase inhibitors have been suggested to not only inhibit TXA_2 production but also to enhance PGI_2 synthesis since unused PGH_2 can be utilized by PGI_2 synthetase. Although a number of TXA_2 synthetase inhibitors have been reported, few have been successfully developed as clinically effective drugs for treatment of cerebral or coronary infarction or atherosclerosis. Superoxides and free radicals are also important pathogenetic factors in ischemic diseases and atherosclerosis.[4,5)] Antioxidants have been investigated as potential anti-ischemic drugs but their clinical efficacy has not been fully established. The arachidonic acid cascade and free radicals interact with each other in a complex manner. Free radicals are involved in enzymatic reactions in the arachidonic acid cascade and are released from endoperoxide metabolites of arachidonic acid. Furthermore, PGI_2 synthetase is more sensitive to inactivation by free radicals compared to TXA_2 synthetase, resulting in imbalance of PGI_2 and TXA_2. Free radicals activate platelets and cause production of TXA_2.[6)] TXA_2 plays a role in oxygen radical generation in cerebral ischemia-reperfusion injury.[7)] Therefore, simultaneous inhibition of TXA_2 and free radicals is expected to be more effective for protection against ischemic, thromboembolic and atherosclerotic disease than either alone. We reported previously that indapamide, a non-thiazide diuretic, has free radical scavenging activity, which is due to its indoline moiety, and protects PGI_2 synthetase from free radicals in vascular smooth muscle cells.[8)] Known TXA_2 synthetase inhibitors have an aromatic ring, such as a benzene, a benzofuran or a benzothiophen, which bears two side chains with a carboxylic acid and a heterocycle, respectively (Fig. 1).[9—13)] We designed a series of indoline derivatives with a carboxyl and an imidazole moiety to find a novel TXA_2 synthetase inhibitor with free radical scavenging activity.

Chemistry

Synthetic routes of the indoline-based imidazole derivatives are shown in Charts 1—3.

Chart 1 showed the general synthetic pathway for preparation of {3-[(imidazol-1-yl)alkyl]indolin-1-yl}alkylcarboxylic acid derivatives. The 5-substituted tryptophols (III, $n=1$) were prepared from the reaction between the appropriate phenyl hydrazines[14)] (I) and 2,3-dihydrofuran with the Grandberg and Moskvina method.[15)] The substituted tryptophols in other positions (4,6,7) (III, $n=1$) were prepared by condensation of oxalyl chloride with the appropriate indole[16)] (II) to give glyoxylates followed by reduction with $LiAlH_4$.[17)] The 5-substituted 3-(3-hydroxypropyl)indole (III, $n=2$) was prepared from ethyl 3-(5-hexyloxyindol-3-yl)propionate[18)] by reduction with $LiAlH_4$. The substituted tryptophols (III, $n=1$) and 3-(3-hydroxypropyl)indole (III, $n=2$) were re-

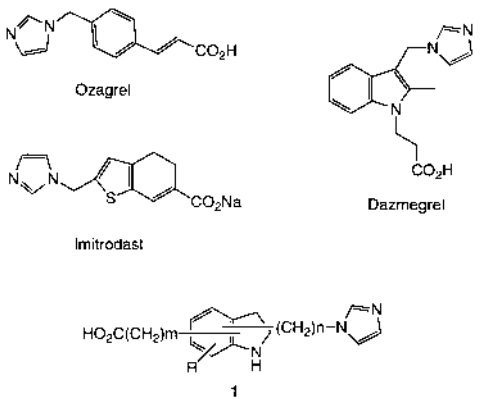

Fig. 1. TXA_2 Inhibitors and General Structure of Indoline-Based TXA_2 Inhibitors

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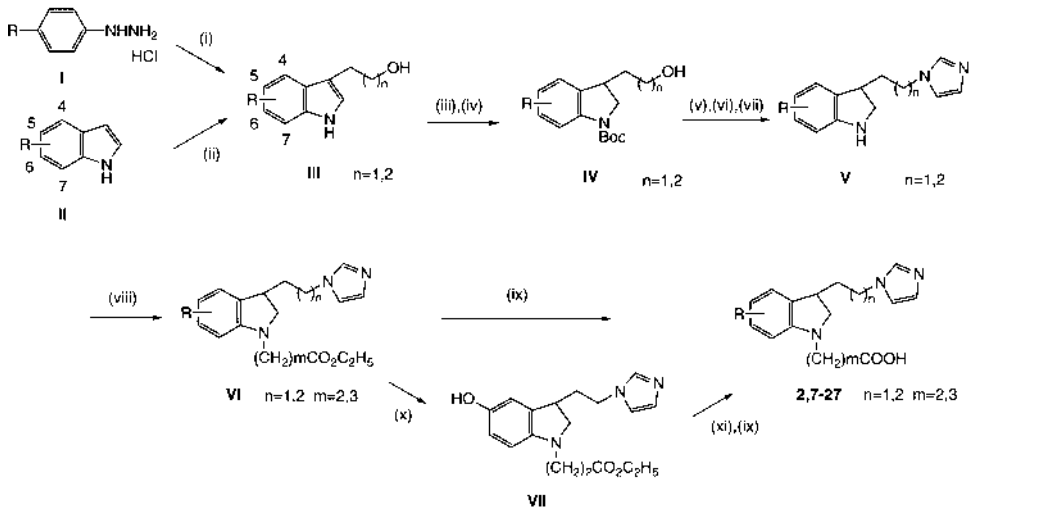
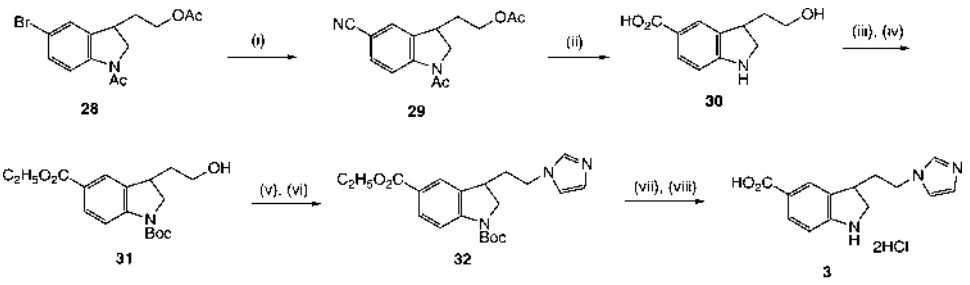


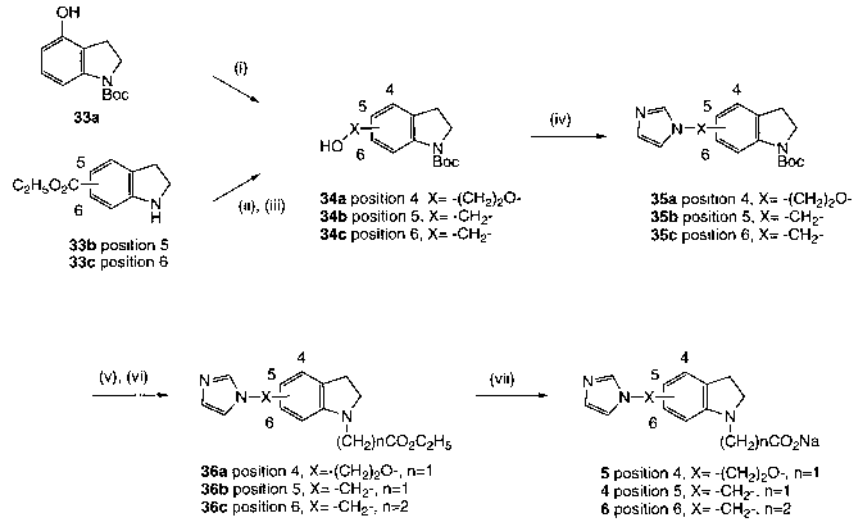
(i)2,3-dihydrofuran; (ii)a)ClCOCOCl, b)EtOH, c)$LiAlH_4$; (iii)$NaBH_3CN$, AcOH; (iv)$(Boc)_2O$; (v)CBr_4, Ph_3P; (vi)imidazole, K_2CO_3; (vii)HCl; (viii)$CH_2{=}CHCO_2C_2H_5$ or $Br(CH_2)_3CO_2C_2H_5$, K_2CO_3; (ix)NaOH; (x)TFA, anisole; (xi)R'X, *t*-BuOK.

Chart 1



(i)CuCN, *N*-methylpyrrolidone; (ii)HCl; (iii)C_2H_5OH, HCl; (iv)$(Boc)_2O$, triethylamine; (v)CBr_4, Ph_3P; (vi) imidazole, K_2CO_3; (vii)NaOH; (viii)HCl.

Chart 2



(i)ethylene bromohydrin, K_2CO_3; (ii)DIBAL-H or $LiAlH_4$; (iii)$(Boc)_2O$; (iv)CBr_4, Ph_3P, imidazole, K_2CO_3; (v)HCl; (vi)$BrCH_2CO_2C_2H_5$ or $CH_2{=}CHCO_2C_2H_5$; (vii)NaOH.

Chart 3

duced with sodium cyanoborohydride ($NaBH_3CN$) to corresponding indolines followed by protection with di-*tert*-butyl dicarbonate ($(Boc)_2O$) to afford *N*-protected indoline derivatives (IV). Bromination of IV with CBr_4 and Ph_3P followed by condensation with imidazole and deprotection with HCl to afford V. Target compounds (**2**, **7**—**9**, **12**—**27**) were obtained by condensation with ethyl acrylate or ethyl 3-bromopropionate followed by hydrolysis of ester precursors (VI) with NaOH.

Compounds **10** and **11** were prepared from the 5-hydroxyindoline derivative (VII). Deprotection of VI ($n=1$, $m=2$, $R=OCH_2C_6H_5$) was performed with trifluoroacetic acid (TFA) in anisole to afford the corresponding phenol (VII), which was condensed with the appropriate bromides followed by hydrolysis of ester precursors with NaOH to give **10** and **11**.

Chart 2 shows the synthetic route of **3**. 5-Bromoindoline derivative (**28**) was reacted with CuCN in *N*-methylpyrrolidone to afford 5-cyanoindoline derivative (**29**). Hydrolysis of **29** with conc. HCl gave the corresponding carboxylic acid (**30**),[19] which was protected with ester and $(Boc)_2O$ to give **31**. Bromination of **31** with CBr_4 and Ph_3P followed by condensation with imidazole gave **32**, which was deprotected with NaOH and HCl to afford **3**.

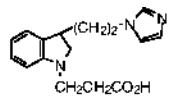
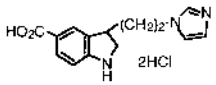
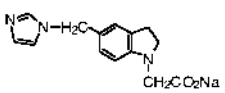
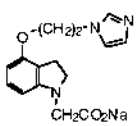
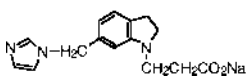
Chart 3 shows the synthetic route of the other indoline derivatives (**4**—**6**). The 4-substituted derivative (**34a**) was prepared from **33a** by condensation with ethylene bromohydrin. The 5- or 6-substituted derivatives (**34b**, **c**) were obtained by reduction of the appropriate indoline carboxylate (**33b**, **c**)[19] with diisobutylaluminum hydride (DIBAL-H) or $LiAlH_4$ and protection with $(Boc)_2O$. Bromination of **34a**—**c** with CBr_4 and Ph_3P followed by condensation with imidazole gave **35a**—**c**, which were deprotected with HCl followed by condensation with ethyl bromoacetate or ethyl acrylate followed by hydrolysis of ester precursors with NaOH to afford target compounds **4**—**6**.

Results and Discussion

In the present study, we synthesized a series of indoline derivatives and examined their biological activities to find novel TXA_2 synthetase inhibitors with free radical scavenging and anti-peroxidative activities. The inhibitory effects of compounds on the arachidonate-induced TXA_2-dependent aggregation of rabbit platelets were determined to estimate their activities against TXA_2 synthetase.[20,21] Selective TXA_2 synthetase inhibitors are known to partially inhibit arachidonate-induced aggregation without affecting adenosine 5′-diphosphate (ADP)-induced aggregation in rabbit platelets, while cyclooxygenase inhibitors abolish the arachidonate-induced aggregation. Free radical scavenging activity was evaluated using α,α-diphenyl-β-picrylhydrazyl (DPPH), a stable free radical. Anti-peroxidative activity was examined using rat brain homogenate.[8]

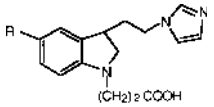
Known TXA_2 synthetase inhibitors have two moieties bearing a heterocycle (imidazole, thiazole, pyridine *etc*.) and a carboxylic acid, attached to an aromatic ring such as benzene, benzofuran, benzothiophene, indole or indane.[9—13] First, we synthesized five compounds, in which imidazole and carboxylic acid moieties were introduced at different positions on the indoline ring and examined their activities (Table 1). Compound **2** with an imidazole moiety at the 3-position and a carboxyl moiety at the 1-position showed an approximately 1.5-fold weaker inhibitory effect on arachidonate-induced aggregation than ozagrel, a selective TXA_2 synthetase inhibitor.[9] The shift of the imidazole moiety to the 5-position (compound **4**) apparently enhanced the anti-platelet activity, but this was probably by inhibition of cyclooxygenase rather than TXA_2 synthetase, since aggregation was

Table 1. Structure and Biological Activities of Indoline Derivatives with Two Moieties Bearing Imidazole and Carboxylic Acid

		Inhibition of AIPA[a] $IC_{20}(\times10^{-6}\,M)$	Radical scavenging[b] % reduction	Inhibition of LPO[c] $IC_{50}(\times10^{-6}\,M)$
2		13.8	67.6	>100
3		>100	21.7	>100
4		8.1	94.0	>100
5		>100	58.4	>100
6		>100	2.6	>100
Ozagrel		9.5	1.9	>100
α-Tocopherol		>100	92.0	70.0
Ascorbic acid		>100	96.5	>100

a) AIPA: arachidonate-induced aggregation of rabbit platelets. *b*) % reduction of DPPH (10^{-4} M) by test compounds (10^{-4} M). *c*) LPO: lipid peroxidation.

Table 2. Structure and Biological Activities of 3-{5-Substituted-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl}propionic Acids



No.	R	Inhibition of AIPA[a] IC_{20} ($\times 10^{-6}$ M)	Radical scavenging[b] % reduction	Inhibition of LPO[c] IC_{50} ($\times 10^{-6}$ M)
2	H	28.0	67.6	>100
7	$CH_3(CH_2)_2O$	4.1	72.3	19.4
8	$(CH_3)_2CH$	2.8	92.3	21.9
9	$CH_3(CH_2)_2$	4.3	92.8	24.8
10	$CH_2=CHCH_2O$	7.5	78.8	20.5
11	$C_2H_5O(CH_2)_2O$	4.0	77.0	170
12	$CH_3(CH_2)_4O$	2.7	76.7	1.84
13	$CH_3(CH_2)_5O$	1.0	77.1	0.53
14		6.6	67.1	2.35
15	$CH_3(CH_2)_5$	4.5	91.8	2.2
16		1.8	93.3	2.05
17	$CH_3(CH_2)_6O$	1.5	78.8	0.25
18		8.2	87.2	2.54
19	$CH_3(CH_2)_7O$	0.7	72.4	0.22
20	$CH_3(CH_2)_8O$	0.7	72.4	0.22
21	$CH_3(CH_2)_{11}O$	2.9	80.7	0.26
Ozagrel		9.5	1.9	>100
α-Tocopherol		>100	92.0	70.0
Ascorbic acid		>100	96.5	>100

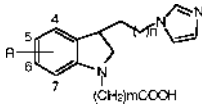
a) AIPA: arachidonate-induced aggregation of rabbit platelets. *b*) % reduction of DPPH (10^{-4} M) by test compounds (10^{-4} M). *c*) LPO: lipid peroxidation.

completely inhibited. The shift of the carboxyl moiety to the 5-position (compound **3**) and the imidazole moiety to the 4- or 6-positions (compounds **5**, **6**), respectively, abolished the inhibitory activity on arachidonate-induced aggregation. The position of the imidazole and the carboxylic acid on the aromatic ring and the distance between these functional groups are important for the activity of TXA_2 synthetase inhibitors.[9—13] Among the indoline-based derivatives, the position of two functional groups and the distance between them in compound **2** may be more favorable for interaction with TXA_2 synthetase than those in the other four compounds (**3—6**). Compound **4** showed potent free radical scavenging activity comparable to those of ascorbic acid and α-tocopherol. Compounds **2** and **5** showed moderate free radical scavenging activity, while compounds **3** and **6** had weak activity. All the compounds in Table 1 showed little effect on lipid peroxidation. We assumed that these compounds were too hydrophilic to interact with the lipid-bilayer of the plasma membrane of rat brain homogenate. Indeed, ascorbic acid, a hydrophilic antioxidant, effectively scavenged free radicals but had no inhibitory effect on lipid peroxidation.

In the next set of experiments, compound **2** was structurally modified to enhance the inhibitory activity on TXA_2 synthesis and lipid peroxidation (Table 2). Introduction of substituents including alkoxy, alkyl, alkenyl and alkylallyl at the 5-position increased the activity on arachidonate-induced aggregation. Among the alkoxy derivatives, the activity increased depending on the carbon number of the alkoxy group from 3 to 6 (compounds **7**, **12**, **13**), while it was similar from 6 to 9 (compounds **13**, **17**, **19**, **20**) and was reduced at 12 (compound **21**). Insertion of a double bond (compound **10**), an oxygen atom (compound **11**) or a cyclic structure (compounds **14**, **18**) tended to reduce the activity. The alkoxy moiety may tightly interact with the lipophilic region of the enzyme and enhance the inhibitory activity. All the compounds listed in Table 2 showed free radical scavenging activity similar to or stronger than that of compound **2**. Introduction of lipophilic substituents markedly increased the inhibitory activity on lipid peroxidation except compound **11**. Especially, the compounds with substituents containing more than 5 carbon atoms showed 24- to 318-fold stronger activity than that of α-tocopherol. Compounds with a longer and straight alkyl chain were more potent than those with a cycloalkyl or allylalkyl moiety. A long alkoxy chain seems to be favorable for interaction with the lipid-bilayer in plasma and microsomal membranes, as described for the synthetic ascorbate analogue CV-3611.[22]

Finally, the effects of distance between the imidazole and carboxyl moieties and the position of the alkoxy side chain on both activities were investigated. The lengths of the alkyl chains bearing imidazole and carboxyl groups were changed (Table 3). Insertion of one methylene between the two functional groups decreased the inhibitory effect on arachidonate-induced platelet aggregation (**13** *vs*. **22**, **23**), and introduction of two methylenes (**13** *vs*. **24**) markedly decreased the effect. For TXA_2 synthetase inhibitors, it is known that there is an optimal distance 8.5—9.0 Å between the nitrogen atom at the 1 position of the imidazole and the carboxyl carbon.[9] Changing the distance between the two functional groups had no effects on the free radical scavenging and anti-peroxidative activities (**13**, **22—24**). In compounds **20**, **25—27**, the effects of the position of the alkoxy moiety on the activities were examined. The shift of the nonyloxy group from the 5-position to the 6- or 7-position (**20** *vs*. **26**, **27**) maintained the inhibitory activity against arachidonate-induced aggregation, but the shift to the 4-position resulted in a marked reduction

Table 3. Structure and Biological Activities of 3-{3-[2-(Imidazol-1-yl)alkyl]indolin-1-yl}alkanoic Acids with Hexyloxy or Nonyloxy Side Chains



No	R	Position	*m*	*n*	Inhibition of AIPA[a] IC_{20} ($\times10^{-6}$ M)	Radical scavenging[b] % reduction	Inhibition of LPO[c] IC_{50} ($\times10^{-6}$ M)
13	$CH_3(CH_2)_5O$	5	2	1	1.0	77.1	0.53
22	$CH_3(CH_2)_5O$	5	2	2	3.0	72.7	0.30
23	$CH_3(CH_2)_5O$	5	3	1	4.5	83.1	0.54
24	$CH_3(CH_2)_5O$	5	3	2	18.5	77.0	0.83
25	$CH_3(CH_2)_8O$	4	2	1	6.0	62.2	2.7
20	$CH_3(CH_2)_8O$	5	2	1	0.7	80.7	0.26
26	$CH_3(CH_2)_8O$	6	2	1	0.5	84.8	2.9
27	$CH_3(CH_2)_8O$	7	2	1	0.7	94.4	1.25

a) AIPA: arachidonate-induced aggregation of rabbit platelets. *b*) % reduction of DPPH (10^{-4} M) by test compounds (10^{-4} M). *c*) LPO: lipid peroxidation.

in this activity (**20** *vs*. **25**). The anti-peroxidative activity was markedly reduced by the shift of the substituent to the 4-, 6- or 7-position. Thus, 3-{5-substituted-3-[2-(imidazol-1-yl)-ethyl]indolin-1-yl}propionic acid was confirmed to be a structure suitable for both inhibition of TXA_2 synthesis and lipid peroxidation.

The present results showed that normal alkoxy derivatives **13**, **17**, **19** and **20** have potent TXA_2 synthetase inhibitory and anti-peroxidative activities, indicating that the position of substituents on the indoline ring, the distance between the imidazolyl and the carboxyl group as well as the length of the alkoxy chain are favorable for both activities in these compounds. Smaller and less lipophilic compounds are desirable in terms of bioavailability and safety. Thus, compound **13** was selected and further evaluated. The IC_{50} values of **13** and ozagrel for inhibition of TXA_2 synthesis in rabbit platelets were 0.043 and 1.50×10^{-6} M ($n=4$), respectively. Compound **13** but not ozagrel at a dose of 30 mg/kg (*p.o.*) significantly prevented the arachidonate-induced sudden death in mice (mortality: vehicle 100% and compound **13** 40%, $n=10$, $p<0.05$, Fisher's exact method). Compound **13** (30 mg/kg, *p.o.*) significantly suppressed the *ex vivo* peroxidation in the rat plasma induced by $FeSO_4$ (malondialdehyde: 3.38 to 1.81 nmol/ml, $n=6$, $p<0.05$, Student's *t*-test). These results indicated that compound **13** exerted both anti-platelet and anti-peroxidative activities at the same dose *in vivo*.

Compound **13** is a potential therapeutic agent for the treatment of ischemic diseases by the dual inhibition of TXA_2 synthesis and lipid peroxidation. Further biological evaluation of **13** should be performed in experimental models of ishchemic disease, in which both TXA_2 and free radicals are considered to be involved.

Experimental

Melting points were measured on a Yamato capillary melting-point apparatus and are given uncorrected. IR spectra were taken with a JASCO IR-800 spectrometer. ^{1}H-NMR spectra were recorded at 90 MHz on a Hitachi R-1900 spectrometer using tetramethylsilane as an internal standard. For column chromatography, silica gel (Daisogel No.1001W, Daiso) was used.

2-(5-Hexyloxyindol-3-yl)ethanol (III, R=5-O(CH_2)$_5CH_3$, *n*=1) To a solution of 4-hexyloxyphenylhydrazine hydrochloride (I, R=4-O(CH_2)$_5CH_3$) (7.0 g) in MeOH (200 ml) was added 2,3-dihydrofuran (4.3 ml) portionwise at 15 °C. The reaction mixture was stirred at 50 °C for 6 h and the solvent was removed *in vacuo*. The residue was purified by column chromatography ($CHCl_3$–MeOH) to yield III (R=5-O(CH_2)$_5CH_3$, $n=1$) (3.1 g, yield 41%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 0.90 (br t, 3H), 1.1—2.2 (m, 8H), 3.00 (t, 2H, $J=6.3$ Hz), 3.90 (t, 2H, $J=6.3$ Hz), 4.00 (t, 2H, $J=6.5$ Hz), 6.7—7.4 (m, 4H), 8.05 (br, 1H). IR (neat): 3420, 1620, 1480 cm^{-1}.

The other 2-(5-substituted-indol-3-yl)ethanols (III) were prepared in a similar manner as described above. Yields and spectral data for these compounds are listed in Table 4.

2-(6-Nonyloxyindol-3-yl)ethanol (III, R=6-O(CH_2)$_8CH_3$, *n*=1) To a solution of 6-nonyloxyindole (II, R=6-O(CH_2)$_8CH_3$) (17.5 g) in Et_2O (200 ml) was added oxalyl chloride (19.9 g) in Et_2O (100 ml) at 5 °C dropwise followed by stirring at 20 °C for 5 h. After removal of the solvent *in vacuo*, the residue was dissolved in EtOH (200 ml) followed by stirring at 20 °C for 15 h. After removal of the solvent *in vacuo*, the residue was dissolved in tetrahydrofuran (THF) (500 ml) and to the solution was added $LiAlH_4$ (18.0 g). The reaction mixture was refluxed for 3 h, then quenched with water and the product was extracted with AcOEt (200 ml). The extracts were washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give III (R=6-O(CH_2)$_8CH_3$, $n=1$) (9.0 g, yield 44%) as a solid. ^{1}H-NMR ($CDCl_3$) δ: 0.90 (br t, 3H), 1.0—2.1 (m, 14H), 2.95 (t, 2H, $J=6.2$ Hz), 3.92 (t, 2H, $J=6.2$ Hz), 4.00 (t, 2H, $J=6.5$ Hz), 6.6—7.0 (m, 3H), 7.45 (d, 1H, $J=8.5$ Hz), 7.9 (br, 1H). IR (Nujol): 3400, 3250, 1480 cm^{-1}.

2-(4-Nonyloxyindol-3-yl)ethanol and 2-(7-nonyloxyindol-3-yl)ethanol were synthesized from corresponding indoles according to the procedure described right above.

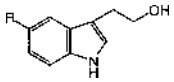
2-(4-Nonyloxyindol-3-yl)ethanol (III, R=4-O(CH_2)$_8CH_3$, $n=1$): Yield 27% as a solid. ^{1}H-NMR ($CDCl_3$) δ: 0.81 (br t, 3H), 1.0—2.1 (m, 14H), 3.18 (t, 2H, $J=6.0$ Hz), 3.95 (t, 2H, $J=6.0$ Hz), 4.09 (t, 2H, $J=6.4$ Hz), 6.41 (dd, 1H, $J=9.0$, 2.0 Hz), 6.7—7.3 (m, 3H), 8.0 (br, 1H). IR (Nujol): 3400, 1505 cm^{-1}.

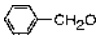
2-(7-Nonyloxyindol-3-yl)ethanol (III, R=7-O(CH_2)$_8CH_3$, $n=1$): Yield 79% as a solid. ^{1}H-NMR ($CDCl_3$) δ: 0.81 (br t, 3H), 1.1—2.1 (m, 14H), 3.02 (t, 2H, $J=6.3$ Hz), 3.90 (t, 2H, $J=6.3$ Hz), 4.10 (t, 2H, $J=6.7$ Hz), 6.67 (dd, 1H, $J=8.5$, 2.0 Hz), 6.8—7.5 (m, 3H), 8.4 (br, 1H). IR (Nujol): 3490, 3430, 1500 cm^{-1}.

3-(5-Hexyloxyindol-3-yl)propan-1-ol (III, R=5-O(CH_2)$_5CH_3$, *n*=2) To a solution of 3-(5-hexyloxyindol-3-yl)propionic acid ethyl ester (18.0 g) in THF (200 ml) was added $LiAlH_4$ (10.5 g) portionwise at 5 °C. The reaction mixture was stirred for 2 h at 20 °C, then quenched with water and the product was extracted with AcOEt (200 ml). The extracts were washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give III (R=5-O(CH_2)$_5CH_3$, $n=2$) (14.5 g, yield 92%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 0.93 (br t, 3H), 1.0—2.2 (m, 10H), 2.82 (t, 2H, $J=6.3$ Hz), 3.75 (t, 2H, $J=6.3$ Hz), 4.04 (t, 2H, $J=6.5$ Hz), 6.7—7.4 (m, 4H), 7.9 (br, 1H). IR (neat): 3400 cm^{-1}.

1-Boc-5-hexyloxy-3-(2-hydroxyethyl)indoline (IV, R=5-O(CH_2)$_5CH_3$, *n*=1) To a suspension of III (R=5-O(CH_2)$_5CH_3$, $n=1$) (3.3 g) in AcOH (21 ml) was added sodium cyanoborohydride (4.0 g) portionwise at 15 °C. The reaction mixture was stirred at the same temperature for 1 h, then poured into ice-water and neutralized with 4 N aq. NaOH. 5-Hexyloxy-3-(2-hydroxyethyl)indoline was extracted with $CHCl_3$ (100 ml) and the extracts were washed with water and dried. To the solution was added $(Boc)_2O$ (2.5

Table 4. Physical Data for 2-(5-Substituted-indol-3-yl)ethanols



R	Yield(%)	^{1}H-NMR ($CDCl_3$, δ, ppm)	IR (cm^{-1})
$CH_3(CH_2)_2O$	35	0.92 (t, 3H, *J*=6.3 Hz), 1.3—2.0 (m, 2H), 2.87 (t, 2H, *J*=6.0 Hz), 3.90 (t, 2H, *J*=6.0 Hz), 4.02 (t, 2H, *J*=6.3 Hz), 6.7—7.4 (m, 4H), 7.9 (br, 1H).	3550, 3420 1490
$(CH_3)_2CH$	11	1.35 (d, 6H, *J*=6.7 Hz), 2.3—2.6 (m, 1H), 3.02 (t, 2H, *J*=6.0 Hz), 3.90 (t, 2H, *J*=6.0 Hz), 6.9—7.5 (m, 4H), 7.9 (br, 1H).	3540, 3410 1480
$CH_3(CH_2)_2$	16	0.94 (t, 3H, *J*=6.3 Hz), 1.3—2.1 (m, 2H), 2.68 (t, 2H, *J*=6.3 Hz), 2.97 (t, 2H, *J*=6.3 Hz), 3.85 (t, 2H, *J*=6.3 Hz), 6.7—7.4 (m, 4H), 7.9 (br, 1H).	3530, 3405 1485
$CH_3(CH_2)_4O$	43	0.90 (br t, 3H), 1.0—2.5 (m, 6H), 2.90 (t, 2H, *J*=6.0 Hz), 3.90 (t, 2H, *J*=6.0 Hz), 3.95 (t, 2H, *J*=6.2 Hz), 6.6—7.4 (m, 4H), 7.8 (br, 1H).	3545, 3420 1485
	35	1.1—2.2 (m, 10H), 2.98 (t, 2H, *J*=6.0 Hz), 3.88 (t, 2H, *J*=6.0 Hz), 3.7—4.0 (m, 1H), 6.8—7.4 (m, 4H), 8.0 (br, 1H).	3540, 3420 1480
$CH_3(CH_2)_5$	16	0.92 (br t, 3H), 1.1—2.2 (m, 8H), 2.73 (t, 2H, *J*=6.5 Hz), 3.02 (t, 2H, *J*=6.2 Hz), 3.91 (t, 2H, *J*=6.2 Hz), 6.9—7.6 (m, 4H), 8.0 (br, 1H).	3540, 3430 1480
	10	1.1—2.2 (m, 10H), 2.3—2.7 (m, 1H), 2.95 (t, 2H, *J*=6.0 Hz), 3.83 (t, 2H, *J*=6.0 Hz), 6.8—7.4 (m, 4H), 7.9 (br, 1H).	3540, 3400 1480
$CH_3(CH_2)_6O$	42	0.89 (br t, 3H), 1.1—2.0 (m, 10H), 2.96 (t, 2H, *J*=6.0 Hz), 3.90 (t, 2H, *J*=6.0 Hz), 3.98 (t, 2H, *J*=6.4 Hz), 6.8—7.4 (m, 4H), 8.0 (br, 1H).	3550, 3420 1490
	46	2.95 (t, 2H, *J*=6.3 Hz), 3.87 (t, 2H, *J*=6.3 Hz), 5.08 (s, 2H), 6.8—7.1 (m, 9H), 7.9 (br, 1H).	3420, 1480
$CH_3(CH_2)_7O$	51	0.87 (br t, 3H), 1.1—2.0 (m, 12H), 2.98 (t, 2H, *J*=6.0 Hz), 3.90 (t, 2H, *J*=6.0 Hz), 4.00 (t, 2H, *J*=6.3 Hz), 6.7—7.4 (m, 4H), 7.9 (br, 1H).	3410, 1470
$CH_3(CH_2)_8O$	40	0.90 (br t, 3H), 1.1—2.1 (m, 14H), 2.90 (t, 2H, *J*=6.2 Hz), 3.92 (t, 2H, *J*=6.2 Hz), 4.02 (t, 2H, *J*=6.5 Hz), 6.6—7.4 (m, 4H), 8.0 (br, 1H)	3530, 3400 1505
$CH_3(CH_2)_{11}O$	37	0.87 (br t, 3H), 1.1—2.2 (m, 20H), 2.95 (t, 2H, *J*=6.2 Hz), 3.90 (t, 2H, *J*=6.2 Hz), 4.03 (t, 2H, *J*=6.5 Hz), 6.7—7.5 (m, 4H), 8.1 (br, 1H).	3530, 3420 1500

g) followed by stirring at 20 °C for 10 h. The solvent was removed *in vacuo* to afford an oil, which was purified by column chromatography (*n*-hexane–AcOEt) to yield IV (R=5-O$(CH_2)_5CH_3$, *n*=1) (4.2 g, yield 91%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 0.91 (br t, 3H), 1.0—2.2 (m, 10H), 1.55 (s, 9H), 3.2—4.4 (m, 7H), 6.65 (d, 1H, *J*=9.0 Hz), 6.70 (s, 1H), 7.26 (d, 1H, *J*=9.0 Hz). IR (neat): 3450, 1700 cm^{-1}.

5-Hexyloxy-3-[2-(imidazol-1-yl)ethyl]indoline (V, R=5-O$(CH_2)_5CH_3$, *n*=1) To a solution of IV (R=5-O$(CH_2)_5CH_3$, *n*=1) (4.3 g) and CBr_4 (5.9 g) in CH_3CN (50 ml) was added Ph_3P (4.6 g). The reaction mixture was stirred for 1 h at 20 °C and the solvent was removed *in vacuo*. The residue was dissolved in acetone (80 ml) and to the solution was added imidazole (8.0 g) and K_2CO_3 (4.9 g) followed by refluxing for 20 h. After removal of the solvent *in vacuo*, the residue was dissolved in AcOEt (100 ml). The solution was washed with water and dried. After removal of the solvent *in vacuo*, the residue was dissolved in EtOH (30 ml). To the solution was added 10 N HCl in EtOH (26 ml) followed by stirring at 20 °C for 2 h. After removal of the solvent *in vacuo*, 5-hexyloxy-3-[2-(imidazol-1-yl)ethyl]indoline hydrochloride was extracted with water (100 ml) and washed with AcOEt (50 ml). After neutralization with 4 N aq. NaOH, the extract with AcOEt (100 ml) was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give V (R=5-O$(CH_2)_5CH_3$, *n*=1) (2.7 g, yield 73%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 0.90 (br t, 3H), 1.0—2.5 (m, 10H), 2.8—3.5 (m, 4H), 3.88 (t, 2H, *J*=6.4 Hz), 4.03 (t, 2H, *J*=6.5 Hz), 6.5—6.8 (m, 3H), 6.94 (s, 1H), 7.09 (s, 1H), 7.52 (s, 1H). IR (neat): 3350, 3250, 1600, 1500 cm^{-1}.

Ethyl 3-{5-Hexyloxy-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl}propionate (VI, R=5-O$(CH_2)_5CH_3$, *n*=1, *m*=2) To a solution of V (R=5-O$(CH_2)_5CH_3$, *n*=1) (2.7 g) in EtOH (10 ml) was added ethyl acrylate (2.3 ml) followed by refluxing for 20 h. After removal of the solvent *in vacuo*, the residue was purified by column chromatography ($CHCl_3$–MeOH) to yield VI (R=5-O$(CH_2)_5CH_3$, *n*=1, *m*=2) (3.0 g, yield 84%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 0.90 (br t, 3H), 1.1—2.4 (m, 10H), 1.16 (t, 3H, *J*=6.2 Hz), 2.56 (t, 2H, *J*=6.3 Hz), 2.8—3.6 (m, 3H), 3.33 (t, 2H, *J*=6.3 Hz), 3.7—4.4 (m, 6H), 6.3—6.9 (m, 3H), 6.93 (s, 1H), 7.10 (s, 1H), 7.53 (s, 1H). IR (neat): 2925, 2850, 1735, 1600 cm^{-1}.

3-{5-Hexyloxy-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl}propionic Acid (13) To a solution of VI (R=5-O$(CH_2)_5CH_3$, *n*=1, *m*=2) (3.0 g) in 90% EtOH (30 ml) was added NaOH (0.92 g) followed by stirring at 20 °C for 3 h. After removal of the solvent *in vacuo*, the residue was dissolved in $CHCl_3$ (50 ml), the solution was washed with 5% aq. citric acid and dried. The solvent was removed *in vacuo* to give an oil, which was recrystallized from EtOH to give **13** (1.8 g, yield 64%) as white crystals. mp 101—102 °C: ^{1}H-NMR (DMSO-d_6) δ: 0.88 (br t, 3H), 1.0—2.3 (m, 10H), 2.50 (br t, 2H), 2.7—3.6 (m, 5H), 3.84 (t, 2H, *J*=6.3 Hz), 4.05 (t, 2H, *J*=6.7 Hz), 6.50 (d, 1H, *J*=8.5 Hz), 6.65 (d, 1H, *J*=8.5 Hz), 6.70 (s, 1H), 6.91 (s, 1H), 7.23 (s, 1H), 7.69 (s, 1H), 8.6 (br, 1H). IR (Nujol): 1700, 1500 cm^{-1}. *Anal*. Calcd for $C_{22}H_{31}N_3O_3$: C, 68.54; H, 8.11; N, 10.90. Found: C, 68.63; H, 8.26; N, 10.89. MS *m/z*: 385 (M^+).

Compounds **2**, **7**—**9**, **12**—**27** were prepared in a similar manner as described just above. Physical properties and spectral data for these compounds are listed in Table 5.

Ethyl 3-{5-Hydroxy-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl}propionate (VII) A solution of ethyl 3-{5-benzyloxy-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl}propionate (VI, R=5-O$CH_2C_6H_5$, *n*=1, *m*=2) (18 g) in anisole (90 ml) and TFA (90 ml) was stirred at 70 °C for 16 h. After removal of solvent *in vacuo*, to the residue was added water (300 ml) and the soluble products were extracted with $CHCl_3$ (100 ml) and discarded. The aqueous solution was neutralized with aq. $NaHCO_3$ and the product was extracted with $CHCl_3$ (300 ml). The extracts were washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give VII (12.1 g, yield 86%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.20 (t, 3H, *J*=6.3 Hz), 1.7—2.3 (m, 2H), 2.52 (br t, 2H), 2.6—3.7 (m, 5H), 3.92 (t, 2H, *J*=6.7 Hz), 4.05 (q, 2H, *J*=6.3 Hz), 6.2—6.9 (m, 3H), 6.85 (s, 1H), 7.00 (s, 1H), 7.42 (s, 1H). IR (neat): 1735, 1500 cm^{-1}.

{5-Allyloxy-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl}propionic Acid (10) To a solution of VII (2.7 g) in EtOH (40 ml) was added potassium *tert*-butoxide (6.0 g) followed by stirring at 20 °C for 0.5 h. To the reaction mixture was added allyl bromide (1.49 g) followed by stirring for 7 h at 90 °C. After removal of solvent *in vacuo*, the residue was dissolved in AcOEt (200 ml). The solution was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give the ester of **10** (0.9 g, yield 30%) as an oil. To a solution of the ester (0.9 g) in 90% EtOH (30 ml) was added NaOH (0.36 g) followed by stirring at 20 °C for 3 h. After removal of the solvent *in vacuo*, the residue was neutralized with 5% aq. citric acid and the product was extracted with $CHCl_3$ (50 ml). The extract was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was recrystallized from EtOH to give **10** (0.65 g, yield 78%) as crystals. mp 68—78 °C: ^{1}H-NMR (DMSO-d_6) δ: 1.5—2.3 (m, 2H), 2.51 (t, 2H, *J*=6.3 Hz), 2.6—3.7 (m, 5H), 4.00 (t, 2H, *J*=6.5 Hz), 4.45 (d, 2H, *J*=6.8 Hz), 5.0—6.3 (m, 3H), 6.38

Table 5. Physical Data for 3-[(Imidazol-1-yl)alkyl]substituted-indoline-1-alkanoic Acids

No.	mp (°C)	^{1}H-NMR (DMSO-d_6, δ, ppm)	IR (cm^{-1})
2	138—141	1.7—2.5 (m, 2H), 2.50 (br t, 2H), 2.7—3.3 (m, 5H), 4.00 (t, 2H, *J*=6.3 Hz), 5.00 (br, 1H), 6.3—7.4 (m, 6H), 7.62 (s, 1H).	1700, 1485
7	68—70	0.97 (t, 3H, *J*=6.4 Hz), 1.3—2.7 (m, 6H), 2.7—3.6 (m, 5H), 3.82 (t, 2H, *J*=6.3 Hz), 4.05 (t, 2H, *J*=6.5 Hz), 4.60 (br, 1H), 6.40 (d, 1H, *J*=9.0 Hz), 6.60 (d, 1H, *J*=9.0 Hz), 6.70 (s, 1H), 6.95 (s, 1H), 7.25 (s, 1H), 7.63 (s, 1H).	1695, 1485
8	135—137	1.15 (d, 6H, *J*=6.8 Hz), 1.2—3.6 (m, 10H), 4.01 (t, 2H, *J*=6.5 Hz), 4.50 (br,1H), 6.35 (d, 1H, *J*=9.3 Hz), 6.7—7.0 (m, 3H), 7.20 (s, 1H), 7.67 (s, 1H).	1700, 1495
9	109—110	0.86 (t, 3H, *J*=6.5 Hz), 1.1—2.7 (m, 6H), 2.48 (t, 2H, *J*=6.3 Hz), 2.7—3.6 (m, 5H), 4.02 (t, 2H, *J*=6.6 Hz), 4.80 (br, 1H), 6.38 (d, 1H, *J*=9.5 Hz), 6.6—7.1 (m, 3H), 7.19 (s, 1H), 7.66 (s, 1H).	1695, 1490
12	92—95	0.88 (br t, 3H), 1.0—2.0 (m, 8H), 2.0—3.5 (m, 7H), 3.80 (t, 2H, *J*=6.3 Hz), 4.00 (t, 2H, *J*=6.5 Hz), 5.00 (br, 1H), 6.3—6.8 (m, 3H), 6.85 (s, 1H), 7.18 (s, 1H), 7.65 (s, 1H).	1700, 1490
14	104—108	1.0—2.4 (m, 12H), 2.46 (br t, 2H), 2.6—3.7 (m, 5H), 3.8—4.0 (m, 1H), 4.05 (t, 2H, *J*=6.5 Hz), 6.40 (d, 1H, *J*=8.6 Hz), 6.70 (d, 1H, *J*=9.6 Hz), 6.75 (s, 1H), 6.95 (s, 1H), 7.25 (s, 1H), 7.75 (s, 1H), 8.60 (br, 1H).	1700, 1495
15	123—124	0.86 (br t, 3H), 1.0—1.6 (m, 8H), 1.6—2.7 (m, 4H), 2.50 (br t, 2H), 2.8—3.7 (m, 5H), 4.06 (t, 2H, *J*=6.5 Hz), 5.80 (br, 1H), 6.43 (d, 1H, *J*=9.3 Hz), 6.6—7.1 (m, 3H), 7.23 (s, 1H), 7.70 (s, 1H).	1700, 1500
16[a]	142—144	1.0—2.5 (m, 13H), 2.60 (t, 2H, *J*=6.0 Hz), 2.9—4.2 (m, 5H), 4.00 (br t, 2H), 6.44 (d, 1H, *J*=9.0 Hz), 6.6—7.2 (m, 4H), 7.58 (s, 1H), 8.40 (br, 1H).	1700, 1495
17	79—82	0.88 (br t, 3H), 1.0—2.4 (m, 12H), 2.50 (br t, 2H), 2.7—3.6 (m, 5H), 3.85 (t, 2H, *J*=6.3 Hz), 4.08 (t, 2H, *J*=6.5 Hz), 5.60 (br, 1H), 6.47 (d, 1H, *J*=9.0 Hz), 6.65 (d, 1H, *J*=9.0 Hz), 6.72 (s, 1H), 6.98 (s, 1H), 7.39 (s, 1H), 7.80 (s, 1H).	1695, 1485
18	109—112	1.6—2.4 (m, 2H), 2.45 (br t, 2H), 2.6—3.7 (m, 5H), 4.00 (t, 2H, *J*=6.8 Hz), 4.70 (br, 1H), 4.95 (s, 2H), 6.38 (d, 1H, *J*=8.7 Hz), 6.6—7.0 (m, 2H), 6.85 (s, 1H), 7.16 (s, 1H), 7.2—7.5 (m, 5H), 7.60 (s, 1H).	1700, 1500
19	71—74	0.90 (br t, 3H), 1.0—2.4 (m, 14H), 2.52 (br t, 2H), 2.7—3.6 (m, 5H), 3.85 (t, 2H, *J*=6.3 Hz), 4.10 (t, 2H, *J*=6.4 Hz), 6.00 (br, 1H), 6.3—6.7 (m, 3H), 6.90 (s, 1H), 7.21 (s, 1H), 7.70 (s, 1H).	1700, 1495
20	72—74	0.85 (br t, 3H), 1.0—2.3 (m, 16H), 2.45 (br t, 2H), 2.7—3.6 (m, 5H), 3.82 (t, 2H, *J*=6.0 Hz), 4.03 (t, 2H, *J*=6.3 Hz), 4.40 (br, 1H), 6.40 (d, 1H, *J*=8.2 Hz), 6.5—6.8 (m, 2H), 6.90 (s, 1H), 7.20 (s, 1H), 7.68 (s, 1H).	1695, 1490
21	64—68	0.88 (br t, 3H), 1.0—2.3 (m, 22H), 2.50 (br t, 2H), 2.7—3.6 (m, 5H), 3.85 (t, 2H, *J*=6.4 Hz), 4.06 (t, 2H, *J*=6.5 Hz), 4.40 (br, 1H), 6.3—6.8 (m, 3H), 6.96 (s, 1H), 7.25 (s, 1H), 7.65 (s, 1H).	1695, 1490
22[a]	80—82	0.88 (br t, 3H), 1.1—2.3 (m, 12H), 2.60 (t, 2H, *J*=6.5 Hz), 2.9—3.6 (m, 5H), 3.6—4.2 (m, 4H), 6.43 (d, 1H, *J*=8.4 Hz), 6.60 (s, 1H), 6.63 (d, 1H, *J*=8.4 Hz), 6.78 (s, 1H), 7.05 (s, 1H), 7.78 (br, 2H).	1710, 1490
23[a]	Oil	0.86 (br t, 3H), 1.0—2.6 (m, 14H), 2.6—3.5 (m, 5H), 3.6—4.2 (m, 4H), 6.2—6.7 (m, 3H), 6.38 (s, 1H), 7.11 (s, 1H), 7.70 (s, 1H), 8.30 (bs, 1H).	1710, 1500
24[a]	Oil	0.86 (br t, 3H), 1.0—2.6 (m, 16H), 2.6—3.5 (m, 5H), 3.5—4.3 (m, 4H), 6.2—6.8 (m, 4H), 7.10 (s, 1H), 7.62 (s, 1H), 8.20 (br, 1H).	1720, 1495
25[a]	80—84	0.85 (br t, 3H), 1.0—2.3 (m, 16H), 2.60 (br t, 2H), 3.0—3.2 (m, 5H), 3.2—4.4 (m, 4H), 6.2—7.2 (m, 4H), 7.13 (s, 1H), 7.65 (s, 1H), 9.0 (br, 1H).	1700
26[a]	86—87	0.90 (br t, 3H), 1.0—2.3 (m, 16H), 2.55 (br t, 2H), 2.9—3.6 (m, 5H), 3.6—4.3 (m, 4H), 6.08 (s, 1H), 6.12 (d, 1H, *J*=8.8 Hz), 6.6—7.1 (m, 3H), 7.64 (s, 1H), 8.7 (br, 1H).	1695, 1485
27[a]	94—95	0.85 (br t, 3H), 1.0—2.3 (m, 16H), 2.53 (t, 2H, *J*=6.0 Hz), 2.9—4.2 (m, 9H), 6.4—6.7 (m, 3H), 6.82 (s, 1H), 6.98 (s, 1H), 7.58 (s, 1H), 7.72 (br, 1H).	1700, 1490

a) Measured in $CDCl_3$.

(d, 1H, *J*=8.0 Hz), 6.59 (d, 1H, *J*=2.0 Hz), 6.80 (dd, 1H, *J*=8.0, 2.0 Hz), 7.05 (s, 1H), 7.32 (s, 1H), 7.90 (s, 1H), 8.50 (br, 1H). IR (Nujol): 1700, 1490 cm^{-1}.

Compound **11** was synthesized from VII and 2-bromoethyl ethyl ether according to the similar procedure described just above.

{5-Ethoxyethoxy-3-[2-(imidazol-1-yl)ethyl]indolin-1-yl} propionc Acid (**11**): Yield 55% as crystals. mp 104—106 °C: ^{1}H-NMR (DMSO-d_6) δ: 1.12 (t, 3H, *J*=6.2 Hz), 1.6—2.3 (m, 2H), 2.50 (br t, 2H), 2.8—3.7 (m, 9H), 3.7—4.3 (m, 4H), 4.4 (br, 1H), 6.3—6.8 (m, 3H), 6.97 (s, 1H), 7.28 (s, 1H), 7.81 (s, 1H). IR (Nujol): 1700, 1495 cm^{-1}.

3-Acetoxyethyl-1-acetyl-5-cyanoindoline (29) A suspension of **28** (13.5 g) and CuCN (7.8 g) in *N*-methylpyrrolidone (40 ml) was stirred at 200 °C for 2 h. The reaction mixture was cooled, diluted with AcOEt (300 ml), the solution was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography (toluene–AcOEt) to yield **29** (6.8 g, yield 60%) as a solid. ^{1}H-NMR ($CDCl_3$) δ: 1.6—2.5 (m, 2H), 2.08 (s, 3H), 2.27 (s, 3H), 3.2—4.1 (m, 3H), 4.18 (t, 2H, *J*=6.5 Hz), 7.35 (s, 1H), 7.42 (d, 1H, *J*=8.5 Hz), 8.22 (br d, 1H). IR (Nujol): 2220, 1740, 1610 cm^{-1}.

3-(2-Hydroxyethyl)indoline-5-carboxylic Acid (30) A solution of **29** (6.5 g) in conc. HCl (50 ml) was refluxed for 3 h. After concentrated to *ca*. 20 ml, the reaction mixture was adjusted to pH 10 with 10 N aq. NaOH. After removal of the insolubles by filtration, the aqueous solution was acidified to pH 4 with 1 N aq. HCl and the product was extracted with AcOEt (300 ml). The extracts were washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography (toluene–AcOEt) to yield **30** (3.2 g, yield 65%) as a solid. ^{1}H-NMR (DMSO-d_6) δ: 1.4—2.2 (m, 2H), 3.0—3.9 (m, 3H), 3.67 (t, 2H, *J*=6.5 Hz), 4.6 (br, 2H), 6.20 (br, 1H), 6.40 (d, 1H, *J*=9.0 Hz), 7.47 (s, 1H), 7.52 (d, 1H, *J*=9.0 Hz). IR (Nujol): 3360, 1700 cm^{-1}.

Ethyl 1-Boc-3-(2-hydroxyethyl)indoline-5-carboxylate (31) To a solution of **30** (3.0 g) in EtOH (100 ml) was added 10 N HCl in EtOH solution

(14.5 ml) and refluxed for 2 h. After removal of EtOH *in vacuo*, the residue was dissolved in $CHCl_3$ (100 ml) and the solution was washed with aq. $NaHCO_3$ and dried. After concentration of the solution to ca. 20 ml *in vacuo*, to the solution was added $(Boc)_2O$ (3.7 g) and triethylamine (1.7 g) followed by stirring at 60 °C for 3 h. The reaction mixture was washed with 5% aq. citric acid, water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to yield **31** (4.8 g, yield 96%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.38 (t, 3H, J=6.3 Hz), 1.57 (s, 9H), 1.6—2.4 (m, 2H), 3.3—4.0 (m, 5H), 4.32 (q, 2H, J=6.3 Hz), 7.5—8.0 (m, 3H). IR (neat): 3350, 1695, 1500 cm^{-1}.

Ethyl 1-Boc-3-[2-(imidazol-1-yl)ethyl]indoline-5-carboxylate (32) To a solution of **31** (4.8 g) and CBr_4 (9.9 g) in CH_3CN (100 ml) was added Ph_3P (4.7 g). The reaction mixture was stirred for 2 h at 20 °C. After removal of the solvent *in vacuo*, the residue was diluted with acetone (200 ml). To the solution was added imidazole (6.6 g) and K_2CO_3 (5.3 g) followed by refluxing for 4 h. After removal of the solvent *in vacuo*, the residue was dissolved in AcOEt (300 ml). The solution was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give **32** (3.2 g, yield 58%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.39 (t, 3H, J=6.3 Hz), 1.59 (s, 9H), 1.9—2.4 (m, 2H), 3.0—4.1 (m, 5H), 4.34 (q, 2H, J=6.3 Hz), 6.78 (s, 1H), 7.03 (s, 1H), 7.43 (s, 1H), 7.5—8.0 (m, 3H). IR (neat): 1705, 1495 cm^{-1}.

3-[2-(Imidazol-1-yl)ethyl]indoline-5-carboxylic Acid Dihydrochloride (3) To a solution of **32** (3.2 g) in 90% EtOH (30 ml) was added NaOH (1.5 g) followed by stirring at 60 °C for 6 h. After neutralization with 5% aq. citric acid, 1-Boc-3-[2-(imidazol-1-yl)ethyl]indoline-5-carboxylic acid was extracted with $CHCl_3$ (200 ml). The extracts were washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give the precursor of **3** (1.4 g, yield 47%) as an oil. To a solution of this oil (1.4 g) in $CHCl_3$ (30 ml) was added 10 N HCl in EtOH solution (20 ml) followed by stirring at 20 °C for 2 h. Crystals from the reaction mixture were recrystallized from EtOH to give **3** (0.8 g, yield 62%) as white crystals. mp 209—212 °C: ^{1}H-NMR (DMSO-d_6) δ: 1.8—3.0 (m, 2H), 3.2—4.0 (m, 3H), 4.40 (t, 2H, J=6.8 Hz), 7.08 (d, 1H, J=9.0 Hz), 7.6—8.0 (m, 4H), 9.25 (s, 1H), 10.4 (br, 4H). IR (Nujol): 1710, 1490 cm^{-1}. *Anal*. Calcd for $C_{14}H_{15}N_3O_2 \cdot 2HCl \cdot 0.25H_2O$: C, 50.23; H, 5.27; N, 12.55. Found: C, 49.94; H, 5.18; N, 12.54.

1-Boc-4-(2-hydroxyethoxy)indoline (34a) A mixture of **33a** (15.2 g), K_2CO_3 (22.0 g) and ethylene bromohydrin (20.7 g) in *N*,*N*-dimethylformamide (DMF) (100 ml) was stirred for 10 h at 60 °C. The reaction mixture was diluted with AcOEt (500 ml), washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give **34a** (10.2 g, yield 56%) as a solid. ^{1}H-NMR ($CDCl_3$) δ: 1.55 (s, 9H), 2.99 (t, 2H, J=7.5 Hz), 3.7—4.3 (m, 6H), 6.52 (d, 1H, J=8.5 Hz), 6.9—7.5 (m, 2H). IR (Nujol): 3320, 1710, 1605 cm^{-1}.

1-Boc-5-hydroxymethylindoline (34b) To a solution of ethyl indoline-5-carboxylate **33b** (19.0 g) in dry benzene (200 ml) was added 0.1 M DIBAL-H in toluene solution (200 ml) dropwise at 20 °C. The reaction mixture was stirred for 30 min at 40 °C, then quenched with water and the product was extracted with $CHCl_3$ (500 ml). The extract was concentrated to *ca*. 100 ml *in vacuo* and to the solution was added $(Boc)_2O$ (43.2 g) followed by stirring for 10 h at 20 °C. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography (benzene–AcOEt) to give **34b** (10 g, yield 40%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.58 (s, 9H), 3.05 (t, 2H, J=7.5 Hz), 3.97 (t, 2H, J=7.5 Hz), 4.58 (s, 2H), 7.05 (br d, 1H), 7.12 (s, 1H), 7.55 (br d, 1H). IR (neat): 3420, 1700, 1495 cm^{-1}.

N-Boc-6-hydroxymethylindoline (**34c**): Yield 55% as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.53 (s, 9H), 3.02 (t, 2H, J=7.5 Hz), 3.95 (t, 2H, J=7.5 Hz), 4.62 (s, 2H), 6.92 (d, 1H, J=8.5 Hz), 7.06 (d, 1H, J=8.5 Hz), 7.70 (s, 1H). IR (neat): 3400, 1700, 1500 cm^{-1}.

1-Boc-5-(imidazol-1-yl)methylindoline (35b) To a solution of **34b** (10.0 g) and CBr_4 (26.7 g) in CH_3CN (200 ml) was added Ph_3P (10.5 g) at 10 °C. The reaction mixture was stirred for 10 min at 20 °C. To the reaction mixture was added imidazole (68.0 g) followed by stirring for 2 h at 20 °C. After removal of the solvent *in vacuo*, the residue was dissolved in AcOEt (500 ml), the solution was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give **35b** (12.0 g, yield 95%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.55 (s, 9H), 3.02 (t, 2H, J=7.5 Hz), 3.96 (t, 2H, J=7.5 Hz), 5.00 (s, 2H), 6.7—7.1 (m, 4H), 6.95 (s, 1H), 7.15 (br d, 1Hz). IR (neat): 1700, 1500 cm^{-1}.

1-Boc-4-[2-(imidazol-1-yl)ethoxy]indoline (**35a**): Yield 37% as a solid. ^{1}H-NMR ($CDCl_3$) δ: 1.57 (s, 9H), 2.96 (t, 2H, J=8.0 Hz), 3.99 (t, 2H, J=8.0 Hz), 4.28 (br s, 4H), 6.40 (d, 1H, J=8.0 Hz), 6.9—7.5 (m, 4H), 7.53 (s, 1H): IR (Nujol): 1700 cm^{-1}.

1-Boc-6-(imidazol-1-yl)methylindoline (**35c**): Yield 92% as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.55 (s, 9H), 3.08 (t, 2H, J=7.5 Hz), 3.99 (t, 2H, J=7.5 Hz), 5.08 (s, 2H), 6.65 (d, 1H, J=8.5 Hz), 6.92 (d, 1H, J=8.5 Hz), 6.95 (s, 1H), 7.05 (s, 1H), 7.60 (s, 1H), 7.70 (s, 1H). IR (neat): 1700, 1500 cm^{-1}.

Ethyl 2-[5-(Imidazol-1-yl)methylindolin-1-yl]acetate (36b) To a solution of **35b** (12.0 g) in $CHCl_3$ (300 ml) was added 10 N HCl in EtOH solution (40 ml). The reaction mixture was stirred for 2 h at 20 °C. After neutralization with aq. $NaHCO_3$, the organic layer was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give 5-(imidazol-1-yl)methylindoline (3.6 g, yield 47%) as a solid. A mixture of 5-(imidazol-1-yl)methylindoline (1.0 g), ethyl bromoacetate (1.0 g) and K_2CO_3 (1.4 g) in DMF (20 ml) was stirred at 20 °C for 20 h. The reaction mixture was diluted with AcOEt (200 ml), the solution was washed with water and dried. The solvent was removed *in vacuo* to give an oil, which was purified by column chromatography ($CHCl_3$–MeOH) to give **36b** (0.8 g, yield 56%) as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.25 (t, 3H, J=6.3 Hz), 2.98 (t, 2H, J=7.5 Hz), 3.56 (t, 2H, J=7.5 Hz), 3.87 (s, 2H), 4.18 (q, 2H, J=6.3 Hz), 4.95 (s, 2H), 6.30 (d, 1H, J=8.0 Hz), 6.7—6.9 (m, 3H), 6.95 (s, 1H), 7.43 (s, 1H). IR (neat): 1740, 1500 cm^{-1}.

Ethyl 2-{4-[2-(Imidazol-1-yl)ethoxy]indolin-1-yl}acetate (**36a**): Yield 13% as a solid. ^{1}H-NMR ($CDCl_3$) δ: 1.27 (t, 3H, J=6.5 Hz), 2.93 (t, 2H, J=8.0 Hz), 3.55 (t, 2H, J=8.0 Hz), 3.85 (s, 2H), 4.07 (q, 2H, J=6.5 Hz), 4.10 (br s, 4H), 6.07 (d, 1H, J=8.5 Hz), 6.19 (d, 1H, J=8.5 Hz), 6.7—7.2 (m, 3H), 7.55 (s, 1H). IR (Nujol): 1730, 1620, 1600 cm^{-1}.

Ethyl 3-[6-(Imidazol-1-yl)methylindolin-1-yl]propionate (**36c**): Yield 58% as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.23 (t, 3H, J=6.3 Hz), 2.55 (t, 2H, J=6.3 Hz), 2.92 (t, 2H, J=7.5 Hz), 3.2—3.6 (m, 4H), 4.15 (q, 2H, J=6.3 Hz), 5.00 (s, 2H), 6.27 (s, 1H), 6.40 (d, 1H, J=8.5 Hz), 6.90 (s, 1H), 6.97 (d, 1H, J=8.5 Hz), 7.05 (s, 1H), 7.53 (s, 1H). IR (neat): 1735, 1505 cm^{-1}.

Sodium 2-[5-(Imidazol-1-yl)methylindolin-1-yl]acetate (4) To a solution of **36b** (0.8 g) in 90% EtOH (40 ml) was added NaOH (0.2 g) followed by stirring at 20 °C for 2 h. After removal of EtOH *in vacuo*, the residue was dissolved in water (300 ml) and washed with AcOEt (100 ml). The solvent was concentrated to ca. 100 ml *in vacuo* and subjected to Diaion HP-21 column chromatography. The fraction eluted with MeOH–water was lyophilized to give **4** (0.25 g, yield 30%) as a solid. ^{1}H-NMR (DMSO-d_6) δ: 2.87 (t, 2H, J=7.6 Hz), 3.47 (t, 2H, J=7.6 Hz), 3.85 (s, 2H), 4.97 (s, 2H), 6.35 (d, 1H, J=8.5 Hz), 6.7—7.1 (m, 3H), 7.12 (s, 1H), 7.70 (s, 1H). IR (Nujol): 1600, 1500 cm^{-1}.

Sodium 2-{4-[2-(Imidazol-1-yl)ethoxy]indolin-1-yl}acetate (**5**): Yield 52% as a solid. ^{1}H-NMR (DMSO-d_6) δ: 2.80 (t, 2H, J=7.8 Hz), 3.2—3.8 (m, 4H), 3.9—4.6 (m, 4H), 6.05 (d, 1H, J=8.5 Hz), 6.20 (d, 1H, J=8.5 Hz), 6.7—7.1 (m, 2H), 7.28 (s, 1H), 7.70 (s, 1H). IR (Nujol): 1600 cm^{-1}.

Sodium 3-[6-(Imidazol-1-yl)methylindolin-1-yl]propionate (**6**): Yield 42% as a solid. ^{1}H-NMR (DMSO-d_6) δ: 2.22 (t, 2H, J=6.5 Hz), 2.82 (t, 2H, J=7.5 Hz), 3.0—3.7 (m, 4H), 5.01 (s, 2H), 6.35 (s, 1H), 6.38 (d, 1H, J=8.5 Hz), 6.85 (s, 1H), 6.92 (d, 1H, J=8.5 Hz), 7.05 (s, 1H), 7.62 (s, 1H). IR (Nujol): 1560 cm^{-1}.

Rabbit Platelet Aggregation (*in Vitro*) Male Japanese white rabbits (approximately 3 kg) were used. Blood was taken from the carotid artery under anesthesia with sodium pentobarbital (20 mg/kg, i.v.). Platelet-rich plasma (PRP) and platelet-poor plasma (PPP) were obtained by centrifugation at 1000 rpm for 10 min and at 3000 rpm for 10 min at 4 °C, respectively. The platelet density of PRP was adjusted to 4×10^5 cells/μl. Platelet aggregation was measured with an NKK Hema Tracer (Niko Bioscience). PRP was incubated with test compounds or ozagrel at 37 °C for 1 min, followed by addition of arachidonic acid or ADP.

TXA_2 Synthesis in Washed Rabbit Platelets Rabbit blood was collected from the carotid artery under anesthesia with sodium pentobarbital and mixed with acid–citrate–dextrose (ACD) solution (sodium citrate 85 mM, citric acid 64 mM and dextrose 100 mM). PRP was obtained and the platelets were washed three times with Tyrode's solution. The platelets were finally suspended in Tyrode's solution at 10^5 cells/μl. The platelet suspension was incubated at 37 °C for 10 min in the presence of aspirin (10^{-5} M) and then further incubated with PGH_2 for 30 min. TXA_2 produced was determined as TXB_2 using a TXB_2[^{3}H]RIA kit.

Free Radical Scavenging DPPH, a stable free radical was dissolved at 10^{-4} M in ethanol and incubated at 25 °C for 20 min in the absence or presence of the synthesized compounds (10^{-4} M) according to the method previously reported.[8] Remaining DPPH in the absence and presence of the test compounds was determined spectrophotometrically at a wavelength of 517

nm and % reduction by the compounds was calculated.

Lipid Peroxidation in Rat Brain Homogenate Production of lipidperoxide was determined as malondialdehyde (MDA) in the rat brain homogenate as reported previously[8]. Briefly, the cerebral cortex isolated from male SD rats (6-week old) was homogenized in ice-cold 50 mM phosphate-buffered saline (pH.7.4). The homogenate was centrifuged at 1300 ***g*** for 10 min at 4 °C. The supernatant was incubated at 37 °C for 30 min in the presence of vehicle or test compounds. The reaction was terminated by addition of 20% trichloroacetic acid, and the mixture was immediately centrifuged at 2000 ***g*** for 15 min at 4 °C. The supernatant was heated at 100 °C for 15 min with thiobarbituric acid solution at pH 7.0. The optical absorbance of MDA was measured at 532 nm. The MDA level in medium before the assay was determined, and the net production of MDA during the incubation period in the presence of vehicle or test compounds was determined. Inhibition (%) of lipid peroxidation was calculated by comparison of MDA produced in the presence of vehicle and test compounds.

Plasma Lipid Peroxidation in Rats (*ex Vivo*) Male Wistar rats (7-week old) were used. Blood was taken from the abdominal aorta under anesthesia with sodium pentobarbital (50 mg/kg, i.p.) 1 h after administration of compound **13** (30 mg/kg) or vehicle. Plasma was seperated and incubated with $FeSO_4$ (5 mM) at 37 °C for 3 h. The lipid peroxide levels were determined as MDA.

Arachidonate-Induced Death in Mice Male ddY mice (5-week old) were used. Arachidonic acid (100 mg/kg) was injected into the tail veins of mice and the mortality was observed for 180 min. Compound **13** and ozagrel (30 mg/kg) were administered orally 30 min before the injection of arachidonate.

References

1) Moncada S., Vane J. R., *Pharmacol. Rev*., **30**, 293—331 (1979).
2) Wennmalm A., *Acta Physiol. Pol*., **39**, 217—224 (1988).
3) Patrono C., Ciabattoni G., Davi G., *Stroke*, **21**, IV130—133 (1990).
4) Clark I. A., Cowden W. B., Hunt N. H. *Med. Res. Rev*., **5**, 297—332 (1985).
5) Roberfroid M. B., Viehe H. G., Remacle J., *Ad. Drug Res*., **16**, 1—84 (1987).
6) Leo R., Pratico D., Iuliano L., Pulcinelli F. M., Ghiselli A., Pignatelli P., Colavita A. R., FitzGerald G. A., Violi F., *Circ.*, **95**, 885—891 (1997).
7) Matsuo Y., Kihara I., Ikeda M., Ninomiya M., Onodera H., Kogure K., *Brain Res*., **709**, 296—302 (1996).
8) Uehara Y., Shirahase H., Nagata T., Ishimitsu T., Morishita S., Osumi S., Matsuoka H., Sugimoto T., *Hyperten.*, **15**, 216—224 (1990).
9) Iizuka K., Akahane K., Momose D., Nakazawa M., *J. Med. Chem*., **24**, 1139—1148 (1981).
10) Tanouchi T., Kawamura M., Ohyama I., Kajiwara I., Iguchi Y., Okada T., Miyamoto T., Taniguchi K., Hayashi M., *J. Med. Chem*., **24**, 1149—1155 (1981).
11) Cross P. E., Dickinson R. P., Parry M. J., Randall M. J., *J. Med. Chem*., **29**, 1637—1643 (1986).
12) Amemiya Y., Terada A., Wachi K., Miyazawa H., Hatakeyama N., Matsuda K., Oshima T., *J. Med. Chem*., **32**, 1265—1272 (1989).
13) Kanao M., Watanabe Y., Kimura Y., Saegusa J., Yamamoto K., Kanno H., Kanaya N., Kubo H., Ashida S., Ishikawa F., *J. Med. Chem*., **32**, 1326—1334 (1989).
14) Kosower E. M., Miyadera T., *J. Med. Chem*., **15**, 307—312 (1972).
15) Grandberg I. I ., Moskvina T. P., *Khim. Geterotsikl. Soedin*, **10**, 1366—1367 (1972).
16) Hunt R. R., Rickard R. L., *J. Chem. Soc*., **1966**, 344—345.
17) Demerson C. A., Humber L. G., Philipp A. H., *J. Med. Chem*., **19**, 391—395 (1976).
18) Rensen M., *Bull. Soc. Chem. Belg*., **68**, 258—269 (1959).
19) Ikan R., Rapaport E., *Tetrahedron*, **23**, 3823—3827 (1967).
20) Tsuruda M., Mikashima H., Oe T., Kawasaki K., Setoguchi S., Naka Y., Tahara T., *Yakugaku Zasshi*, **109**, 33—45 (1989).
21) Kamiya S., Matsui H., Shirahase H., Nakamura S., Wada K., Kanda M., Shimaji H., Kakeya N., *Chem. Pharm. Bull*., **43**, 1692—1695 (1995).
22) Kato K., Terao S., Shimamoto N., Hirata M., *J. Med. Chem*., **31**, 793—798 (1988).