STUDIES ON BENZOHETEROCYCLIC DERIVATIVES (IX)
STRUCTURE-ACTIVITY RELATIONSHIPS OF 5 OR 7
SUBSTITUTED 2-(3'-ALKOXYPROPYLAMINOMETHYL)
2.3-DIHYDROBENZOFURAN ANALOGUES ON
ANALGESIC, SPINAL REFLEX DEPRESSING
AND ADRENERGIC α-BLOCKING EFFECTS

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It has been reported that benzodioxan derivatives, e.g. 2-alkoxymethyl 1.4-benzo-
dioxan (1) and 2-(3'-methoxypropylaminomethyl) 1.4-benzodioxan (Quiloflex) (2, 3) have
adrenergic α-blocking, sedative and spinal reflex depressing properties.

Through our intensive pharmacological screening of benzofuran derivatives which
possess quite similar chemical structure with 1.4-benzodioxan ring, we found that most
of these compounds revealed the similar pharmacological properties to the benzodioxan
derivatives (4). It is very interesting that 2-(3'-alkoxypropylaminomethyl) 2.3-dihydro-
benzofuran analogues which showed relatively potent skeletal muscle relaxant, sedative,
analgesic and adrenergic α-blocking actions, were very similar to chemical structure of
Quiloflex.

The purpose of our present work was to investigate the structure-activity relations-
ships of 5 or 7 substituted 2-(3'-alkoxypropylaminomethyl) 2.3-dihydrobenzofuran ana-
logues, particularly of the substituents and their position on the ring concerning analgesic,
spinal reflex depressing and adrenergic α-blocking effects.

Their compound numbers and chemical structures listed in Table 1.

| Table 1. Chemical structures and compound numbers of benzofuran derivatives. |
|-----------------------------------------------|---|---|---|
| X                                             | R   | -CH₃ | -C₃H₇ | -C₃H₇ (iso) |
| 5-CH₃                                         | No. 29 | No. 30 | No. 31 |
| 7-CH₃                                         | 32   | 33    | 34    |
| 5-Cl                                          | 35   | 36    | 37    |
| 7-Cl                                          | 38   | 39    | 40    |
| 5-OCH₃                                        | 41   | 42    | 43    |
| 5,7-H                                         | 45   | 46    | 47    |
METHODS

1) Analgesic action

Groups of 10 young male mice weighing 14 to 16 were employed in the experiments. Analgesic effect was evaluated by the electric stimulating method by Ozawa et al. (5), modified in our laboratories (6).

The pain threshold was determined by measuring the onset time of vocalization induced by electric stimulus (50 volt, 20 msec and 1 Hz) applied on the tail of mouse. Having been determined the normal reaction time (normal pain threshold), the drug was injected intraperitoneally and subsequently, the reaction time was measured at 10 to 30 minute intervals. With some of the drugs AD-50 values were estimated using Litchfield and Wilcoxon method (7).

2) Spinal reflex depressing action

Experiments were performed on unanesthetized, high spinal, adult cats of 2.5 to 4.0 kg in body weight. Under ether anesthesia the trachea was canulated and the spinal cord transected at the level of C-1. After spinal transection, respiration was artificially maintained. Animals were mounted on a “Noken” type stereotaxic instrument designed to allow immobilization of the cord. The exposed lumbosacral region of the cord was bathed in mineral oil maintained at 36 to 37°C by the use of an infrared lamp. The tibial or peroneal nerve was placed on bipolar platinum hook electrodes and stimulated by supra-maximal (5 times the threshold) square waves, 0.5 msec in duration, with a frequency of 12/min. The ipsilateral L-7 or S-1 ventral root was isolated and placed on bipolar platinum hook recording electrodes. Spinal monosynaptic (M.S.R.) and polysynaptic reflexes (P.S.R.) were photographically recorded by a cathode ray oscilloscope. The tested drugs and mephenesin as a standard were injected into the radial vein at least 2 hours after cessation of ether anesthesia.

3) Adrenergic α-blocking action

Experiments were carried out on adult mongrel dogs weighing 7.0 to 15.0 kg of both sexes. Animals were anesthetized with morphine (10 mg/kg s.c.) and urethane (1,000 mg/kg s.c.). The blood pressure was recorded from a common carotid artery with a mercury manometer on a smoked kimograph paper or with a high pressure transducer on a direct writing oscillogram. Having been determined the normal pressore response produced by adrenaline (10 μg/kg i.v.), the tested drugs were intravenously injected into the juglar vein and subsequently the same dose of adrenaline was injected at 30 min. intervals.

Some drugs which showed relatively potent adrenolytic effect on the morphine-urethane anesthetized dogs were also investigated in pentobarbital (40 mg/kg i.p.) and α-chloralose (100 mg/kg i.p.)-urethane (1,000 mg/kg s.c.) anesthetized dogs.

Ilidal, 6-allyl 6,7-dihydro-5 H dibenzo (c.a.) azepine phosphate was used as the reference drug of adrenergic α-blocking drug.
TABLE 2. Analgesic effects AD-50 of benzofuran derivatives tested by means of electric stimulating method in mice.

| X, R | -CH₃ | -C₂H₅ | -C₃H₇ (iso) |
|------|------|-------|------------|
| 5,7-H | 80.5 (65.5-97.6) | 57.0 (38.5-84.5) | 104 (87.4-124) |
| 5-CH₃ | 100 [6/11] | 100 [5/10] | 50 [2/10] |
| 5-Cl | 100 [3/10] | 82.5 (63.5-107) | 50 [3/10] |
| 5-OCH₃ | 100 [1/10] | 100 [1/10] | 100 [1/10] |
| 7-CH₃ | 72.0 (58.5-88.5) | 59.2 (42.5-82.4) | 100 [5/10] |
| 7-Cl | 73.0 (52.0-102) | 55.0 (43.6-69.2) | 100 [6/10] |

[ ] Number of effective animals/Number of treated animals
Aminopyrine (i.p.) AD-50 151 mg/kg (122-188)

RESULTS

1) Analgesic action

Analgesic effects (AD-50) of benzofuran derivatives are shown in Table 2. No. 33, 39 and 46 of the compounds which have respectively CH₃, Cl and H at the position of 7 on the benzofuran ring and ethoxy group at 3' position as substituents were shown relatively potent analgesic effect in this analogues. AD-50 values of these three compounds are about 60 mg/kg (i.p.) and almost as 3 times potent as that of aminopyrine. On the other hand, CH₃ or Cl substitution at 5 position of benzofuran ring reduced their analgesic actions. Compounds which have methoxy or isoproxy groups at 3' position as alkoxy substituents showed less potent analgesic effect than that of ethoxy groups.

Fig. 1 shows the time course of analgesic effect of No. 33, 39 and 46 of the compounds. The peak analgesic effect of these compounds appeared at 10 minutes after the administration and then their effects were gradually reduced and disappeared within 90 to 120 minutes.
TABLE 3. Depressing effect of benzofuran derivatives on spinal monosynaptic (M.S.R.) and polysynaptic reflex (P.S.R.) in spinal cats.

| X       | M.S.R. | P.S.R. | M.S.R. | P.S.R. | M.S.R. | P.S.R. |
|---------|--------|--------|--------|--------|--------|--------|
| 5,7-H   | 50-60  | 35-40  | 75-100 | 40-100 | 30-60  | 40-70  |
| 5-CH₃   | 0-10   | 0      | 0-20   | 0      | 0-15   | 0      |
| 5-Cl    | 0-20   | 0-10   | 0-20   | 0      | 15-50  | 0-20   |
| 5-OCH₃  | 0      | 0      | 0      | 0      | 0      | 0      |
| 7-CH₃   | 0-10   | 20-50  | 30-50  | 20-60  | 20-40  | 10-50  |
| 7-Cl    | 0-20   | 10-30  | 20-40  | 30-80  | 10-35  | 35-70  |

Fig. 2. Time course of depressing effects of benzofuran derivatives on spinal monosynaptic (M.S.R.) and polysynaptic reflex (P.S.R.) in spinal cats.
2) *Spinal reflex depressing action*

Within a few minutes after the intravenous administration of 2 to 5 mg/kg of benzofuran derivatives, M.S.R. and P.S.R. were apparently depressed. The rate of maximal depression of M.S.R. and P.S.R. after intravenous administration of 5 mg/kg of each compounds were illustrated in Table 3.

The spinal reflex depressing effects of benzofuran derivatives were slightly decreased by replacement with CH₃ or Cl at 7 position of benzofuran ring and markedly decreased by replacement with CH₃, Cl or OCH₃ at 5 position. The compounds having ethoxy group at 3' position as alkoxy substituents showed more potent activities than those having methoxy or isopropoxy group. The depression time course of the M.S.R. and P.S.R. produced by No. 45, 46 and 47 were shown in Fig. 2. These compounds depressed the both of M.S.R. and P.S.R. by same degree. The compound No. 46 had most potent depressing effect on spinal reflex, which depressed the both the M.S.R. and P.S.R. by 60 to 70% and it's effect lasted at least for 150 minutes at dose of 5 mg/kg (i.v.).

The depressing effect of mephenesin (30 mg/kg i.v.) on spinal reflex was illustrated in Fig. 3, which was apparently less potent and short acting than No. 45, 46 or 47 were.

3) *Adrenergic α-blocking action*

In urethane-morphine anesthetized dogs, benzofuran derivatives in 2 mg/kg (i.v.) produced a temporary fall of blood pressure in 30 to 50 mmHg and recovered to normal range within 5 to 10 minutes after administration. The pressor responses produced by

### Table 4. Adrenolytic effects of benzofuran derivatives on blood pressure of dogs and vas deferens of rats*

| Compound | Depression percent (pressor response in dogs) | Depression potency (contraction of rat's vas deferens) |
|----------|---------------------------------------------|------------------------------------------------------|
|          | -CH₃ | -C₂H₅ | -C₁H₇ (iso) | -CH₃ | -C₂H₅ | -C₁H₇ (iso) |
| X        | 2 mg/kg | 2 mg/kg | 2 mg/kg | 1 x 10⁻⁵ | 1 x 10⁻⁵ | 1 x 10⁻⁵ | 1 x 10⁻⁵ | 1 x 10⁻⁵ |
| 5,7-H    | 42-67 | 56-82 | 42-70 | ± | 0 | ++ | ++ | ++ | + |
| 5-CH₃    | 13-21 | 39-42 | 23-29 | 0 | 0 | + | 0 | + | 0 |
| 5-Cl     | 20-49 | 22-40 | 21-39 | 0 | 0 | 0 | 0 | 0 | 0 |
| 5-OCH₃   | 22-41 | 55-71 | 30-59 | 0 | 0 | + | 0 | 0 | 0 |
| 7-CH₃    | 42-52 | 55-64 | 42-70 | ++ | ++ | ++ | ++ | ++ | ++ |
| 7-Cl     | 31-56 | 65-67 | 29-47 | + | + | + | + | + | + |

* These data were cited from the previous report (4).

**: g/ml

**: 70-100% inhibition, + : 30-70% inhibition, 0: 0-30% inhibition, n=3-5.
10 μg/kg (i.v.) of adrenaline were clearly depressed by all of these benzofuran derivatives at intravenous dose of 2 mg/kg. Concerning to adrenergic β-blocking action, structure activity relationship of these compounds was not so distinct like analgesic and spinal reflex depressing effect of them. However, the compounds which have CH₃, Cl or OCH₃ at 5 position of benzofuran ring were less potent than the others on adrenolytic effect except compound No. 42. Exchange of alkoxy substituents at 3' position in these analogues is seemed to be not affected on their adrenolytic effects. In order to compare with Ilidal, the compounds No. 33, 34, 39, 42 and 46 were tested on adrenolytic activity in pentobarbital and α-chloralose-urethane anesthetized dogs at intravenous doses of 0.5 and 1.0 mg/kg respectively.

The results obtained were shown in Table 5. The compound No. 46 was of most potent in benzofuran derivatives being tested and as potent as the effect of Ilidal. Adrenolytic effects of No. 46 lasted more than 120 minutes. Adrenolytic effects of benzofuran derivatives were not so varied under the different anesthetics, but it was found the tendency to show more potent action under urethane-morphine anesthesia than under pentobarbital or α-chloralose-urethane anesthesia do.

**TABLE 5. Adrenolytic effects of benzofuran derivatives and Ilidal on blood pressure of dogs anesthetized with different anesthetics.**

| Compound No. | Depression percent (pressor response induced by adrenaline) |
|--------------|------------------------------------------------------------|
|              | Urethane-morphine  | Pentobarbital  | α-Chloralose-urethane |
|              | 1.0 mg/kg 0.5 mg/kg | 1.0 mg/kg 0.5 mg/kg | 1.0 mg/kg 0.5 mg/kg |
| No. 33       | 8-39 0-22          | 10-17 5-15     | 0-30 0            |
| No. 34       | 10-44 10-20        | 17-25 0        | 0-24 0            |
| No. 39       | 19-43 7-19         | 11-25 0        | 6-24 0-12         |
| No. 42       | 20-60 16-40        | 16-39 0-10     | 4-26 0-15         |
| No. 46       | 36-67 24-43        | 21-33 21-29    | 29-46 23-36       |
| Ilidal       | 40-70 29-54        | 32-46 15-24    | 64-72 16-32       |

n = 3-5

**DISCUSSION**

In previous papers (4), we reported the result of pharmacological screening of benzofuran derivatives and pointed out that pharmacological properites of 2-(3'-alkoxypropylaminomethyl) 2,3-dihydrobenzofuran analogues extremely resemble to those of 1,4-benzodioxan derivatives which described by Bovet et al. (1), Kulupp et al. (2) and Henatch et al. (3). These benzofuran analogues depressed the spontaneous motility of mice examined by means of photocell method and caused sedative and muscle relaxant effects. They also revealed analgesic action at intraperitoneal dose of 50 to 100 mg/kg when the action was examined by electric stimulating method in mice. The another characteristic effect of these compounds were potent adrenolytic effect and some of them were evaluated to be about as 100 times potent as that of Tolaxolin (2-benzyl-2-imidazoline) when the effect was examined in the isolated vas deference of rats.
On the other hand, Kulupp et al. (2) and Henatch et al. (3) have reported that Quiloflex depressed spinal reflex in α-chloralose anesthetized or spinal cats and showed sedative effect, potentiating effect of barbiturate induced sleep and depressing effect of reticular EEG arousal responses. They also described that Quiloflex showed marked adrenolytic effect, which suppressed pressor response induced by adrenaline or noradrenaline at intravenous dose of 0.5 mg/kg in α-chloralose anesthetized cats.

Therefore, we examined on analgesic, spinal reflex depressing and adrenergic α-blocking effects of 5 or 7 substituted 2-(3'-alkoxypropylaminomethyl) 2,3-dihydrobenzofuran analogues and tried to discuss the structure activity relationships of them respectively.

Regarding to the analgesic effect, the compound No. 33, 39 and 46 which have ethoxy group at 3' position as a alkoxy substituents and have H, CH₃ or Cl at 7 position of benzofuran ring were more potent than the others. Intraperitoneal AD-50 values of these compounds were in 55 to 60 mg/kg and were almost as 3 times potent as that of aminopyrine. Intraperitoneal toxic doses of these benzofuran analogues, however, were in 100 to 200 mg/kg (4) and their safety margin (the ratio of toxic dose to effective dose) were not so large on analgesic effect.

The structure activity relationship of these analogues concerning spinal reflex depressing effect was quite similar to which was found in analgesic effect. Similar relationship had been observed on spontaneous motility depressing effect in mice (4). Spinal reflex depressing effect of the compound No. 46, the most potent one in this analogues, seems to be slightly less potent than that of Quiloflex comparing with the data reported by Kulupp et al. (2). Comparing with mephenesin, however, the compound No. 46 was apparently potent than it.

In the previous report on the pharmacological screening of benzofuran derivatives, we described that adrenolytic effect of 5 or 7 substituted 2-(3'-alkoxypropylaminomethyl) 2,3-dihydrobenzofuran analogues were markedly reduced by replacement with CH₃, Cl or OCH₃ at 5 position and increased by replacement with CH₂ or Cl at 7 position when the effect was examined using rat's vas deference. In present experiments using urethane-morphine anesthetized dogs, however, all of the compounds being tested supressed the pressor responses induced by adrenaline at intravenous dose of 5 mg/kg. Although the structure activity relationship of this action was not so distinct like analgesic and spinal reflex depressing actions, it was also found a tendency that the compounds having H, CH₂ or Cl at 7 position showed relatively potent effect. From the results of further detail experiments on adrenergic α-blocking effect by the use of pentobarbital or α-chloralose-urethane anesthetized dogs, the compounds No. 42 and 46 were evaluated to be as potent as lidal at least.

It was found that the compounds which have potent analgesic effect, e.g. No. 33, 39 and 46, also have potent spinal reflex depressing and adrenolytic effects.

Since many investigators have reported that morphine and the other potent analgesics depressed spinal reflex, particularly polysynaptic reflex (8-11), it was considered that spinal reflex depressing effect of benzofuran analogues might contribute to appearance of anal-
gesic effect of them. In present experiment, the compound which have potent depressing
effect on central nervous system also showed potent adrenolytic effect, and these results
stimulated to the fact that phenothiazine (12-14) and butyrophenone (15, 16) derivatives
widely known as major tranquilizer possess potency adrenolytic and spinal reflex depressing
effects.

Recently, Andén et al. (17, 18) have suggested that chlorpromazine and haloperidol
might block the dopamine and noradrenaline receptors in central nervous system, and
their tranquilizing effect were attributed to their adrenolytic activities in central nervous
system.

Therefore, it would be reasonable to assume that these benzofuran derivatives have
adrenolytic activity in central nervous system like chlorpromazine and haloperidol.

SUMMARY

Analgesic, spinal reflex depressing and adrenergic \(\alpha\)-blocking effects of 5 or 7 sub-
stituted 2-(3'-alkoxypropylaminomethyl) 2.3-dihydrobenzofuran derivatives were investi-
gated. The relationship between the pharmacological activities and the chemical struc-
tures was also discussed.

In general, the compounds which have H, CH\(_3\) or Cl as a substituent at 7 position
on benzofuran ring showed relatively potent analgesic, spinal reflex depressing and ad-
renergic \(\alpha\)-blocking actions, but CH\(_3\), Cl or OCH\(_3\) substitution at 5 position reduced the
pharmacological activities.

While, the compounds which have ethoxy group as a terminal alkoxy group in side
chain of benzofuran ring have more potent actions than methoxy or isopropoxy groups
have.

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