2,2′-Disubstituted Biphenyls: Synthesis and Suppressive Effect against Carbon Tetrachloride-Induced Liver Injury

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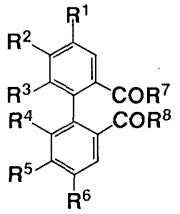
2,2′-Disubstituted biphenyl compounds (1—15) were synthesized by using the Ullmann coupling reaction as a key step. The suppressive effect of these compounds against CCl_4-induced liver injuries in mice was evaluated. An unsymmetrical biphenyl (14f) exhibited the most potent activity. The structure–activity relationship is discussed.

Keywords 2,2′-disubstituted biphenyl; Ullmann coupling reaction; liver-protective activity; carbon tetrachloride-induced liver injury

Lignans of the dibenzo[*a*,*c*]cyclooctene series, *e.g.*, gomisins A, B and C, deoxyschizandrin, and wuweizisu C have recently attracted considerable interest because of their protective activities against liver injury.[1—5] These compounds have a 2,2′-disubstituted biphenyl skeleton which appears to be an essential structure for exhibiting the liver-protective activity. Furthermore, diphenic acid derivatives, represented by dimethyl 4,4′-dimethoxy-5,6,5′,6′,-bis(methylenedioxy)biphenyl-2,2′-dicarboxylate (DDB, **1a**), are also well-known to show potent liver-protective activity.[2,5] Thus, 2,2′-disubstituted biphenyl compounds are particularly attractive candidates for new liver-protective agents. However, due to the difficulties encountered in the synthesis of these compounds, their liver-protective activities have not yet been studied systematically. In the course of our synthetic studies of lignans, we have developed a new method for the synthesis of 2,2′-disubstituted unsymmetrical biphenyls based on the intramolecular Ullmann coupling reaction directed by salicyl alcohol as a template.[6] These synthetic studies prompted us to examine the liver-protective activity of the 2,2′-disubstituted biphenyls systematically. We now report the synthesis of a number of 2,2′-disubstituted biphenyls (**1**—**15**) (Chart 1) and their suppressive effect against CCl_4-induced liver injury.

Chemistry The symmetrical biphenyl compounds (**1a**—**e**) were synthesized by the Ullmann coupling reaction of the corresponding methyl 2-halobenzoate derivatives (**16a**—**e**) according to the reported method[7,8] (Chart 2).

The unsymmetrical biphenyl compounds (**1f**—**i**, **2f**, **4**—**15**) were synthesized from the corresponding cyclic biphenyls (**17**, **18**) which were prepared by the template-directed intramolecular Ullmann coupling reaction developed by us[6] (Chart 2). Thus, the ester exchange reaction of **17f**—**h**[6c] and **17j** took place cleanly under acidic conditions to afford the corresponding diesters (**1f**—**h**, **j**, **2f**) in 85—90% yields. Hydrogenolysis of **1j** by using palladium on charcoal afforded the phenol derivative (**1i**) in 90% yield. The diamide derivatives (**4f**—**6f**) were synthesized by the condensation of amines and **19f**,[6c]



1a–i: R^7=R^8=OMe
2f: R^7=R^8=OEt
3f: R^7,R^8=-O(CH_2)$_3$O-
4f: R^7=R^8=NHEt
5f: R^7=R^8=NEt_2
6f: R^7=R^8=NHC(Me)$_2$$CH_2$OH
7f: R^7=NEt_2, R^8=OEt
8f: R^7=NEt_2, R^8=O-(2-Me-Ph)
9f: R^7=NHC(Me)$_2$$CH_2$OH, R^8=O-(2-Me-Ph)
10f: R^7=NHCH_2CH_2OH, R^8=O-(2-Me-Ph)
11f: R^7=NH(CH_2)$_5$$CH_3$, R^8=O-(2-Me-Ph)
12f: R^7=N⟨⟩NCH_2CH_2OH, R^8=O-(2-Me-Ph)
13f: R^7=N⟨⟩NMe, R^8=O-(2-Me-Ph)
14f: R^7=O-(2-Me-Ph), R^8=NHC(Me)$_2$$CH_2$OH
15f: R^7=O-(2-Me-Ph), R^8=N⟨⟩NCH_2CH_2OH

a: R^1=R^6=OMe, R^2,R^3=R^4,R^5=-OCH_2O-; **b**: R^1=R^6=OMe, R^2=R^3=R^4=R^5=OAc; **c**: R^1=R^2=R^3=R^4=R^5=R^6=OMe; **d**: R^1,R^2=R^5,R^6=-OCH_2O-, R^3=R^4=H; **e**: R^1=R^6=H, R^2,R^3=R^4,R^5=-OCH_2O-; **f**: R^1=H, R^2,R^3=-OCH_2O-, R^4=R^5=R^6=OMe; **g**: R^1=H, R^2,R^3=-OCH_2O-, R^4=R^5=R^6=OMe; **h**: R^1=R^4=H, R^2,R^3=R^5,R^6=-OCH_2O-; **i**: R^1=R^4=H, R^2,R^3=-OCH_2O-, R^5=OH, R^6=OMe; **j**: R^1=R^4=H, R^2,R^3=-OCH_2O-, R^5=OCH_2Ph, R^6=OMe

Chart 1

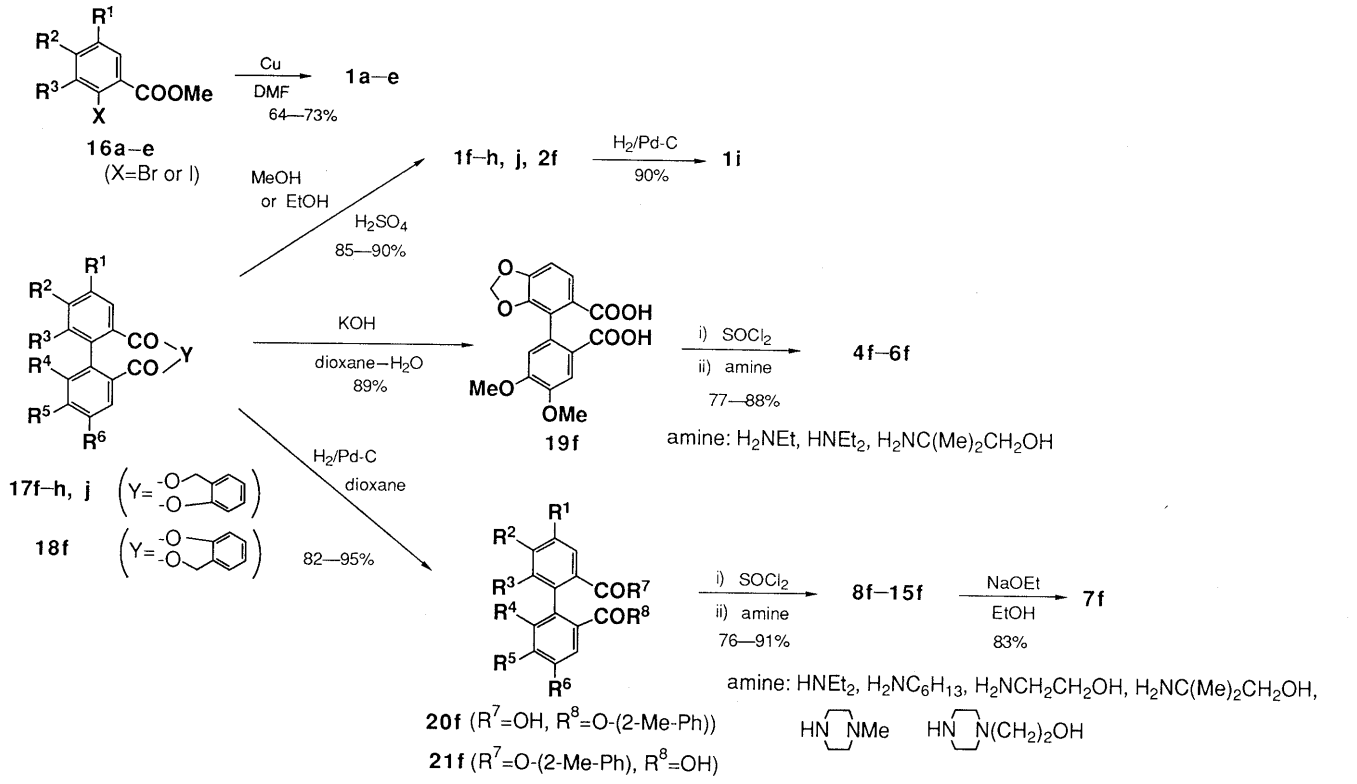


Chart 2

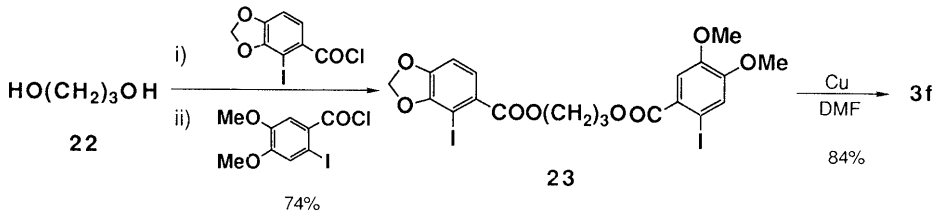


Chart 3

which was prepared by the saponification of the cyclic biphenyl (**18f**).[6c] Furthermore, the monoamide derivatives (**7f**—**15f**) were also synthesized from the cyclic biphenyls (**17f**, **18f**).[6c] Thus, hydrogenolysis of **17f** and **18f** in dioxane using palladium on charcoal proceeded regioselectively to afford the corresponding monoesters (**20f**, **21f**)[6c] in 82—95% yields. Condensations of **20f** and **21f** with a variety of amines were carried out by the usual method to afford the corresponding monoamides (**8f**—**15f**) in 76—91% yields. The ethyl ester (**7f**) was obtained in a good yield by treatment of **8f** with NaOEt in EtOH. Furthermore, the cyclic biphenyl compound (**3f**) was prepared by the intramolecular Ullmann coupling reaction of **23**, which was synthesized by the acylation of **22** (Chart 3).

Biological Results and Discussion The suppressive effect of the 2,2′-disubstituted biphenyls prepared above was evaluated against CCl_4-induced liver injury in mice after oral administration at a dose of 100 mg/kg. The results are summarized in Table I. The activity was evaluated in terms of the suppressive effect of these compounds on the CCl_4-induced elevation of the glutamate pyruvic transaminase (GPT) activity in mice.

We first evaluated the suppressive effect of the biphenyls (**1b**—**i**) having two methoxycarbonyl groups at the 2 and 2′ positions in order to examine the effect of the oxygen functionalities attached to the aromatic rings on the activity. Among the symmetrical biphenyls (**1b**—**e**), only **1e** having methylenedioxy groups at the 5,6 and 5′,6′ positions was active. We next examined the activity of the unsymmetrical biphenyls (**1f**—**i**) having a 5,6-methylenedioxy group on one benzene ring and oxygen functionalities other than the methylenedioxy group on the other benzene ring. We found that all compounds exhibited the activity except for **1i** (having a phenolic hydroxyl group). These results clearly indicate that the biphenyl skeleton having a 5,6-methylenedioxy group on at least one of the two benzene rings is important for exhibiting the activity. Among the unsymmetrical biphenyls, **1g** showed the most potent activity, which was comparable to that of DDB. This result is in marked contrast to that in the case of the cyclic biphenyl lignans, such as gomisin A, in which a methylenedioxy group at the 4,5 or 4′,5′ position is essential for exhibiting the activity.[3]

In order to find compounds having much more potent activity, we further examined the effect of the substituents

Table I. Effect of 2,2′-Disubstituted Biphenyls on CCl_4-Induced Liver Injury in Mice[a]

Compd. No.	Suppressive activity[b]
1a (DDB)	+++
1b	−
1c	−
1d	−
1e	++
1f	+++
1g	++
1h	++
1i	−
2f	++++
3f	−
4f	+++
5f	++++
6f	+++
7f	++
8f	+++
9f	++
10f	+
11f	++
12f	++++
13f	+
14f	++++
15f	++

a) A test compound was orally administered at a dose of 100 mg/kg. *b*) Suppression (%) of the CCl_4-induced elevation of GPT in mice: $-\leq 20$, $20\leq +\leq 40$, $40<++\leq 60$, $60<+++\leq 80$, $80<++++$.

at the 2 and 2′ positions on the activity. Thus, we examined the activity of the diesters (**2f**, **3f**) having ester groups other than methoxycarbonyl groups at the 2 and 2′ positions. Of these compounds, **2f** showed potent activity, while **3f** lacked the activity. It is noteworthy that the activity of **2f** is more potent than that of DDB.

We also examined the activity of the diamide derivatives (**4f**—**6f**). All of them showed activity comparable to or stronger than that of DDB. The 2,2′-bis(*N*,*N*-diethylcarboxamide) derivative (**5f**) showed the most potent activity. These results prompted us to examine the activity of the monoamide (**7f**). However, **7f** unexpectedly showed only weak activity. On the other hand, **8f**, an *o*-tolyl analog of **7f**, showed almost the same activity as that of DDB. We further evaluated **9f**—**15f** having an *o*-tolyloxycarbonyl group and a carbamoyl group. All of these compounds were found to have the activity, and **12f** and **14f** showed very potent activity.

On the basis of these results, we selected **2f**, **5f**, **12f** and **14f** for further evaluation. The results are summarized in Table II. The activity of these compounds was evaluated at doses of 100, 30 and 10 mg/kg. Compound **14f** showed the most potent activity, being more potent than DDB. The acute toxicity of these compounds was also examined. No acute toxicity was detected with compound **14f**. Thus, **14f** is considered to be a potential candidate for a liver-protective agent. Further studies of **14f** are in progress, and the results will be reported elsewhere.

Table II. Suppressive Effect and Acute Toxicity of **1a**, **2f**, **5f**, **12f** and **14f**

Compd. No.	Dose (mg/kg)	Suppression (%) of the CCl_4-induced elevation of GPT	Acute toxicity[a]
2f	10	−2.3	0/3
	30	26.2	
	100	84.7[b]	
5f	10	5.4	1/3
	30	31.6	
	100	90.9[b]	
12f	10	−5.4	3/3
	30	14.9	
	100	97.8[b]	
14f	10	12.2	0/3
	30	61.5[c]	
	100	95.5[b]	
1a (DDB)	10	13.3	0/3
	30	35.3	
	100	74.5[b]	

a) Mortality (number of mice that died/number of mice tested) when a compound was orally administered at a dose of 1000 mg/kg. *b*) Significantly different from control, $p<0.01$. *c*) $p<0.05$.

Experimental

Melting points were determined in open capillary tubes on a Yamato MP-21 melting point apparatus, without correction. Infrared (IR) spectra were obtained using a Perkin Elmer 1640 IR spectrometer. NMR spectra were recorded on a Hitachi R-90 or a Bruker AC-200 instrument using Me_4Si as the internal standard. Mass spectra (MS) were obtained on a Hitachi M-60 or Hitachi M-2000A spectrometer. Thin layer chromatography was carried out on silica gel (Merck type 60H). Dimethyl formamide (DMF), purchased from Katayama Kagaku, was dried over molecular sieves (4 A) and used without further purification. All other solvents were purchased from Katayama Kagaku and used without purification. Copper powder was purchased from Katayama Kagaku and used immediately after purification.[9]

Preparation of the Diesters (1a—e) The diesters (**1a**—**e**) were prepared according to the reported method.[7,8] Compound **1a**: mp 158—159 °C (lit.[5] 154—156 °C). Compound **1b** was prepared from methyl 3,4-bis(acetoxy)-2-bromo-5-methoxybenzoate. Compound **1b**: colorless needles, mp 165—166 °C. IR (KBr): 1788, 1776, 1720, 1336, 1096 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 1.92 (6H, s), 2.25 (6H, s), 3.58 (6H, s), 3.90 (6H, s), 7.52 (2H, s). MS *m/z*: 562 (M^+). *Anal.* Calcd for $C_{26}H_{26}O_{14}$: C, 55.52; H, 4.66. Found: C, 55.46; H, 4.71. Compound **1c**: mp 151—152 °C (lit.[10] 152—153 °C). Compound **1d**: mp 157 °C (lit.[11] 160—160.5 °C). Compound **1e** was prepared from methyl 2-iodo-3,4-methylenedioxybenzoate. Compound **1e**: colorless needles, mp 186 °C. IR (Nujol): 1702 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.64 (6H, s), 5.94 (4H, s), 6.84 (2H, d, J=2 Hz), 7.71 (2H, d, J=2 Hz). MS *m/z*: 358 (M^+). *Anal.* Calcd for $C_{18}H_{14}O_8$: C, 60.34; H, 3.74. Found: C, 60.33; H, 3.75.

17-Benzyloxy-16-methoxy-8*H*-benzo[*c*]-1,3-benzodioxolo[4,5-*e*][1,8]-benzodioxacycloundecin-6,14-dione (17j) 2-Iodo-3,4-methylenedioxybenzoyl chloride (3.1 g, 10 mmol) was added in portions to a solution of salicyl alcohol (1.24 g, 10 mmol), Et_3N (3.34 ml, 24 mmol) and 4-dimethylaminopyridine (12 mg, 0.1 mmol) in dimethylacetamide (50 ml) over a period of 30 min at −30—−20 °C. The reaction mixture was warmed to room temperature and stirred for 6 h at the same temperature, then again cooled to −30—−20 °C. To this mixture was added dropwise a solution of 2-iodo-4-benzyloxy-5-methoxybenzoyl chloride (4.03 g, 10 mmol) in methylene chloride (20 ml) over a period of 30 min at −30—−20 °C. The reaction mixture was warmed to room temperature and stirred for 14 h at the same temperature, then poured into water (300 ml) and extracted with ethyl acetate (3×200 ml). The combined organic layer was washed with aqueous $NaHCO_3$ solution and brine, and dried over $MgSO_4$. The organic layer was evaporated under reduced pressure and the residue was purified by column chromatography on silica gel (ethyl acetate : hexane = 2 : 1) to afford 2-(2-iodo-3,4-methylenedioxybenzoyloxymethyl)phenyl 4-benzyloxy-2-iodo-5-metoxybenzoate (6.1 g, 8.0 mmol): colorless needles, mp 134—136 °C. IR (Nujol): 1740, 1725 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.86 (3H, s), 5.16 (2H, s), 5.36 (2H, s), 6.04 (2H, s), 6.66 (1H, d, J=8 Hz), 7.15—7.65 (11H, m), 7.69 (1H, s). MS *m/z*: 764 (M^+). *Anal.* Calcd for $C_{30}H_{22}I_2O_8$: C, 47.14; H, 2.90; I, 33.21. Found: C, 47.37; H, 2.85; I, 33.09. A solution of the diester

(1.53 g, 2.0 mmol) in DMF (10 ml) was added dropwise over a period of 3 h to refluxing DMF (10 ml) containing activated copper powder[9] (1.27 g, 20 mmol). After the addition was over, the reaction mixture was refluxed for an additional 1 h. It was then cooled and the insoluble materials were filtered off. The filtrate was evaporated to dryness under reduced pressure. The residue was dissolved in ethyl acetate (50 ml) and the solution was washed with water, dried over $MgSO_4$, and evaporated to dryness under reduced pressure. The residue was purified by column chromatography on silica gel (ethyl acetate : hexane = 2 : 1) to afford **17i** (888 mg, 1.74 mmol): a colorless amorphous powder. IR (Nujol): 1750, 1725 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.97 (3H, s), 4.72 (1H, d, J = 11 Hz), 5.0—5.9 (2H, m), 5.75—6.2 (3H, m), 6.69 (1H, d, J = 8.5 Hz), 7.05—7.5 (12H, m). MS m/z: 510 (M^+). *Anal.* Calcd for $C_{30}H_{22}O_8$: C, 70.58; H, 4.34. Found: C, 70.69; H, 4.51.

Preparation of the Diesters (1f—h, j, 2f) A solution of the cyclic biphenyl (**17f**) (870 mg, 2.0 mmol) and sulfonic acid (0.1 ml) in methanol (200 ml) was refluxed for 14 h. The mixture was evaporated to dryness *in vacuo* and the residue was poured into a mixture of water (50 ml) and ethyl acetate (50 ml). The organic layer was separated and the aqueous layer was further extracted with ethyl acetate (100 ml). The combined organic layer was washed with water, and dried over $MgSO_4$. The solvent was evaporated off *in vacuo*. The residue was crystallized from diisopropyl ether to afford **1f** (670 mg, 1.8 mmol): colorless needles, mp 121 °C. IR (KBr): 1720, 1600 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.59 (3H, s), 3.65 (3H, s), 3.86 (3H, s), 3.94 (3H, s), 5.94 (2H, s), 6.67 (1H, s), 6.82 (1H, d, J = 8 Hz), 7.60 (1H, s), 7.65 (1H, d, J = 8 Hz). MS m/z: 374 (M^+). *Anal.* Calcd for $C_{19}H_{18}O_8$: C, 60.96; H, 4.85. Found: C, 60.92; H, 4.86.

Compounds **1g**, **h**, **j** and **2f** were prepared by the same procedure as described above. Compound **1g**: colorless needles, mp 115 °C. IR (KBr): 1722 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.61 (9H, s), 3.92 (6H, s), 5.93 (2H, s), 6.82 (1H, d, J = 8 Hz), 7.38 (1H, s), 7.68 (1H, d, J = 8 Hz). MS m/z: 404 (M^+). *Anal.* Calcd for $C_{20}H_{20}O_9$: C, 59.41; H, 4.99. Found: C, 59.38; H, 4.89. Compound **1h**: colorless needles, mp 139—141 °C. IR (KBr): 1720, 1718 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.64 (6H, s), 5.94 (2H, s), 6.04 (2H, s), 6.64 (1H, s), 6.79 (1H, d, J = 8 Hz), 7.51 (1H, s), 7.64 (1H, d, J = 8 Hz). MS m/z: 358 (M^+). *Anal.* Calcd for $C_{18}H_{14}O_8$: C, 60.34; H, 3.94. Found: C, 60.41; H, 3.89. Compound **1j**: colorless needles, mp 118—119 °C. IR (KBr): 1705, 1695 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.53 (3H, s), 3.65 (3H, s), 3.95 (3H, s), 5.13 (2H, s), 5.95—6.15 (2H, m), 6.70 (1H, s), 6.79 (1H, d, J = 8 Hz), 7.2—7.5 (5H, m), 7.60 (1H, s), 7.61 (1H, d, J = 8 Hz). MS m/z: 450 (M^+). *Anal.* Calcd for $C_{25}H_{22}O_8$: C, 66.66; H, 4.92. Found: C, 66.77; H, 5.01. Compound **2f**: colorless needles, mp 82—83 °C. IR (Nujol): 1722 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 1.01 (3H, t, J = 7 Hz), 1.03 (3H, t, J = 7 Hz), 3.88 (3H, s), 3.96 (3H, s), 4.01 (2H, q, J = 7 Hz), 4.10 (2H, q, J = 7 Hz), 5.95 (2H, s), 6.68 (1H, s), 6.84 (1H, d, J = 8 Hz), 7.61 (1H, s), 7.65 (1H, d, J = 8 Hz). MS m/z: 402 (M^+). *Anal.* Calcd for $C_{21}H_{22}O_8$: C, 62.68; H, 5.51. Found: C, 62.39; H, 5.66.

Dimethyl 5-Hydroxy-4-methoxy-5′,6′-methylenedioxybiphenyl-2,2′-dicarboxylate (1i) Hydrogenolysis of **1j** (900 mg, 2.0 mmol) using 10% palladium on charcoal (100 mg) was carried out in dioxane (50 ml) for 5 h under a hydrogen atmosphere (2.0 kg/cm^2). The insoluble materials were filtered off and the filtrate was evaporated to dryness *in vacuo*. The residue was crystallized from diisopropyl ether to give **1i** (650 mg, 1.8 mmol): colorless needles, mp 172—173 °C. IR (KBr): 3400, 1710 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.61 (1H, s), 3.64 (6H, s), 3.96 (3H, s), 5.93 (2H, s), 6.75 (1H, s), 6.80 (1H, d, J = 7 Hz), 7.59 (1H, s), 7.63 (1H, d, J = 7 Hz). MS m/z: 360 (M^+). *Anal.* Calcd for $C_{18}H_{16}O_8$: C, 60.00; H, 4.48, Found: C, 60.02; H, 4.46.

Preparation of Diamides (4—6) $SOCl_2$ (2.4 g, 20 mmol) and DMF (1 drop) were added to a solution of **19f**[6c] (690 mg, 2.0 mmol) in dioxane (10 ml), and the mixture was refluxed for 0.5 h, then evaporated to dryness *in vacuo*. The residue was taken up in toluene (20 ml) and the solution was evaporated to dryness *in vacuo*. The residue were dissolved in methylene chloride (20 ml) containing 4-dimethylaminopyridine (490 mg, 4.0 mmol), then ethylamine (230 mg, 5.0 mmol) was added dropwise at 0—5 °C. The mixture was stirred for 14 h at room temperature, and washed with 5% aqueous citric acid solution and water. The organic layer was dried over $MgSO_4$ and the mixture was evaporated to dryness *in vacuo*. The residue was crystallized from diisopropyl ether to afford **4f** (680 mg, 1.7 mmol): colorless needles, mp 196—197 °C. IR (Nujol): 3280, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.90 (3H, t, J = 6 Hz), 0.93 (3H, t, J = 6 Hz), 3.0—3.45 (4H, m), 3.81 (3H, s), 3.91 (3H, s), 5.89 (2H, s), 6.60 (1H, s), 6.7—7.2 (5H, m). MS m/z: 400 (M^+). *Anal.* Calcd for $C_{21}H_{24}N_2O_6$: C, 62.99; H, 6.04; N, 7.00. Found: C, 62.78; H, 6.23; N, 6.81.

The diamides (**5f**, **6f**) were prepared from **19f** by the same procedure as described above. Compound **5f**: colorless needles, mp 148—149 °C. IR (Nujol): 1630, 1615 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.85 (3H, t, J = 7 Hz), 0.89 (3H, t, J = 7 Hz), 1.17 (6H, t, J = 7 Hz), 2.8—3.9 (8H, m), 3.89 (6H, s), 5.8—5.95 (2H, m), 6.7—7.0 (3H, m), 7.13 (1H, s). MS m/z: 456 (M^+). *Anal.* Calcd for $C_{25}H_{32}N_2O_6$: C, 65.77; H, 7.07; N, 6.15. Found: C, 65.64; H, 6.92; N, 5.93. Compound **6f**: colorless needles, mp 182—184 °C. IR (Nujol): 3280, 1640 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 1.05 (6 H, s), 1.12 (6H, s), 3.4 (4H, br s), 3.83 (3H, s), 3.93 (3H, s), 4.40 (1H, br s), 4.65 (1H, br s), 5.93 (2H, s), 6.7—7.2 (6H, m). *Anal.* Calcd for $C_{25}H_{32}N_2O_8$: C, 61.46; H, 6.60; N, 5.73. Found: C, 61.52; H, 6.71; N, 5.49.

Preparation of the Monoamides (8—15) The monoamides (**8f—15f**) were prepared from the corresponding carboxylic acids (**20f**, **21f**) by the same procedure as described above. Compound **8f**: colorless needles, mp 145—146 °C. IR (Nujol): 1705, 1640 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.79 (3H, t, J = 6.8 Hz), 0.83 (3H, t, J = 6.8 Hz), 2.22 (3H, s), 2.7—3.15 (2H, m), 3.2—3.75 (2H, m), 3.95 (3H, s), 4.00 (3H, s), 5.90 (2H, s), 6.80 (1H, d, J = 7.9 Hz), 6.85 (1H, d, J = 7.9 Hz), 6.95—7.3 (5H, m), 7.76 (1H, s). MS m/z: 491 (M^+). *Anal.* Calcd for $C_{28}H_{29}NO_7$: C, 68.41; H, 5.95; N, 2.85. Found: C, 68.16; H, 5.91; N, 3.15. Compound **9f**: colorless needles, mp 177—181 °C. IR (Nujol): 1705, 1640 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.84 (3H, s), 0.99 (3H, s), 2.17 (3H, s), 3.2—3.6 (3H, m), 3.90 (3H, s), 4.01 (3H, s), 4.9—6.3 (3H, m), 6.7—7.35 (7H, m), 7.69 (1H, s). *Anal.* Calcd for $C_{28}H_{29}NO_8$: C, 66.26; H, 5.76; N, 2.76. Found: C, 66.27; H, 5.8; N, 2.63. Compound **10f**: colorless needles, mp 148—150 °C. IR (Nujol): 3370, 1695, 1660 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 2.16 (3H, s), 2.1—2.3 (1H, m), 3.1—3.5 (4H, m), 3.90 (3H, s), 3.99 (3H, s), 5.90—5.95 (2H, m), 6.6 (1H, br s), 6.77 (1H, d, J = 8.5 Hz), 6.80 (1H, s), 6.9—7.3 (5H, m), 7.69 (1H, s). *Anal.* Calcd for $C_{26}H_{25}NO_8$: C, 65.13; H, 5.26; N, 2.92. Found: C, 65.33; H, 5.30; N, 2.84. Compound **11f**: colorless needles, mp 117—118 °C. IR (Nujol): 3400, 1735, 1660 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.7—1.4 (11H, m), 2.16 (3H, s), 2.8—3.4 (2H, m), 3.89 (3H, s), 3.98 (3H, s), 5.91 (2H, s), 6.0 (1H, br s), 6.7—7.3 (7H, m), 7.67 (1H, s). MS m/z: 519 (M^+). *Anal.* Calcd for $C_{30}H_{33}NO_7$: C, 69.34; H, 6.40; N, 2.69. Found: C, 69.50; H, 6.41; N, 2.65. Compound **12f**: colorless needles, mp 92—95 °C. IR (Nujol): 3400, 1730, 1600 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 2.1—2.6 (5H, m), 2.22 (3H, s), 2.8—3.7 (8H, m), 3.91 (3H, s), 4.00 (3H, s), 5.90 (2H, s), 6.7—7.3 (7H, m), 7.73 (1H, s). *Anal.* Calcd for $C_{30}H_{32}N_2O_8$: C, 65.68; H, 5.88; N, 5.11. Found: C, 65.42; H, 5.97; N, 4.86. Compound **13f**: colorless needles, mp 156—158 °C. IR (Nujol): 1745, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 1.5—2.4 (4H, m), 2.12 (3H, s), 2.21 (3H, s), 3.0—3.7 (4H, m), 3.90 (3H, s), 3.97 (3H, s), 5.88 (2H, s), 6.0—7.35 (6H, m), 7.66 (1H, s), 7.99 (1H, s). MS m/z: 518 (M^+). *Anal.* Calcd for $C_{29}H_{30}N_2O_7$: C, 67.17; H, 5.83; N, 5.40. Found: C, 67.45; H, 5.88; N, 5.29. Compound **14f**: colorless needles, mp 155—156 °C. IR (Nujol): 3320, 1700, 1660 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.91 (3H, s), 1.00 (3H, s), 2.12 (3H, s), 3.2—3.65 (3H, m), 3.84 (3H, s), 3.90 (3H, s), 4.85—5.1 (1H, m), 6.00 (2H, s), 6.47 (1H, br s), 6.7—7.35 (6H, m), 7.73 (1H, d, J = 8 Hz). MS m/z: 507 (M^+). *Anal.* Calcd for $C_{28}H_{29}NO_8$: C, 66.26; H, 5.76; N, 2.76. Found: C, 66.54; H, 5.58; N, 2.54. Compound **15f**: colorless needles, mp 102—103 °C. IR (Nujol): 3400, 1720, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 2.0—2.6 (5H, m), 2.17 (3H, s), 3.05—3.7 (8H, m), 3.85 (3H, s), 3.88 (3H, s), 6.00 (1H, d, J = 0.9 Hz), 6.04 (1H, d, J = 0.9 Hz), 6.7—7.3 (7H, m), 7.86 (1H, d, J = 8.3 Hz). MS m/z: 548 (M^+). *Anal.* Calcd for $C_{30}H_{32}N_2O_8$: C, 65.68; H, 5.88; N, 5.11. Found: C, 65.66; H, 5.84; N, 5.22.

Ethyl 2-(*N*,*N*-Diethylcarbamoyl)-4,5-dimethoxy-5′,6′-methylenedioxybiphenyl-2-carboxylate (7f) Compound **8f** (750 mg, 2.0 mmol) was added to a solution of sodium ethoxide (50 mg) in ethanol (20 ml), and the mixture was stirred at room temperature for 4 h, then poured into 5% aqueous citric acid solution. The whole was extracted with ethyl acetate (2 × 100 ml). The combined organic layer was washed with water, dried over $MgSO_4$, and evaporated *in vacuo*. The residue was crystallized from ether to afford **7f** (710 mg, 1.7 mmol): colorless needles, mp 86—87 °C. IR (Nujol): 1715, 1640 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.82 (6H, t, J = 7.1 Hz), 1.14 (3H, t, J = 7.1 Hz), 2.65—3.1 (2H, m), 3.25—3.65 (2H, m), 3.91 (3H, s), 3.95 (3H, s), 4.19 (2H, br q, J = 7.1 Hz), 5.86 (1H, d, J = 1.1 Hz), 5.94 (1H, d, J = 1.1 Hz), 6.85 (1H, d, J = 8.0 Hz), 6.89 (1H, d, J = 8.0 Hz), 7.26 (1H, s), 7.50 (1H, s). MS m/z: 429 (M^+). *Anal.* Calcd for $C_{23}H_{27}NO_7$: C, 64.32; H, 6.34; N, 3.26. Found: C, 64.13; H, 6.35; N, 3.17.

1-(2-Iodo-4,5-dimethoxybenzoyloxy)-3-(2-iodo-3,4-methylenedioxy-

benzoyloxy)propane (23) 2-Iodo-3,4-methylenedioxybenzoyl chloride (3.1 g, 10 mmol) was added to a solution of **22** (7.6 g, 100 mmol), Et_3N (3.34 ml, 24 mmol) and 4-dimethylaminopyridine (12 mg, 0.1 mmol) in methylene chloride (50 ml) at 5 °C and the mixture was stirred at room temperature for 14 h, then washed with water, saturated aqueous $NaHCO_3$ solution and brine. The organic layer was dried over $MgSO_4$ and evaporated to dryness *in vacuo*. The residue was dissolved in methylene chloride (50 ml) containing triethylamine (3.34 ml, 24 mmol) and 4-dimethylaminopyridine (12 mg, 0.1 mmol). To this solution was added 2-iodo-4,5-dimethoxybenzoyl chloride (3.3 g, 10 mmol) at 5 °C, and the mixture was stirred at room temperature for 14 h, then washed with water, saturated aqueous $NaHCO_3$ solution and brine, and dried over $MgSO_4$. The solvent was evaporated to dryness *in vacuo* and the residue was purified by column chromatography on silica gel (ethyl acetate : hexane = 2 : 1) to give **23** (4.7 g, 7.4 mmol): colorless needles, mp 132—133 °C. IR (Nujol): 1720 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 2.28 (2H, q, J = 6 Hz), 3.82 (3H, s), 3.88 (3H, s), 4.52 (4H, t, J = 6 Hz), 6.10 (2H, s), 6.74 (1H, d, J = 8.5 Hz), 7.38 (1H, s), 7.43 (1H, s), 7.55 (1H, d, J = 8.5 Hz). MS *m/z*: 640 (M^+). *Anal.* Calcd for $C_{20}H_{18}I_2O_8$: C, 37.52; H, 2.83. Found: C, 37.59; H, 2.77.

8,9-Dihydro-13,14-dimethoxy-1,2-methylenedioxy-7*H*-dibenzo[*g*,*i*]-[1,5]dioxacycloundecine-5,11-dione (3f) A solution of the diester **23** (1.17 g, 2.0 mmol) in DMF (10 ml) was added dropwise over a period of 3 h to refluxing DMF (10 ml) containing activated copper powder (1.27 g, 20 mmol). After the addition was over, the reaction mixture was refluxed for an additional 1 h, then cooled, and the insoluble materials were filtered off. The filtrate was evaporated to dryness *in vacuo*. The residue was dissolved in ethyl acetate (50 ml) and the solution was washed with water dried over $MgSO_4$. After the solvent had been removed *in vacuo*, the residue was purified by column chromatography on silica gel (hexane : chloroform : ethylacetate = 3 : 3 : 1) to afford **3f** (650 mg, 1.68 mmol) as colorless needles, mp 197—199 °C. IR (Nujol): 1725 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 2.0—2.4 (2H, m), 3.9—4.35 (2H, m), 3.90 (6H, s), 4.6—4.95 (2H, m), 5.9—6.1 (2H, m), 6.76 (1H, d, J = 8 Hz), 6.95 (1H, s), 7.16 (1H, d, J = 8 Hz), 7.19 (1H, s). MS *m/z*: 386 (M^+). *Anal.* Calcd for $C_{20}H_{18}O_8$: C, 62.17; H, 4.70. Found: C, 62.19; H, 4.74.

Evaluation of Biological Activity Groups of 3 ddY male mice, weighing 25—30 g, were used. Test compounds were orally administered to mice at a dose of 100, 30 or 10 mg/kg, 3 h before oral administration of 75 ml/kg of CCl_4. Mice were starved for 24 h after the injection of CCl_4, then killed, and blood samples were collected from the abdominal aorta. The plasma GPT activity was measured according to the method of Reimann and Frankel.[12] The suppressive effect of each compound against CCl_4-induced liver injury was evaluated in terms of the suppression (%) of elevation of GPT activity. The suppression (%) was calculated from the following equation (Tables I and II).

$$\text{suppression (\%) of elevation of GPT activity} = \{1-(A-B)/(C-B)\} \times 100$$

where A is the mean GPT activity of the test compound group, B is the mean GPT activity of the normal control group, and C is the mean GPT activity of the CCl_4-treated control group.

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