

137. Antiplatelet α -Methylidene- γ -butyrolactones: Synthesis and Evaluation of Quinoline, Flavone, and Xanthone Derivatives

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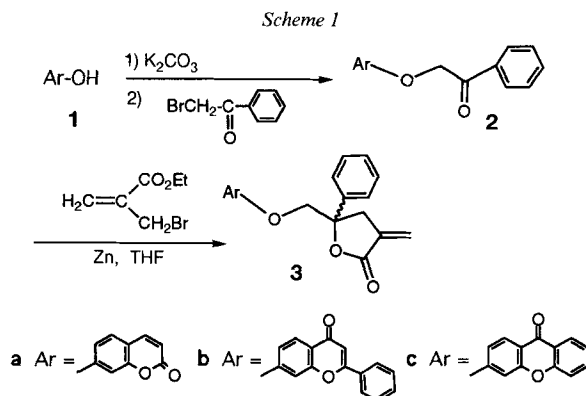
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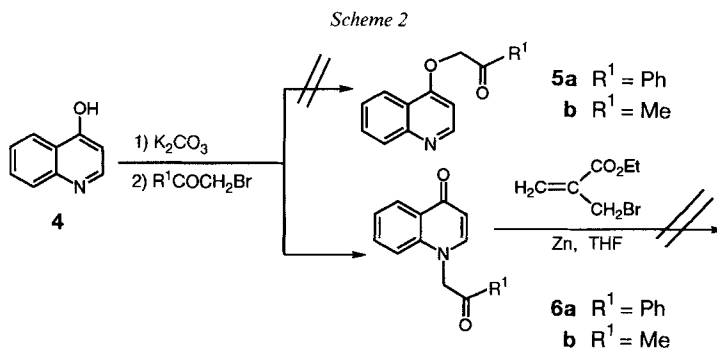
As a continuation of our previous studies on the synthesis and antiplatelet activity of coumarin derivatives of α -methylidene- γ -butyrolactones, certain quinoline, flavone, and xanthone derivatives were synthesized and evaluated for antiplatelet activity against thrombin (Thr)-, arachidonic acid (AA)-, collagen (Col)-, and platelet-activating factor (PAF)-induced aggregation in washed rabbit platelets. These compounds were synthesized from quinolin-8-ol, flavon-7-ol, and xanthon-3-ol, respectively, *via* alkylation and *Reformatsky*-type condensation (Schemes 1–3). By the comparison with coumarin α -methylidene- γ -butyrolactone **3a**, flavone and xanthone derivatives, **3b** and **3c**, respectively, are more selective in which only AA- and collagen-induced aggregation are strongly inhibited. Most of the quinoline derivatives (**9a–e**) exhibited broad-spectrum antiplatelet activities.

Introduction. – A number of natural products bearing an α -methylidene- γ -butyrolactone functionality exhibit wide-ranging biological activities which include antitumoral, bactericidal, fungicidal, and anthelmintic properties [1–5]. Recently, we have synthesized certain coumarins and naphthalenes containing α -methylidene- γ -butyrolactones as potential antiplatelet agents [6] [7]. Among them, 7-[(2,3,4,5-tetrahydro-4-methylidene-5-oxo-2-phenylfuran-2-yl)methoxy]-2*H*-1-benzopyran-2-one (**3a**) and its 4-substituted counterpart have shown potent and broad-spectrum antiplatelet effects in which thrombin (Thr)-, arachidonic acid (AA)-, collagen (Col)-, and platelet-activating factor (PAF)-induced aggregation of rabbit platelets were inhibited. To determine the effect of structural modification with respect to the optimal antiplatelet activity, we have synthesized analogs of **3a** in which coumarin (= 2*H*-1-benzopyran-2-one) moiety was replaced by flavone (= 4*H*-1-benzopyran-4-one) and xanthone (= 9*H*-xanthen-9-one) to increase the lipophilic property of α -methylidene- γ -butyrolactones and thus increase their penetration into the target sites of platelets. The flavonoids are a ubiquitous family of phytochemicals that possess a wide variety of biological activities. A number of natural and synthetic xanthenes were also found to exhibit antiplatelet activity [8–11]. On the other hand, the hydrophilic analogs of **3a** were synthesized in which coumarin was replaced by quinoline, a constituent of many therapeutic drugs such as antimalarial agents. Their antiplatelet structure-activity relationships are also discussed.

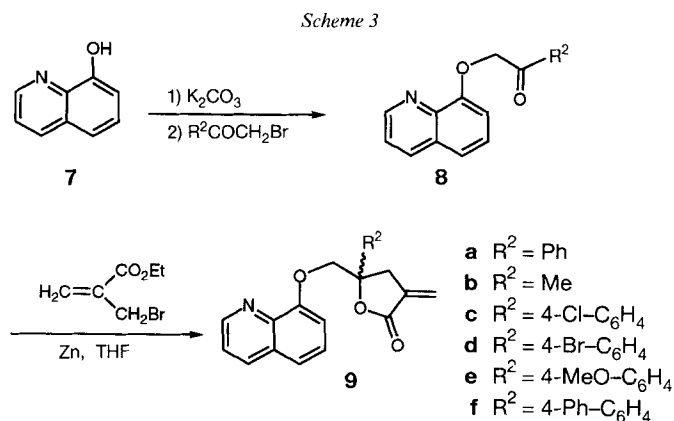
Results and Discussion. – The preparation of aryl- α -methylidene- γ -butyrolactones **3a–c** is illustrated in *Scheme 1*. Alkylation of **1a–c** with 2-bromoacetophenone under basic conditions afforded 2-(aryloxy)acetophenones **2a–c** which were then reacted with ethyl 2-(bromomethyl)acrylate and Zn powder in dry THF (*Reformatsky*-type condensation) to give the target compounds **3a–c** in 47–75% overall yield.



Since 4- and 8-substituted quinolines constitute many chemotherapeutic drugs such as antimalarial agents, chloroquine and primaquine, respectively, both quinolin-4-ol (**4**) and quinolin-8-ol (**7**) were chosen as starting materials. Compound **4** was treated with K_2CO_3 and 2-bromoacetophenone in dry DMF (*Scheme 2*). The product thus obtained was 1-(2-oxo-2-phenylethyl)quinolin-4-one (**6a**) but not 4-(2-oxo-2-phenylethoxy)quinoline (**5a**) based on the 1H , ^{13}C heteronuclear-correlation (HETCOR) NMR experiments. The singlet $C(1')H_2$ protons (δ at 6.06 ppm) were clearly coupled to C-atoms with resonances of 140.69 (3J), 145.24 (3J), and 193.37 (2J) corresponding to C(8a), C(2), and C(2'), respectively. Besides aromatic protons resonated at 7.34–8.25 ppm, there are two doublet peaks ($J = 7.6$ Hz) appeared at 6.15 and 7.95 ppm. The downfield peak (δ 7.95) in coupling with C(8a) (140.69, 3J), C(2) (145.24, 1J), and C(4) (176.62, 3J) was assigned to H–C(2) and the upfield peak (δ 6.15) assigned to H–C(3). The *N*- but not *O*-alkylation was also observed when the alkylating agent was bromoacetone, and 1-(2-oxo-propyl)quinolin-4-one (**6b**) was obtained. Attempts to achieve the *Reformatsky*-type



condensation of **6** under different reaction conditions were not successful. However, treatment of **7** with K_2CO_3 and 2-bromoacetophenone in dry THF afforded 8-(2-oxo-2-phenylethoxy)quinoline (**8a**), an O-alkylated product (Scheme 3). The structure of **8a** was determined through the 1H , ^{13}C heteronuclear-correlation NMR experiments in which the singlet $C(1')H_2$ protons (δ at 5.65 ppm) were clearly coupled to C-atoms with resonances of 153.82 (3J) and 194.43 (2J) corresponding to C(8) and C(2'), respectively. Reformatsky-type condensation of **8a** gave the desired 8-[(2,3,4,5-tetrahydro-4-methylidene-5-oxo-2-phenylfuran-2-yl)methoxy]quinoline (**9a**) in 75% yield. Accordingly, compounds **9b–f** were prepared from **7** via O-alkylation and Reformatsky-type condensation in good overall yield.



The antiplatelet activities of α -methylidene- γ -butyrolactones were evaluated in washed rabbit platelets. Platelet aggregation was induced by thrombin (Thr, 0.1 U/ml), arachidonic acid (AA, 100 μM), collagen (Col, 10 $\mu g/ml$), and platelet-activating factor (PAF, 2 nM). The final concentration of test compounds was 100 $\mu g/ml$, and the results are shown in Table 1. Compound **3a** inhibited the platelet aggregation caused by four inducers, while its flavone and xanthone analogs, **3b** and **3c**, respectively, inhibited AA- and Col-induced platelet aggregation completely, and those by thrombin and PAF partially. The compound **9a** and its 2-Me counterpart **9b** exhibited potent and broad-spectrum antiplatelet effects in which Thr-, AA-, Col-, and PAF-induced platelet aggregation were inhibited. Significant antiplatelet activities were also observed when the 2-Ph group of **9a** was replaced by a 4-Cl-, 4-Br-, or 4-MeO-substituted phenyl group (see **9c–e**). However, a 4-Ph-substituted phenyl substituent (**9f**) decreased antiplatelet spectrum in which only AA- and Col-induced aggregation were inhibited. The inhibitory concentrations for 50% aggregation (IC_{50}) induced by AA and PAF are given in Table 2. The coumarin derivative **3a** showed the most significant antiplatelet effects on AA- and PAF-induced platelet aggregation, while its flavone and xanthone counterparts, **3b** and **3c**, respectively, were more selective. The quinoline derivative **9a** possessing a 2-Ph substituent was more active than its 2-Me counterpart **9b**. The poor inhibitory potency of **9f** against PAF-induced platelet aggregation implies that a bulky substituent at aromatic benzene reduced its antiplatelet potency. In contrary, **9a** and **9c** are two of the best.

Table 1. Effect of α -Methylidene- γ -butyrolactones on the Platelet Aggregation Induced by Thrombin (Thr), Arachidonic Acid (AA), Collagen (Col), and Platelet-Activating Factor (PAF) in Washed Rabbit Platelets^{a)}

	Aggregation [%]			
	Thr (0.1 U/ml)	AA (100 μ M)	Col (10 μ g/ml)	PAF (2nM)
Control	92.8 $\pm$ 1.5	87.2 $\pm$ 1.0	88.8 $\pm$ 1.5	90.3 $\pm$ 1.6
3a	0 ^{b)} ^{c)}	0	0	0
3b	65.6 $\pm$ 9.3 ^{d)}	0	0	36.8 $\pm$ 4.2 ^{c)}
3c	81.7 $\pm$ 3.7 ^{d)}	0	0	70.3 $\pm$ 4.1 ^{c)}
9a	0	0	0	0
9b	17.4 $\pm$ 7.9 ^{c)}	0	0	0
9c	0	0	0	0
9d	0	0	0	0
9e	0	0	0	0
9f	70.5 $\pm$ 4.1 ^{c)}	0	0	78.0 $\pm$ 1.6 ^{c)}
Aspirin	91.9 $\pm$ 1.4	0	85.4 $\pm$ 3.9	90.5 $\pm$ 1.2

^{a)} Platelets were preincubated with DMSO (0.5%, control), aspirin (10 μ g/ml), or α -methylidene- γ -butyrolactones (100 μ g/ml) at 37° for 3 min, and the inducer was then added. Percentages of aggregation are presented as means $\pm$ standard errors of the mean ($n = 3-7$).

^{b)} Complete inhibition in all experiments.

^{c)} Significantly different from control value at $p < 0.001$.

^{d)} Significantly different from control value at $p < 0.01$.

Table 2. IC₅₀ Values (μ M) of α -Methylidene- γ -butyrolactones on the Platelet Aggregation Induced by AA and PAF

	AA	PAF
3a	3.7	16.4
3b	6.1	> 200
3c	18.5	> 200
9a	14.2	33.6
9b	88.9	177.9
9c	17.3	33.9
9d	19.1	60.0
9e	41.9	128.3
9f	25.4	> 200

As a result of these studies, compound **3a** was found to possess broad and potent *in vitro* antiplatelet activity. Its lipophilic flavone and xanthone analogs, **3b** and **3c**, respectively, were less active in potency but were more selective. The hydrophilic quinoline analogs **9a-f** were also less active than **3a** in the inhibitory potency against platelet aggregation induced by Thr, AA, Col, and PAF.

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Experimental Part

General. TLC: precoated (0.2 mm) silica gel 60 F-254 plates from EM Laboratories, Inc.; detection by UV light (254 nm). M.p.: YANACO micromelting-point apparatus; uncorrected. UV Spectra ($\lambda_{\max}$ (log ϵ) in nm): Beckman UV-VIS spectrophotometer. ¹H- and ¹³C-NMR spectra: Varian-Gemini-200 spectrometer, chemical shifts δ in ppm with Me₄Si as an internal standard. Elemental analyses were carried out on a Heraeus CHN-O-Rapid elemental analyzer and results were within $\pm 0.4\%$ of theoretical values.

7-[2-Oxo-2-phenylethoxy]-2-phenyl-4H-1-benzopyran-4-one (**2b**). Flavon-7-ol (**1b**) (1.19 g, 5 mmol), K_2CO_3 (0.69 g, 5 mmol), and dry DMF (40 ml) were stirred at r.t. for 30 min. To this soln. was added 2-bromoacetophenone (1.00 g, 5 mmol) in dry DMF (10 ml) in one portion. The resulting mixture was continued to stir at r.t. for 12 h (TLC monitoring) and then poured into ice-water (100 ml). The pale-yellow solid thus obtained was collected and crystallized from CH_2Cl_2/Et_2O 1:10: **2b** (0.99 g, 73%). Pale-yellow needle crystals. M.p. 174–175°. 1H -NMR ($CDCl_3$): 5.45 (s, 2 H—C(1')); 6.75 (s, H—C(3)); 6.98–8.17 (m, 13 arom. H). ^{13}C -NMR ($CDCl_3$): 70.77 (C(1')); 101.78, 109.59, 114.48, 118.56, 126.21, 127.40, 128.12, 128.47, 128.68, 129.03, 131.47, 134.19, 134.27, 157.77, 162.47, 163.18 (arom. C); 177.71 (C(4)); 193.12 (C(2')). Anal. calc. for $C_{23}H_{16}O_4$: C 77.51, H 4.53; found: C 77.15, H 4.57.

3-(2-Oxo-2-phenylethoxy)-9H-xanthen-9-one (**2c**). Prepared by the same procedure as described for **2b**: 97% yield. M.p. 174–175°. 1H -NMR ($CDCl_3$): 5.43 (s, 2 H—C(1')); 6.87–8.33 (m, 12 arom. H). ^{13}C -NMR ($CDCl_3$): 70.74 (C(1')); 101.51, 113.31, 116.52, 117.72, 121.94, 123.96, 126.68, 128.11, 128.61, 129.04, 134.22, 134.26, 134.42, 156.21, 157.84, 163.32 (arom. C); 176.21 (C(9)); 193.10 (C(2')). Anal. calc. for $C_{21}H_{14}O_2 \cdot 0.2 H_2O$: C 75.52, H 4.35; found: C 75.78, H 4.31.

2-Phenyl-7-[(2,3,4,5-tetrahydro-4-methylidene-5-oxo-2-phenylfuran-2-yl)methoxy]-4H-1-benzopyran-4-one (**3b**). To a soln. of **2b** (1.07 g, 3 mmol) in dry THF (60 ml) were added activated Zn powder (0.26 g, 3.9 mmol), hydroquinone (6 mg), and ethyl 2-(bromomethyl)acrylate (0.78 g, 4 mmol). The mixture was refluxed under N_2 atmosphere for 6 h (TLC monitoring). After cooling, it was poured into an ice-cold 5% HCl soln. (300 ml) and extracted with CH_2Cl_2 (3 $\times$ 60 ml). The CH_2Cl_2 extracts were combined and washed with H_2O , dried (Na_2SO_4), and then evaporated to give a brown solid which was purified by column chromatography on silica gel using CH_2Cl_2 to afford a residual solid which was crystallized from CH_2Cl_2/Et_2O 1:10: **3b** (0.82 g, 64%). M.p. 190–191°. UV: 250 (4.25), 305 (4.34; 0.1N HCl/MeOH); 251 (4.23), 305 (4.33) (MeOH); 305 (4.35; 0.1N NaOH/MeOH). 1H -NMR ($CDCl_3$): 3.26 (dt, $J = 17.0, 2.9$, 1 H—C(3')); 3.69 (dt, $J = 17.0, 2.5$, 1 H—C(3')); 4.27 (AB type, CH_2O); 5.73 (t, $J = 2.5$, 1 H, $CH_2=C(4')$); 6.34 (t, $J = 2.8$, 1 H, $CH_2=C(4')$); 6.75 (s, H—C(3)); 6.91–8.14 (m, 13 arom. H). ^{13}C -NMR ($CDCl_3$): 37.33 (C(3')); 74.56 (CH_2O); 83.78 (C(2')); 101.63, 107.57, 114.36, 118.53, 122.03, 125.08, 126.16, 127.36, 128.78, 128.93, 129.02, 131.51, 131.72, 134.58, 139.91, 157.71, 162.37, 163.15 (arom. C); 169.05 (C(5')); 177.68 (C(4)). Anal. calc. for $C_{27}H_{20}O_5$: C 76.40, H 4.75; found: C 76.13, H 4.80.

3-[(2,3,4,5-Tetrahydro-4-methylidene-5-oxo-2-phenylfuran-2-yl)methoxy]-9H-xanthen-9-one (**3c**). Prepared by the same procedure as described for **3b**: 77% yield. M.p. 160–161°. UV: 235 (sh, 4.62), 265 (4.05), 298 (4.19; 0.1N HCl/MeOH); 235 (sh, 4.64), 266 (4.08), 297 (4.22; MeOH); 265 (4.27), 298 (4.25; 0.1N NaOH/MeOH). 1H -NMR ($CDCl_3$): 3.25 (dt, $J = 16.9, 2.9$, 1 H—C(3')); 3.68 (dt, $J = 16.9, 2.5$, 1 H—C(3')); 4.26 (AB type, CH_2O); 5.72 (t, $J = 2.6$, 1 H, $CH_2=C(4')$); 6.33 (t, $J = 2.9$, 1 H, $CH_2=C(4')$); 6.82–8.33 (m, 12 arom. H). ^{13}C -NMR ($CDCl_3$): 37.35 (C(3')); 74.47 (CH_2O); 83.79 (C(2')); 101.34, 113.20, 116.47, 117.72, 121.91, 122.02, 124.00, 125.08, 126.67, 128.54, 128.77, 128.93, 134.44, 134.59, 139.92, 156.18, 157.79, 163.25 (arom. C); 169.06 (C(5')); 176.19 (C(9)). Anal. calc. for $C_{25}H_{18}O_5$: C 75.37, H 4.55; found: C 75.30, H 4.60.

1-(2-Oxo-2-phenylethyl)quinolin-4-one (**6a**). Quinolin-4-ol (1.45 g, 10 mmol), K_2CO_3 (1.37 g, 10 mmol), and dry DMF (50 ml) were stirred at r.t. for 30 min. To this soln. was added 2-bromoacetophenone (1.99 g, 10 mmol) in dry DMF (10 ml) in one portion. The resulting mixture was stirred at r.t. for 24 h (TLC monitoring) and then poured into ice-water (100 ml). The pale-yellow solid thus obtained was collected and crystallized from CH_2Cl_2/Et_2O 1:10: **6a** (2.21 g, 84%). M.p. 219–220°. 1H -NMR (DMSO): 6.06 (s, 2 H—C(1')); 6.15 (d, $J = 7.6$, H—C(3)); 7.95 (d, $J = 7.6$, H—C(2)); 7.34–8.25 (m, 9 arom. H). ^{13}C -NMR (DMSO): 57.94 (C(1')); 109.08 (C(3)); 116.72, 123.22, 125.58, 126.43, 128.22, 128.95, 131.96, 134.22, 134.28 (arom. C); 140.69 (C(8a)); 145.24 (C(2)); 176.62 (C(4)); 193.37 (C(2')). Anal. calc. for $C_{17}H_{13}NO_2$: C 77.55, H 4.98, N 5.32; found: C 77.55, H 5.03, N 5.35.

1-(2-Oxopropyl)quinolin-4-one (**6b**). From bromoacetone as described for **6a**: 75% yield. M.p. 161–162°. 1H -NMR (DMSO): 2.29 (s, Me); 5.32 (s, 2 H—C(1')); 6.10 (d, $J = 7.6$, H—C(3)); 7.81 (d, $J = 7.6$, H—C(2)); 7.33–8.21 (m, 4 arom. H). ^{13}C -NMR (DMSO): 27.02 (C(3')); 60.46 (C(1')); 108.91 (C(3)); 116.69, 123.26, 125.55, 126.32, 131.94 (arom. C); 140.48 (C(8a)); 144.98 (C(2)); 176.55 (C(4)); 202.36 (C(2')). Anal. calc. for $C_{12}H_{11}NO_2$: C 71.62, H 5.51, N 6.96; found: C 71.47, H 5.57, N 7.03.

8-(2-Oxo-2-phenylethoxy)quinoline (**8a**). Quinolin-8-ol (**7**; 0.73 g, 5 mmol), K_2CO_3 (0.69 g, 5 mmol), and dry DMF (40 ml) were stirred at r.t. for 30 min. To this soln. was added 2-bromoacetophenone (1.0 g, 5 mmol) in dry DMF (10 ml) in one portion. The resulting mixture was stirred at r.t. for 24 h (TLC monitoring) and then poured into ice-water (100 ml). The pale-yellow solid thus obtained was collected and crystallized from CH_2Cl_2/Et_2O (1:10): **8a** (0.91 g, 69%). Pale-yellow needle crystals. M.p. 124–125°. 1H -NMR ($CDCl_3$): 5.65 (s, 2 H—C(1')); 6.95–8.97 (m, 11 arom. H). ^{13}C -NMR ($CDCl_3$): 71.85 (C(1')); 110.09, 120.81, 121.72, 126.41, 128.17, 128.84, 129.60, 133.88, 134.53, 135.93, 140.24, 149.41 (arom. C); 153.82 (C(8)); 194.43 (C(2')). Anal. calc. for $C_{17}H_{13}NO_2$: C 77.55, H 4.98, N 5.32; found: C 77.59, H 4.97, N 5.37.

8-(2-Oxopropoxy)quinoline (**8b**). From bromoacetone as described for **8a**: 67% yield. M.p. 58–59°. ¹H-NMR (CDCl₃): 2.32 (s, Me); 4.88 (s, 2 H–C(1')); 6.88–8.97 (m, 6 arom. H). ¹³C-NMR (CDCl₃): 26.46 (C(3')); 74.13 (C(1')); 109.51, 121.03, 121.90, 126.46, 129.69, 136.02, 140.12, 149.53 (arom. C); 153.69 (C(8)); 206.21 (C(2')). Anal. calc. for C₁₂H₁₁NO₂·2 H₂O: C 60.75, H 6.37, N 5.90; found: C 60.80, H 6.33, N 5.90.

8-[2-(4-Chlorophenyl)-2-oxoethoxy]quinoline (**8c**). From 2-bromo-4'-chloroacetophenone as described for **8a**: 76% yield. M.p. 111–112°. ¹H-NMR (CDCl₃): 5.56 (s, 2 H–C(1')); 6.96–8.96 (m, 10 arom. H). ¹³C-NMR (CDCl₃): 72.23 (C(1')); 110.30, 121.05, 121.77, 126.40, 129.14, 129.64, 129.83, 132.88, 135.95, 140.26, 140.35, 149.47, 153.71 (arom. C); 193.96 (C(2')). Anal. calc. for C₁₇H₁₂ClNO₂: C 68.58, H 4.06, N 4.70; found: C 68.54, H 4.08, N 4.77.

8-[2-(4-Bromophenyl)-2-oxoethoxy]quinoline (**8d**). From 2-bromo-4'-bromoacetophenone as described for **8a**: 75% yield. M.p. 109–110°. ¹H-NMR (CDCl₃): 5.55 (s, 2 H–C(1')); 6.96–8.96 (m, 10 arom. H). ¹³C-NMR (CDCl₃): 72.20 (C(1')); 110.30, 121.06, 121.77, 126.39, 129.13, 129.64, 129.89, 132.13, 133.26, 135.95, 140.23, 149.46, 153.68 (arom. C); 194.18 (C(2')). Anal. calc. for C₁₇H₁₂BrNO₂·0.5 H₂O: C 58.14, H 3.73, N 3.99; found: C 57.89, H 3.81, N 4.03.

8-[2-(4-Methoxyphenyl)-2-oxoethoxy]quinoline (**8e**). From 2-bromo-4'-methoxyacetophenone as described for **8a**: 89% yield. M.p. 121–122°. ¹H-NMR (CDCl₃): 3.86 (s, MeO); 5.56 (s, 2 H–C(1')); 6.92–8.97 (m, 10 arom. H). ¹³C-NMR (CDCl₃): 55.43 (Me); 71.86 (C(1')); 109.99, 113.94, 120.60, 121.61, 126.39, 127.60, 129.53, 130.60, 135.81, 140.25, 149.33, 153.92, 164.00 (arom. C); 193.11 (C(2')). Anal. calc. for C₁₈H₁₅NO₃: C 73.70, H 5.16, N 4.78; found: C 73.67, H 5.17, N 4.81.

8-[2-Oxo-2-(4-phenylphenyl)ethoxy]quinoline (**8f**). From 2-bromo-4'-phenylacetophenone as described for **8a**: 73% yield. M.p. 135–136°. ¹H-NMR (CDCl₃): 5.66 (s, 2 H–C(1')); 6.99–8.98 (m, 15 arom. H). ¹³C-NMR (CDCl₃): 72.07 (C(1')); 110.16, 120.80, 121.68, 126.40, 127.24, 127.40, 128.34, 128.84, 128.94, 129.61, 133.22, 135.87, 139.67, 140.29, 146.50, 149.40, 153.88 (arom. C); 194.21 (C(2')). Anal. calc. for C₂₃H₁₇NO₂: C 81.39, H 5.05, N 4.13; found: C 81.15, H 5.11, N 4.13.

8-[(2,3,4,5-Tetrahydro-4-methylidene-5-oxo-2-phenylfuran-2-yl)methoxy]quinoline (**9a**). To a soln. of **8a** (0.79 g, 3 mmol) in dry THF (60 ml) were added activated Zn powder (0.26 g, 3.9 mmol), hydroquinone (6 mg), and ethyl 2-(bromomethyl)acrylate (0.78 g, 4 mmol). The mixture was refluxed under N₂ atmosphere for 6 h (TLC monitoring). After cooling, it was poured into an ice-cold 5% HCl soln. (300 ml), neutralized with 1.0N NaHCO₃, and extracted with CH₂Cl₂ (3 × 60 ml). The CH₂Cl₂ extracts were combined and washed with H₂O, dried (Na₂SO₄), and then evaporated to give a residual solid which was crystallized from a mixed solvent of CH₂Cl₂/Et₂O 1:10: **9a** (0.75 g, 75%). M.p. 101–102°. UV: 250 (4.69; 0.1N HCl/MeOH); 237 (4.62; MeOH); 237 (4.67; 0.1N NaOH/MeOH). ¹H-NMR (CDCl₃): 3.25 (dt, *J* = 16.8, 2.8, 1 H–C(3')); 4.09 (dt, *J* = 16.9, 2.6, 1 H–C(3')); 4.54 (AB type, CH₂O); 5.68 (t, *J* = 2.6, 1 H, CH₂=C(4')); 6.25 (t, *J* = 2.9, 1 H, CH₂=C(4')); 7.16–8.90 (m, 11 arom. H). ¹³C-NMR (CDCl₃): 37.31 (C(3')); 76.68 (CH₂O); 85.08 (C(2')); 113.49, 121.45, 121.64, 121.88, 125.20, 126.55, 128.30, 128.64, 129.62, 134.89, 135.82, 140.70, 141.06, 149.31, 154.73 (arom. C); 169.48 (C(5')). Anal. calc. for C₂₁H₁₇NO₃: C 76.11, H 5.17, N 4.23; found: C 76.10, H 5.19, N 4.27.

The same procedure was used to convert each of the compounds **8b–g** to **9b–g**, respectively. Compounds **9b** was obtained as a residual oil which was purified by column chromatography on silica gel using CH₂Cl₂.

8-[(2,3,4,5-Tetrahydro-2-methyl-4-methylidene-5-oxofuran-2-yl)methoxy]quinoline (**9b**). Yield: 82%. UV: 250 (4.69; 0.1N HCl/MeOH); 237 (4.60; MeOH); 238 (4.64; 0.1N NaOH/MeOH). ¹H-NMR (CDCl₃): 1.64 (s, Me); 2.84 (dt, *J* = 17.2, 2.8, 1 H–C(3')); 3.48 (dt, *J* = 17.2, 2.6, 1 H–C(3')); 4.29 (AB type, CH₂O); 5.66 (t, *J* = 2.5, 1 H, CH₂=C(4')); 6.26 (t, *J* = 2.9, 1 H, CH₂=C(4')); 7.13–8.91 (m, 6 arom. H). ¹³C-NMR (CDCl₃): 24.33 (Me); 36.81 (C(3')); 74.86 (CH₂O); 82.18 (C(2')); 112.11, 121.28, 121.53, 122.36, 126.56, 129.62, 135.31, 135.89, 140.77, 149.36, 154.62 (arom. C); 169.69 (C(5')). Anal. calc. for C₁₆H₁₅NO₃·0.125 H₂O: C 70.77, H 5.66, N 5.16; found: C 70.80, H 5.75, N 5.08.

8-[[2-(4-Chlorophenyl)-2,3,4,5-tetrahydro-4-methylidene-5-oxofuran-2-yl]methoxy]quinoline (**9c**). Yield: 63%. M.p. 107–108°. UV: 249 (4.75; 0.1N HCl/MeOH); 236 (4.69; MeOH); 238 (4.28; 0.1N NaOH/MeOH). ¹H-NMR (CDCl₃): 3.20 (dt, *J* = 16.9, 2.6, 1 H–C(3')); 4.06 (dt, *J* = 16.9, 2.6, 1 H–C(3')); 4.42 (AB type, CH₂O); 5.70 (t, *J* = 2.5, 1 H, CH₂=C(4')); 6.28 (t, *J* = 2.9, 1 H, CH₂=C(4')); 7.13–8.91 (m, 10 arom. H). ¹³C-NMR (CDCl₃): 37.43 (C(3')); 76.39 (CH₂O); 84.46 (C(2')); 113.40, 121.56, 121.80, 122.47, 126.56, 126.81, 128.82, 129.66, 134.35, 134.45, 135.89, 139.30, 141.02, 149.40, 154.57 (arom. C); 169.24 (C(5')). Anal. calc. for C₂₁H₁₆ClNO₃: C 68.95, H 4.41, N 3.83; found: C 68.91, H 4.44, N 3.87.

8-[[2-(4-Bromophenyl)-2,3,4,5-tetrahydro-4-methylidene-5-oxofuran-2-yl]methoxy]quinoline (**9d**). Yield: 58%. M.p. 123–124°. UV: 250 (4.68; 0.1N HCl/MeOH); 236 (4.66; MeOH); 238 (4.55; 0.1N NaOH/MeOH). ¹H-NMR (CDCl₃): 3.20 (dt, *J* = 17.0, 2.8, 1 H–C(3')); 4.06 (dt, *J* = 17.0, 2.5, 1 H–C(3')); 4.49 (AB type, CH₂O); 5.70 (t, *J* = 2.5, 1 H, CH₂=C(4')); 6.28 (t, *J* = 2.9, 1 H, CH₂=C(4')); 7.13–8.91 (m, 10 arom. H). ¹³C-NMR

(CDCl₃): 37.36 (C(3')); 76.26 (CH₂O); 84.44 (C(2')); 113.37, 121.53, 121.78, 122.48, 126.53, 127.09, 129.63, 131.75, 134.37, 135.86, 139.81, 140.98, 149.37, 154.53 (arom. C); 169.19 (C(5')). Anal. calc. for C₂₁H₁₆BrNO₃: C 61.48, H 3.93, N 3.41; found: C 61.32, H 3.96, N 3.43.

8-{[2,3,4,5-Tetrahydro-2-(4-methoxyphenyl)-4-methylidene-5-oxofuran-2-yl]methoxy}quinoline (**9e**). Yield: 56%. M.p. 125–126°. UV: 250 (4.29; 0.1N HCl/MeOH); 236 (4.37; MeOH); 238 (4.18; 0.1N NaOH/MeOH). ¹H-NMR (CDCl₃): 3.22 (*dt*, *J* = 16.9, 2.8, 1 H–C(3')); 3.81 (*s*, MeO); 4.05 (*dt*, *J* = 16.8, 2.6, 1 H–C(3')); 4.50 (*AB* type, CH₂O); 5.67 (*t*, *J* = 2.5, 1 H, CH₂=C(4')); 6.24 (*t*, *J* = 2.8, 1 H, CH₂=C(4')); 6.90–8.90 (*m*, 10 arom. H). ¹³C-NMR (CDCl₃): 37.34 (C(3')); 55.33 (MeO); 76.67 (CH₂O); 85.02 (C(2')); 113.35, 114.01, 121.48, 121.59, 121.89, 126.58, 129.64, 132.73, 135.06, 135.84, 141.08, 149.35, 154.77, 159.52 (arom. C); 169.60 (C(5')). Anal. calc. for C₂₂H₁₉NO₄: C 73.11, H 5.30, N 3.88; found: C 73.08, H 5.34, N 3.92.

8-{[2,3,4,5-Tetrahydro-4-methylidene-5-oxo-2-(4-phenylphenyl)furan-2-yl]methoxy}quinoline (**9f**). Yield: 65%. M.p. 154–155°. UV: 251 (4.88; 0.1N HCl/MeOH); 238 (4.86; MeOH); 240 (4.71; 0.1N NaOH/MeOH). ¹H-NMR (CDCl₃): 3.29 (*dt*, *J* = 16.9, 2.8, 1 H–C(3')); 4.11 (*dt*, *J* = 16.9, 2.6, 1 H–C(3')); 4.57 (*AB* type, CH₂O); 5.70 (*t*, *J* = 2.5, 1 H, CH₂=C(4')); 6.28 (*t*, *J* = 2.8, 1 H, CH₂=C(4')); 7.19–8.92 (*m*, 15 arom. H). ¹³C-NMR (CDCl₃): 37.35 (C(3')); 76.57 (CH₂O); 85.02 (C(2')); 113.43, 121.49, 121.66, 122.09, 125.73, 126.57, 127.08, 127.35, 127.57, 128.82, 129.63, 134.82, 135.85, 139.63, 140.29, 141.05, 141.27, 149.35, 154.71 (arom. C); 169.50 (C(5')). Anal. calc. for C₂₇H₂₁NO₃: C 79.59, H 5.20, N 3.44; found: C 79.53, H 5.25, N 3.50.

Pharmacological Evaluation. Reagents: Collagen (type I, bovine Achilles tendon) obtained from *Sigma Chem. Co.* was homogenized in 25 mM AcOH and stored (1 mg/ml) at –70°. Platelet-activating factor (PAF) was purchased from *Calbiochem-Behring Co.* and dissolved in CHCl₃. Arachidonic acid (AA), EDTA, and bovine serum albumin were purchased from *Sigma Chem. Co.*

Platelet Aggregation. Blood was collected from the rabbit marginal ear vein, anticoagulated with EDTA (6 mM) and centrifuged for 10 min at 90 × *g* and r.t. Platelet suspension was prepared from this EDTA-anticoagulated platelet-rich plasma according to the washing procedures described in [12]. Platelet numbers were counted with a *Coulter* counter (Model *ZM*) and adjusted to 4.5 × 10⁸ platelets/ml. The platelet pellets were finally suspended in *Tyrodé's* soln. of the following composition (mM): NaCl (136.8), KCl (2.8), NaHCO₃ (11.9), MgCl₂ (2.1), NaH₂PO₄ (0.33), CaCl₂ (1.0), and glucose (11.2), containing bovine serum albumin (0.35%). The platelet suspension was stirred at 1200 rpm and the aggregation was measured at 37° by the turbidimetric method as described by *O'Brien* [13] using a *Chrono-Log Lumi-aggregometer*. To eliminate the effect of the solvent on the aggregation, the final concentration of DMSO was fixed at 0.5%. Percentage of aggregation was calculated using the absorbance of platelet suspension as 0% aggregation and the absorbance of *Tyrodé's* soln. as 100% aggregation. The inhibitory concentration for 50% aggregation (*IC*₅₀) was calculated from computerization of *CA-Cricket Graph III* for five or six dose-effect levels.

REFERENCES

- [1] K. H. Lee, I. H. Hall, E. C. Mar, C. O. Starnes, S. A. Elgebaly, T. G. Waddell, R. I. Hadgraft, C. G. Ruffner, I. Weidner, *Science* **1977**, 196, 533.
- [2] N. D. Heindel, J. A. Minatelli, *J. Pharm. Sci.* **1981**, 70, 84.
- [3] H. M. R. Hoffmann, J. Rabe, *Angew. Chem. Int. Ed.* **1985**, 24, 94.
- [4] K. H. Lee, G. K. Rice, I. H. Hall, V. Amarnath, *J. Med. Chem.* **1987**, 30, 586.
- [5] B. R. Huang, K. H. Lee, T. L. Tseng, H. M. Wang, C. F. Chen, C. C. Tzeng, *Kaohsiung J. Med. Sci.* **1993**, 22, 2206.
- [6] Y. L. Chen, T. C. Wang, K. H. Lee, Y. L. Chang, C. M. Teng, C. C. Tzeng, *Helv. Chim. Acta* **1996**, 79, 651.
- [7] Y. L. Chen, T. C. Wang, S. C. Liang, C. M. Teng, C. C. Tzeng, *Chem. Pharm. Bull.* **1996**, in press.
- [8] T. C. Teng, C. N. Lin, F. N. Ko, K. L. Cheng, *Biochem. Pharmacol.* **1989**, 38, 3791.
- [9] C. N. Lin, S. S. Liou, F. N. Ko, C. M. Teng, *J. Pharm. Sci.* **1993**, 82, 11.
- [10] W. Y. Chen, F. N. Ko, C. N. Lin, C. M. Teng, *Thromb. Res.* **1994**, 75, 81.
- [11] C. N. Lin, S. C. Fang, H. C. Lin, F. N. Ko, B. J. Shieh, H. W. Liu, C. M. Teng, *J. Pharm. Pharmacol.* **1994**, 46, 917.
- [12] S. C. Kuo, C. M. Teng, J. G. Lee, F. N. Ko, S. C. Chen, T. S. Wu, *Planta Med.* **1990**, 56, 164.
- [13] J. R. O'Brien, *J. Clin. Pathol.* **1962**, 15, 452.