ELSEVIER

Eur. J. Med. Chem. 37 (2002) 77–82

EUROPEAN JOURNAL OF MEDICINAL CHEMISTRY

Short communication

6-Arylamino-5,8-quinazolinediones as potent inhibitors of endothelium-dependent vasorelaxation

Chung-Kyu Ryu [a,*], Keun-Hwa Shin [a], Ji-Hui Seo [a,b], Hwa-Jung Kim [a,b]

[a] *College of Pharmacy, Ewha Womans University, Sodaemun-Ku, Seoul 120-750, South Korea*
[b] *Center for Cell Signaling Research, Ewha Womans University, Seoul 120-750, South Korea*

Received 18 July 2001; received in revised form 29 October 2001; accepted 31 October 2001

Abstract

6-(Substituted-phenyl)amino-5,8-quinazolinediones (**3**) were synthesised by regioselective substitution of 5,8-quinazolinedione (**5**) with appropriate arylamines in the presence of Ce(III) ions. All synthesised 5,8-quinazolinediones **3** showed a potent and efficacious inhibitory effect on the acetylcholine (ACh)-induced vasorelaxation of rat aorta with the endothelium. The quinones **3**, at a low concentration of 0.1 μM, reduced the maximal response with increase of EC_{50} values for ACh. The results indicate that quinones **3** are potent inhibitors of endothelium-dependent vasorelaxation.

Keywords: 6-arylamino-5,8-quinazolinedione; isosteres; inhibitors; endothelium-dependent vasorelaxation

1. Introduction

6-Phenylamino-5,8-quinolinedione (LY83583, **1**), a heterocyclic quinone, is a potent inhibitor of endothelium-dependent vasorelaxation [1–4] and lowers intracellular cGMP in several tissues [1] due to inhibitions of endothelial nitric oxide synthase (eNOS) activity [5]. LY83583 (**1**) interacts with the reductase domain of NOS, leading to a decrease in NO formation [4]. Additional quinones such as 1,4-naphthoquinones have also been shown to inhibit the endothelium-dependent vasorelaxation [6,7] (Fig. 1). On the line, in our previous report [8,9], 6-(substituted-phenyl)amino-5,8-quinolinediones **2**, inhibited the acetylcholine (ACh)-induced vasorelaxation of phenylephrine (PE)-precontracted rat aorta with the intact endothelium and the presence of nitrogen on the heterocyclic quinones **2** might be an important factor in the inhibitory activities. Based on the heterocyclic quinones, with several pharmacological activities, such as inhibitory effect on the vasorelaxation, 6-arylamino-5,8-quinazolinediones **3**, as bioisosteres of quinones **1** and **2**, were newly synthesised and evaluated on the inhibitory potential of endothelium-dependent vasorelaxation (Fig. 1). A variety of heterocyclic quinones with different substituents can inhibit the vasorelaxation through different action [8]. The 6-arylamino-5,8-quinazolinediones **3** were incorporated with fluorine, chlorine or bromine, respectively, to alter their pharmacological properties.

There have been a few report [10–13] on synthesis and cytotoxicities of some 5,8-quinazolinedione derivatives against cancer cell lines. However, the inhibitory properties of 5,8-quinazolinediones **3** on vasorelaxation have not been reported. We report, therefore, the synthesis and inhibitory activities of quinazolinediones **3a–j** on the endothelium-dependent vasorelaxation (Fig. 2).

2. Chemistry

The method used to synthesise the quinones **3** is shown in Fig. 2. 5,8-Quinazolinedione (**5**) that has been previously described [13,14] was synthesised by oxidising

* Correspondence and reprints.
E-mail address: ckryu@mm.ewha.ac.kr (C.-K. Ryu).

0223-5234/02/$ - see front matter
PII: S0223-5234(01)01290-9

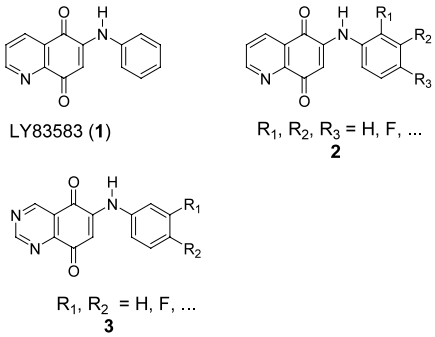


Fig. 1. Quinones as inhibitors of endothelium-dependent vasorelaxation.

the 5,8-quinazolinediol (**4**) with Fremy's salt (potassium nitrosodisulfonate) resulting in a 69% yield. 6-Arylamino-5,8-quinazolinediones **3a**–**j** (Table 1) were synthesised by regioselective nucleophilic substitution of the quinone **5** with appropriate arylamines in the presence of Ce(III). These substitutions were similar to the Michael-type regioselective substitution of arylamines on 6,7-dichloro-5,8-quinolinedione in the presence of Ce(III) [15], and had overall high yields of 50–95%.

3. Pharmacology

The quinazolinediones **3** were tested for their inhibitory activities on the ACh-induced vasorelaxation of PE-precontracted rat aortas with the intact endothelium according to the previously reported method [9]. LY83583 (**1**) and N^G-nitro-L-arginine (L-NA) [16], well-known inhibitors of the endothelium-dependent vasorelaxation, were used as standard agents. Maximal effect (E_{max}) values for E_{max} and EC_{50} values of Ach were obtained from the mean concentration–response curves to ACh (0.01–10 μM) for aortic rings treated with vehicle and quinones **3** (0.1 μM) (Table 1). The EC_{50} value denotes the ACh concentration producing 50% vasorelaxation in the presence of quinones, and the E_{max} value represents percent of the maximal vasorelaxation.

The percent inhibition of PE responses by ACh in control tissues used in our study was quite consistent within tissue preparations (Table 1). The tissue preparations that did not produce appropriate control ACh responses were omitted, and control ACh responses in the absence of test agent were always tested prior to test ACh responses in the presence of test agent using an identical tissue preparation.

Most of the 6-arylamino-5,8-quinazolinediones **3a**–**j** at the test concentration of 0.1 μM strongly inhibited the ACh-induced vasorelaxation and increased the EC_{50} value for ACh, whereas, L-NA inhibited the vasorelaxation at the higher concentration of 1 μM (Table 1). Among them, the quinazolinediones **3a**, **3c**, **3e**, and **3h** significantly reduced the E_{max} of ACh compared with LY83583, and also these compounds caused an increase in EC_{50} values by 11.8–12.7 times greater than the control values (Fig. 3). LY83583 (**1**), however, increased the EC_{50} value by only 3.4-fold relative to the control value. Even, these three quinones **3a**, **3e** and **3h** as low as 0.1 μM, increasing the ACh concentration to 10 μM did not restore the maximal relaxant response.

When potent inhibitors (e.g. **3a**, **3e**, **3h**) and weak inhibitor **3g** are compared for their augmenting effect of PE-elicited contraction in aortic rings with endothelium, these potent inhibitors produced significant effects, whereas, the weak inhibitor did not (Table 2).

4. Results and discussion

Most of the quinones **3** significantly reduced the EC_{50} and E_{max} of ACh-induced vasorelaxation and showed more potent inhibition in the vasorelaxation in comparison with LY83583 (**1**) and L-NA. In terms of structural viewpoint, however, the 5,8-quinazolinedione (**5**) without the arylamino group at the test concentration of 0.1 μM exhibited poor inhibitory activities on the ACh-induced vasorelaxation.

The quinones **3** with the inhibitory effect on endothelium-dependent vasorelaxation produced moderate increases in basal tone of the aorta, although, statistically not significant, and augmented contractions elicited by PE, but only in aortic rings with endothelium. However, in aortic rings lacking endothelium, there was no change in basal tonus and in the PE-induced contractions, even at higher concentrations of these

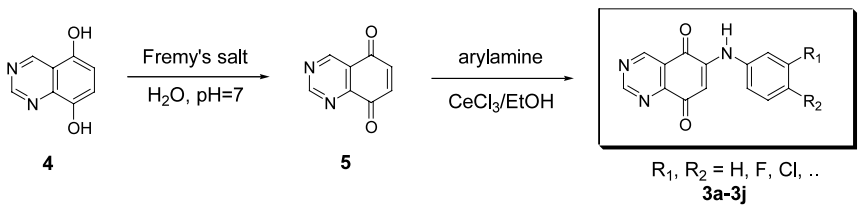
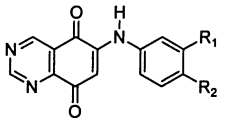


Fig. 2. Synthesis of 5,8-quinazolinediones.

Table 1
The inhibitory effects of the quinones **3** on ACh-induced vasorelaxation of rat aorta



Compounds	Substituent		EC_{50} [a](μM)	(*N*) [b]	E_{max} [a] (% of control)
	R_1	R_2			
Control	–	–	0.100 ± 0.018	(11)	100 ± 1
L-NA	–	–	1.100 ± 0.227*	(4)	69 ± 3*
1	–	–	0.345 ± 0.032*	(7)	77 ± 5*
3a	H	H	1.181 ± 0.290**	(4)	28 ± 6**
3b	H	F	0.846 ± 0.194*	(4)	69 ± 4*
3c	H	Cl	1.036 ± 0.414*	(4)	31 ± 10**
3d	H	Br	0.534 ± 0.118*	(3)	80 ± 5*
3e	H	I	1.118 ± 0.607**	(4)	40 ± 12**
3f	F	H	0.437 ± 0.091*	(4)	69 ± 9*
3g	F	F	0.184 ± 0.105	(4)	93 ± 2
3h	H	CF_3	1.269 ± 0.438**	(4)	41 ± 2**
3I	H	OCF_3	0.504 ± 0.142*	(3)	68 ± 2*
3j	I	H	0.664 ± 0.151*	(4)	69 ± 3*
5	–	–	0.220 ± 0.017	(4)	96 ± 5

*$P<0.05$ relative to the control group; **$P<0.05$ relative to LY83583 (**1**) by student's *t*-test.
[a] EC_{50} is the ACh concentration producing 50% of the vasorelaxation of PE-precontraction (0.3 μM) after preincubation of each quinone (0.1 μM) or L-NA (1 μM) for 20 min and E_{max} is the percent of the maximal ACh-induced vasorelaxation (control).
[b] Data are mean ± S.E.M. using *n* numbers of aortic rings from separate animals.

compounds. In previous report [9], sodium nitroprusside (1 nM–0.3 μM) elicited concentration-dependent relaxations in PE-precontracted aortic rings devoid of endothelium. Under control conditions, sodium nitroprusside as a NO-donor produced almost complete relaxation (98.7% $n = 8$) in the aortic ring with a mean EC_{50} value of 17 nM ($n = 8$). In the presence of a concentration (1 μM) of compounds **3**, the compounds were equally potent with LY83583 in inhibiting sodium nitroprusside-induced relaxations. These observations strongly suggest that the compounds **3** interfere with the relaxant action of endogenously released NO, as indicated for the similar actions of OQ1 and OQ21 in our previous report [9] and LY83583 [2,3] on the PE-induced contraction.

In terms of structure–activity relationship, observations presented in Table 1 and previously reported data [8], the quinazolinedione skeletons **3** showed, in general, more potent inhibitory activities than the bioisosteres such as 5,8-quinolinedione **2**, 5,8-isoquinolinedione and 1,4-naphthoquinone skeletons. In addition, the quinazolinedione (**5**) without an arylamino group exhibited weak inhibitory activities. Thus, the arylamino moiety is not essential for the inhibitory activities of these quinones, but it improves the activities significantly. The structure–activity relationship may not exist between positions of substituents (R_1, R_2 and R_3) for the 6-arylamino moieties of quinones **3**.

5. Conclusions

The 6-arylamino-5,8-quinazolinediones **3** were synthesised by regioselective substitution of 5,8-quinazolinedione (**5**) in the presence of Ce(III) ions with arylamines. All of the synthesised quinones **3** showed a

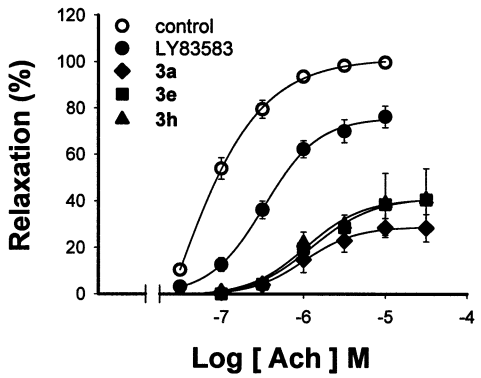


Fig. 3. Concentration-dependent inhibition of ACh-induced endothelium-dependent relaxation by quinazolinedione compounds. Rat aortic rings were isolated with intact endothelium and preincubated with indicated quinazolinedione compounds or vehicle at concentrations of 0.1 μM for 20 min prior to contraction with 0.3 μM PE. Cumulatively increasing concentrations of ACh were added to organ baths. Relaxation responses are expressed as percentage of decrease in submaximal contraction elicited by PE. All concentrations represent final bath concentrations. Values represent mean values ± S.E.M. from 4 to 11 aortic rings from different animals.

Table 2
Effects of the quinones 3 on phenylephrine (0.3 μM)-induced contractions of aortic rings with intact endothelium.

Compounds	Concentration (μM)	Tension (g) [a]
Control	–	1.36 ± 0.04 (11) [b]
3a	0.1	1.84 ± 0.19 (9)*
3e	0.1	1.88 ± 0.29 (4)*
3h	0.1	1.83 ± 0.16 (4)*
3k	0.1	1.59 ± 0.06 (6)*
3g	0.1	1.41 ± 0.08 (4)

*Significantly different ($P<0.05$) from corresponding control by student's t-test.
[a] Responses were expressed as 'g'.
[b] Data are the mean ± S.E.M. of n observations in parentheses.

potent and efficacious inhibitory effect on the ACh-induced vasorelaxation of PE-precontracted rat aorta with intact endothelium. The quinones, at a low concentration of 0.1 μM, significantly reduced the maximal response with an increase of EC_{50} values for ACh. The results indicate that 6-arylamino-5,8-quinazolinediones are potent inhibitors of endothelium-dependent vasorelaxation. Further pharmacological investigations of these quinones as inhibitors of endothelial and neuronal NO syntheses are in progress.

6. Experimental protocols

6.1. Chemistry

All melting points (m.p.) were measured in open capillary tubes with Buchi melting point B-545 and were uncorrected. The TLC was performed on precoated silica gel (60G 254, Merck) using chloroform for the solvent. The compounds were detected under UV light (254 nm) or by heating to 110 °C after spraying with a 30% H_2SO_4–vanillin solution. Column chromatography was performed on silica gel G60 (70–230 mesh, ASTM, Merck). The purity of 5,8-quinazolinediones (**3a**–**j**) was also verified by GC (Hewlett Packard 5890A, HP-5 capillary column at 260 °C, N_2 gas, 17 mL min^{-1} as carrier gas, FID). The IR spectra were taken from Perkin–Elmer 1420r IR spectrometer with KBr pellets. ^{1}H-NMR spectra were recorded on Brucker DPX 250 MHz spectrometer using DMSO-d_6 as solvents, and chemical shifts were given in ppm with TMS as a standard. Mass spectra were obtained on JMS AX 505 WA spectrometer (electronic impact at 70 eV). Elemental analyses were performed by CE instruments EA1110 with sulfanilamide as standard material, and analytical results for C, H and N were within ± 0.4% of theoretical values. Arylamines, DMSO-d_6 and other reagents were obtained from Aldrich Chemical Co. L-NA and reagents for biological screening were obtained from Sigma Co.

6.1.1. The synthesis of 5,8-quinazolinedione (5)

Briefly, 5,8-quinazolinediol (**4**) was prepared according to known procedure [13]. To a solution of compound **4** (1.62 g, 10 mmol) in 400 mL of acetone, a solution of potassium nitrosodisulfonate (5 g, 18.65 mmol) in NaH_2PO_4 buffer (0.3 M, 800 mL, pH 7) was added. The mixture was stirred at room temperature (r.t.) for 1 h and was extracted twice with CH_2Cl_2. The extract was evaporated and then was purified by silica gel column chromatography with $CHCl_3$. 5,8-Quinazolinedione (**5**) was crystallised from CH_2Cl_2 (1.10 g, 69%); m.p. 355–356 °C (dec.); ^{1}H-NMR (DMSO-d_6): δ 9.7 (s, 1H), 9.7 (s, 1H), 7.1 (d, 1H, $J = 7.1$ Hz), 6.9 (d, 1H, $J = 7.1$ Hz) ppm; MS (m/z): 160 ($[M]^+$); $C_8H_4N_2O_2$ (160.13).

6.1.2. General procedure for the synthesis of 6-arylamino-5,8-quinazolinediones (3a–j)

A solution of 5,8-quinazolinedione (**5**, 0.160 g, 1 mmol) and $CeCl_3 \cdot 7H_2O$ (0.373 g, 1 mmol) in 15 mL of 95% EtOH was added to the solution of the arylamine (1.1 mmol) in 5 mL of 95% EtOH and stirred at r.t. for 2 h and then refluxed for 4–5 h. After the mixture was kept overnight in the refrigerator or poured into 20 mL of ice water, the precipitate was collected by filtration. The filtered crude product was purified by silica gel column chromatography with $CHCl_3$ or crystallised from 95% EtOH. Purity of reaction products was verified by both TLC and GC, and the results confirmed that a single compound was contained.

*6.1.2.1. 6-[(Phenyl)amino]-5,8-quinazolinedione (**3a**).* The yield was 60% as dark brown powder (aqueous (aq.) EtOH); m.p.: 209–211 °C; IR (KBr): ν 3400 (s, NH), 3050 (m), 2800 (m), 1640 (s, C=O), 1440–1520, 1380 (s) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 9.6 (s, 1H, NH), 9.3 (s, 2H, H2, H4), 7.3–7.2 (m, 4H, Ph–H), 6.2 (s, 1H, H7) ppm; MS (m/z): 251 ($[M]^+$), 222, 195, 144, 76; $C_{14}H_9N_3O_2$ (251.24).

*6.1.2.2. 6-[N-(4-Fluorophenyl)amino]-5,8-quinazolinedione (**3b**).* The yield was 55% as dark brown powder ((aq.) EtOH); m.p.: 260–262 °C; IR (KBr): ν 3400 (s, NH), 3010 (m, aromatic ring), 1640 (s, C=O), 1180 (m) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 10.0 (s, 1H, NH), 9.3 (s, 2H, H2, H4), 7.6–6.8 (m, 4H, Ph–H), 6.1 (s, 1H, H7) ppm; MS (m/z): 269 ($[M]^+$), 240, 213, 162, 95, 76; $C_{14}H_7FN_3O_2$ (269.23).

*6.1.2.3. 6-[N-(4-Chlorophenyl)amino]-5,8-quinazolinedione (**3c**).* The yield was 72% as black powder ((aq.) EtOH); m.p.: 242–243 °C; IR (KBr): ν 3400 (m, NH), 3100 (m, aromatic), 1690 (m, C=O), 1400–1500, 1380 (s), 1270 (m), 1180 (s) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 9.6 (s, 1H, H), 9.4 (s, 2H, H2, H4), 7.5–7.4 (m, 4H, Ph–H), 6.2 (s, 1H, H7) ppm; MS (m/z): 285 ($[M]^+$), 250, 229, 178, 111, 76; $C_{14}H_8ClN_3O_2$ (285.69).

*6.1.2.4. 6-[N-(4-Bromophenyl)amino]-5,8-quinazolinedione (**3d**).* The yield was 75% as dark brown powder ((aq.) EtOH); m.p.: 209–210 °C; IR (KBr): ν 3470 (s, NH), 3050 (m), 2900 (m), 1720 (s, C=O), 1380 (s), 1270 (m), 690 (m) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 10.9 (s, 1H, NH), 9.6 (s, 1H, H4), 9.4 (s, 1H, H2), 7.6–7.2 (m, 4H, Ph–H), 6.3 (s, 1H, H7) ppm; MS (m/z): 329 ($[M]^+$), 300, 273, 222, 155, 76; $C_{14}H_8BrN_3O_2$ (330.14).

*6.1.2.5. 6-[N-(4-Iodophenyl)amino]-5,8-quinazolinedione (**3e**).* The yield was 81% as dark yellow brown powder ((aq.) EtOH); m.p.: 228–230 °C; IR (KBr): ν 3290 (s, NH), 3050 (m), 2905 (s), 1650 (s, C=O), 1010 (m) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 9.6 (s, 1H, NH), 9.4 (s, 2H, H2, 4), 7.8 (m, 2H, Ph–H), 7.2 (m, 2H, Ph–H), 6.3 (s, 1H, H7) ppm; MS (m/z): 377 ($[M]^+$), 348, 321, 270, 250, 203, 76; $C_{14}H_8IN_3O_2$ (377.14).

*6.1.2.6. 6-[N-(3-Fluorophenyl)amino]-5,8-quinazolinedione (**3f**).* The yield was 67% as brown powder ((aq.) EtOH); m.p.: 245–256 °C; IR (KBr): ν 3339 (s, NH), 3065 (s, aromatic ring), 2900 (m), 1660 (s, C=O), 1450–1550, 1432 (m) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 9.6 (s, 1H, NH), 9.4 (s, 2H, H2, 4), 7.0–7.6 (m, 4H, benzene), 6.4 (s, 1H, H6) ppm; MS (m/z): 269 ($[M]^+$), 260, 240, 213, 162, 262, 95, 76; $C_{14}H_7FN_3O_2$ (269.23).

*6.1.2.7. 6-[N-(3,4-Difluorophenyl)amino]-5,8-quinazolinedione (**3g**).* The yield was 50% as violet powder ((aq.) EtOH); m.p.: 386–388 °C; IR (KBr): ν 3329 (s, NH), 3020 (m), 2900 (m), 1660 (s, C=O), 1430–1550, 1345 (s) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 9.6 (s, 1H, NH), 9.4 (s, 2H, H2, 4), 7.2–7.6 (m, 4H, benzene), 6.3 (s, 1H, H6) ppm; MS (m/z): 287 ($[M]^+$), 231, 213, 180, 152, 113, 76; $C_{14}H_7F_2N_3O_2$ (287.22).

*6.1.2.8. 6-[N-(4-Trifluoromethylphenyl)amino]-5,8-quinazolinedione (**3h**).* The yield was 85% as dark orange powder ((aq.) EtOH); m.p.: 202–204 °C; IR (KBr): ν 3400 (s, NH), 3040 (m), 2920 (m), 1640 (s, C=O), 1380 (s), 1010 (m), 740 (s) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 10.0 (s, 1H, NH), 9.6 (s, 2H, H2, H4), 7.3–6.8 (m, 4H, Ph–H), 6.2 (s, 1H, H7) ppm; MS (m/z): 319 ($[M]^+$), 300, 263, 250, 212, 145, 76; $C_{15}H_8F_3N_3O_2$ (319.24).

*6.1.2.9. 6-[N-(4-Trifluoromethoxyphenyl)amino]-5,8-quinazolinedione (**3i**).* The yield was 87% as dark orange powder ((aq.) EtOH); m.p.: 202–203 °C; IR (KBr): ν 3400 (s, NH), 3060 (m), 2910 (m, 1670 (s, C=O), 1380 (s), 1010 (m), 740 (s) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 10.0 (s, 1H, NH), 9.6 (s, 2H, H2, 4), 7.3–6.8 (m, 4H, benzene), 6.2 (s, 1H, H7) ppm; MS (m/z): 335 ($[M]^+$), 300, 263, 250, 212, 145, 76; $C_{15}H_8F_3N_3O_3$ (335.24).

*6.1.2.10. 6-[N-(3-Iodophenyl)amino]-5,8-quinazolinedione (**3j**).* The yield was 61% as dark purple powder ((aq.) EtOH); m.p.: 166–168 °C; IR (KBr): ν 3400 (s, NH), 2900 (m, aromatic ring), 2300 (s), 1640 (s, C=O), 1450–1550 (v, benzene), 1250 (m) cm^{-1}; ^{1}H-NMR (DMSO-d_6): δ 9.6 (s, 1H, NH), 9.4 (s, 2H, H2, 4), 7.0–7.8 (m, 4H, benzene), 6.3 (s, 1H, H7) ppm; MS (m/z): 377 ($[M]^+$), 368, 321, 270, 250, 203, $C_{14}H_8IN_3O_2$ (377.14).

6.2. Pharmacological studies on vasorelaxant responses in organ bath experiment

The quinazolinediones **3** and **5** were tested for their inhibitory activities on the ACh-induced vasorelaxation of PE-precontracted rat aortas according to a previously reported method [9]. Briefly, male Sprague–Dawley rats (200–300 g) were sacrificed by decapitation and the thoracic aortas were removed. The aortic rings placed in oxygenated (95% O_2–5% CO_2) KR buffer (pH 7.4) with the following composition (in mM): NaCl, 118; KCl, 4.7; $CaCl_2$, 2.5; $MgSO_4$, 1.2; KH_2PO_4, 1.2; $NaHCO_3$, 25; glucose, 10; and EDTA, 0.01. Aortas were cut into rings, ca. 2–3 mm long. In some preparations, the endothelium was removed. Rings were suspended in organ baths, attached to force displacement transducers (Grass Model FT03) and connected to Grass model 79E polygraph. They were equilibrated for 1 h at 37 °C under a resting tension of 2.0 g in a KR buffer. Preparations were oxygenated by continuous bubbling with 95% O_2–5% CO_2 and incubation media was routinely changed every 30 min. Following equilibration period, 60 mM KCl was added to the bathing solution. Tissues were then washed until a steady baseline was reached and tension adjusted to 2 g. Functional integrity of the endothelium was assessed by the relaxation of the pre-contracted rings in response to ACh (3 μM), and the successful removal of endothelium was ascertained by the lack of relaxation to ACh given up to a concentration of 10 μM. Relaxant agents were added as 100 μL aliquots to 10 mL bath chambers at the peak of submaximal (70–80%) precontractions elicited by 0.3 μM PE. Drugs were added directly to the organ baths. In each experiment, a single concentration of new quinone compounds was added, and left in contact with the tissue for 20 min. Tissues were contracted with 0.3 μM PE. Responses to each concentration of the relaxants were expressed as percent reductions of PE contraction.

Acknowledgements

This study was supported by a grant of the Minister of Health and Welfare (No HMP-00 B-21500-00111).

References

[1] A. Muelsch, R. Busse, S. Lieb, U. Fostermann, J. Pharmacol. Exp. Ther. 247 (1988) 283–288.
[2] E. Malta, P.S. Macdonald, G.J. Dusting, Naunyn-Schmiedeber's Arch. Pharmacol. 337 (1988) 459–464.
[3] D. Luo, S.R. Vincent, Eur. J. Pharmacol. Mol. Pharmacol. Sect. 290 (1995) 247–251.
[4] Y. Kumagai, K. Midorikawa, Y. Nakai, T. Yoshikawa, K. Kushida, S. Homma-Takeda, N. Shimojo, Eur. J. Pharmacol. 360 (1998) 213–218.
[5] A.P. Garner, M.J.I. Paine, I. Rodriguez-Crespo, E.C. Chinje, P.O. De Montellano, I.J. Stratford, D.G. Tew, C.R. Wolf, Cancer Res. 59 (1999) 1929–1934.
[6] J.Y. Lee, M.Y. Lee, S.M. Chung, J.H. Chung, Toxicol. Appl. Pharmacol. 161 (1999) 140–145.
[7] Y. Kumagai, H. Nakajima, K. Midorikawa, S. Homma-Takeda, N. Shimojo, Chem. Res. Toxicol. 11 (1998) 608–613.
[8] C.K. Ryu, S.H. Jung, J.A. Lee, H.J. Kim, S.H. Lee, J.H. Chung, Bioorg. Med. Chem. Lett. 9 (1999) 2469–2472.
[9] J.A. Lee, S.H. Jung, M.K. Bae, C.K. Ryu, J.Y. Lee, J.H. Chung, H.J. Kim, Gen. Pharmacol. 34 (2000) 33–42.
[10] I.A. Shaikh, F. Johnson, A.P. Grollman, J. Med. Chem. 29 (1986) 1329–1340.
[11] J. Renault, S. Giorgi-Renault, M. Baron, P. Mailliet, C. Paoletti, S. Cros, E. Viosin, J. Med. Chem. 26 (1983) 1715–1719.
[12] S. Giorgi-Renault, J. Renault, P. Gebel-Servolles, M. Baron, C. Paoletti, S. Cros, M.-C. Bissery, F. Lavelle, G. Atassi, J. Med. Chem. 34 (1991) 38–46.
[13] G. Melesani, F. Marcolin, G. Rodighiero, J. Med. Chem. 13 (1970) 161–162.
[14] K.T. Potts, D. Bhattacharjee, E.B. Walsh, J. Org. Chem. 51 (1986) 2011–2017.
[15] Y.T. Pratt, J. Org. Chem. 27 (1962) 3905–3910.
[16] S. Moncada, A. Higgs, R. Furchgott, Pharmacol. Rev. 49 (1997) 137–142.