

Communications to the Editor

[Chem. Pharm. Bull.]
30(2) 744-749 (1982)

NEW HYPOTENSIVE AGENTS¹⁾

Masayuki Ishikawa,^{*,a} Hiroshi Azuma,^a Yukuo Eguchi,^a Akiko Sugimoto,^a
Shigeru Ito,^a Yoshimi Takashima,^a Hisashi Ebisawa,^b Soyao Moriguchi,^b
Izumi Kotoku^b and Hiroshi Suzuki^b

Institute for Medical and Dental Engineering, Tokyo Medical and Dental
University,^a 2-3-10 Surugadai, Kanda, Chiyoda-ku, Tokyo 101, Japan and
Life Science Research Laboratory, Showa Denko Co., Ltd.,^b 2-24-60 Tama-
gawa, Ohta-ku, Tokyo 146, Japan

The N-alkyl derivatives (1a-c) of 2-carbamoyloxymethyl-3-(2-chloro-phenyl)-6-ethoxycarbonyl-5,7-dimethyl-4(3H)-quinazolinone exert a potent hypotensive activity in anesthetized rabbits and in conscious spontaneously hypertensive rats (SHR). The hypotensive activity of these compounds is attributed to a calcium antagonistic action, inhibition of norepinephrine release, potentiating effect on adenosine response, effects on central nervous system, nicotinic blocking action and to functional beta-blocking action. The hypotensive effect is not mediated through excitation of adrenergic beta-receptors or muscarinic receptors.

KEYWORDS — hypotensive agent; 3-(2-chlorophenyl)-6-ethoxycarbonyl-5,7-dimethyl-2-(N-methylcarbamoyloxymethyl)-4(3H)-quinazolinone; EG1088; calcium antagonist; vasodilator; potentiator of adenosine; CNS action; nicotinic blocking action; functional beta-blocking action

We report a new class of hypotensive agents, the 4(3H)-quinazolinone derivative represented by structure 1. 3-(2-Chlorophenyl)-6-ethoxycarbonyl-5,7-dimethyl-2-(N-methylcarbamoyloxymethyl)-4(3H)-quinazolinone (1a) coded as EG1088, and its homologs (1b and 1c) demonstrate a high degree of hypotensive activity in anesthetized rabbits (i.v.) and in conscious SHR (p.o.).

The synthesis of the compounds was accomplished as outlined in Chart 1. Treatment of anthranilic acid (2)²⁾ with acetic anhydride, and subsequent heating of the resulting crude product with o-chloroaniline in the presence of PCl₃ in xylene at 140-150°C for 4 h by the method of Grimmel et al.³⁾ gave 2-methylquinazolinone (3) [mp 131-132°C; MS m/e 370 (M⁺); PMR (in CDCl₃) δ: 1.39 (3H, t), 2.17 (3H, s, 2-CH₃), 2.24 (3H, s, 7-CH₃), 2.78 (3H, s, 5-CH₃), 4.42 (2H, q), 7.25-7.75 (5H, m, aromatic H)] in 75% yield. Bromination of 3 with eq bromine in acetic acid gave the corresponding 2-monobromomethyl derivative, mp 108-110°C, which was treated with sodium acetate in DMF at 80°C. The resulting crude product was then treated with sodium ethoxide in ethanol with ice cooling to give 2-hydroxymethylquinazolinone (4) [mp 151-152°C; MS m/e 386 (M⁺); PMR (in CDCl₃) δ: 1.46 (3H, t), 2.50 (3H, s, 7-CH₃), 2.85 (3H, s, 5-CH₃), 4.17 (3H, s, 2-CH₂OH), 4.54 (2H, q), 7.40-7.80 (5H, m, aromatic H) in an overall yield of 55%. Treatment of 4 with methyl, ethyl and isopropyl isocyanate in

pyridine at 80°C respectively afforded desired carbamates, 1a [mp 183-184°C; MS m/e 443 (M^+); PMR (in $CDCl_3$) δ : 1.40 (3H, t), 2.42 (3H, s, 7- CH_3), 2.69 (3H, d, N- CH_3), 2.71 (3H, s, 5- CH_3), 4.43 (2H, q), 4.70 (2H, d, 2- CH_2O -), 4.6-4.8 (1H, bs), 7.35-7.75 (5H, m, aromatic H)], 1b [mp 108-110°C; MS m/e 457 (M^+); monohydrochloride: mp 168-172°C (dec.)] and 1c [mp 134.5-135.5°C; MS m/e 471 (M^+)] in 80-85% yield.

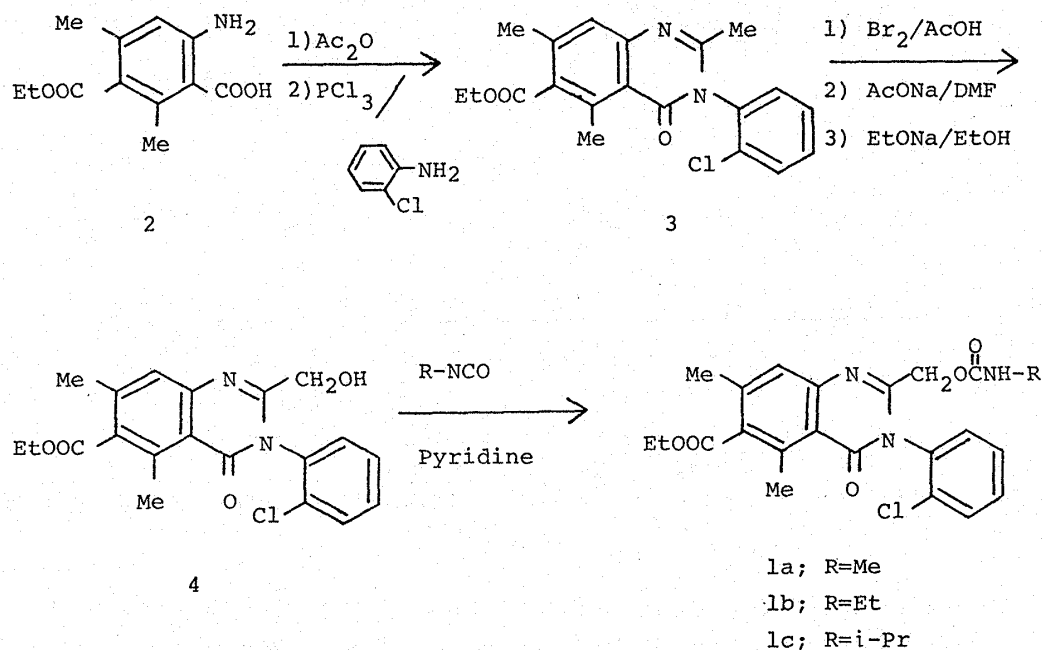


Chart 1

The compounds (1a-c) produced dose-dependent and long lasting hypotensive effects when given orally to conscious SHR. The most pronounced effect was observed 4 to 6 h after ingestion of the drug, as illustrated in Fig. 1. The pd_{30} (dose which decreases systolic blood pressure by 30 mmHg) was calculated to be 2.8, 0.35 and 0.42 mg/kg, p.o. for 1a, 1b and 1c, respectively, compared with 65 μ g/kg, p.o. for clonidine. The hypotensive effect of diltiazem was less potent (pd_{30} =30 mg/kg, p.o.) and shorter lasting than the effects of 1a-c and clonidine (Fig. 1 and Table I). The systolic blood pressure was all but unchanged when 1a was successively administered p.o. in daily doses of 3 and 10 mg/kg for 15 days. Rebound hypertension did not occur after termination of the administration (Fig. 2).

The compounds (1a-c) produced a dose-dependent hypotension in anesthetized rabbits. The pd_{30} (dose which decreases mean arterial blood pressure by 30 mmHg) and half maximum duration of 1a-c, clonidine, nifedipine and diltiazem are shown in Table I. The hypotensive effect and duration of action of 1a-c were more potent and longer lasting than the effects of calcium antagonists such as nifedipine⁴⁾ and diltiazem. In this respect, the hypotensive action of 1a-c was similar to that of clonidine.

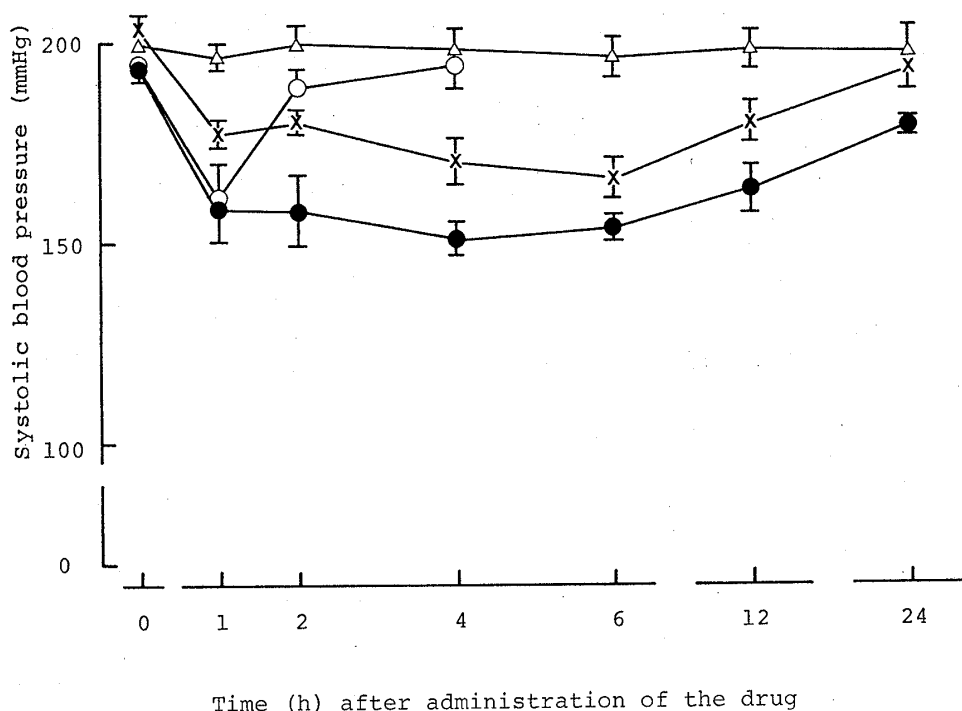


Fig. 1. Anti-hypertensive Effects of EG1088, Diltiazem and Clonidine in spontaneously Hypertensive Rats

Systolic blood pressure was measured in the tail using a plethysmograph. After determination of the predrug value, the pressure was measured 1, 2, 4, 6, 12 and 24 h after administration of drugs in groups of 4 to 6 male SHR (Wistar Kyoto origin) weighing about 300 g. The drugs tested were suspended in 0.5% aqueous solution of sodium carboxymethyl cellulose (CMC) and given p.o. ●—● : 10 mg/kg of EG1088, ○—○ : 10 mg/kg of diltiazem, x—x : 300 μg/kg of clonidine, Δ—Δ : Vehicle (0.5% CMC) Vertical bars represent S.E.

Table I. Comparison of the Hypotensive Activities of the Compounds (1a-c), Clonidine, Nifedipine and Diltiazem in conscious spontaneously Hypertensive Rats (SHR) and Anesthetized Rabbits

Agents	SHR ^{a)}		Rabbit ^{b)}	
	pD ₃₀ ^{c)} (mg/kg, p.o.)	Time to peak (h)	pD ₃₀ ^{c)} (μg/kg, i.v.)	Half maximum duration (min) (dose, i.v.)
1a	2.8	4	8.2 ± 1.4	10.5 ± 2.12 (10μg/kg)
1b	0.35	6	0.79 ± 0.10	10.4 ± 1.57 (1μg/kg)
1c	0.42	4	0.62 ± 0.16	17.0 ± 2.75 (1μg/kg)
Clonidine	0.065	6	12.5 ± 4.4	16.5 ± 2.05 (10μg/kg)
Nifedipine	1.1 ^{d)}	0.5 ^{d)}	93.8 ± 15.6	3.5 ± 0.64 (100μg/kg)
Diltiazem	30.0	0.5	1120.0 ± 190.0	2.8 ± 0.48 (1000μg/kg)

a) Systolic blood pressure in conscious SHR was measured in the tail using a plethysmograph. After determination of the predrug value, the pressure was measured 1, 2, 4, 6, 12 and 24 h (0.5 h only for diltiazem) after oral administration of drugs to groups of 4 to 6 male SHR (Wistar Kyoto origin) weighing about 300 g. b) Changes in mean arterial blood pressure in rabbits anesthetized with sodium pentobarbital were determined with pressure transducer and a cannula inserted into the carotid artery. The hypotensive activities of the drugs were determined using groups of 5 to 6 rabbits of either sex, weighing about 3 kg. c) The pd_{30} was graphically estimated from dose-response curves. d) Estimated from the data reported by H. Ishii, K. Itoh and T. Nose in *Europ. J. Pharmacol.*, 64, 21 (1980).

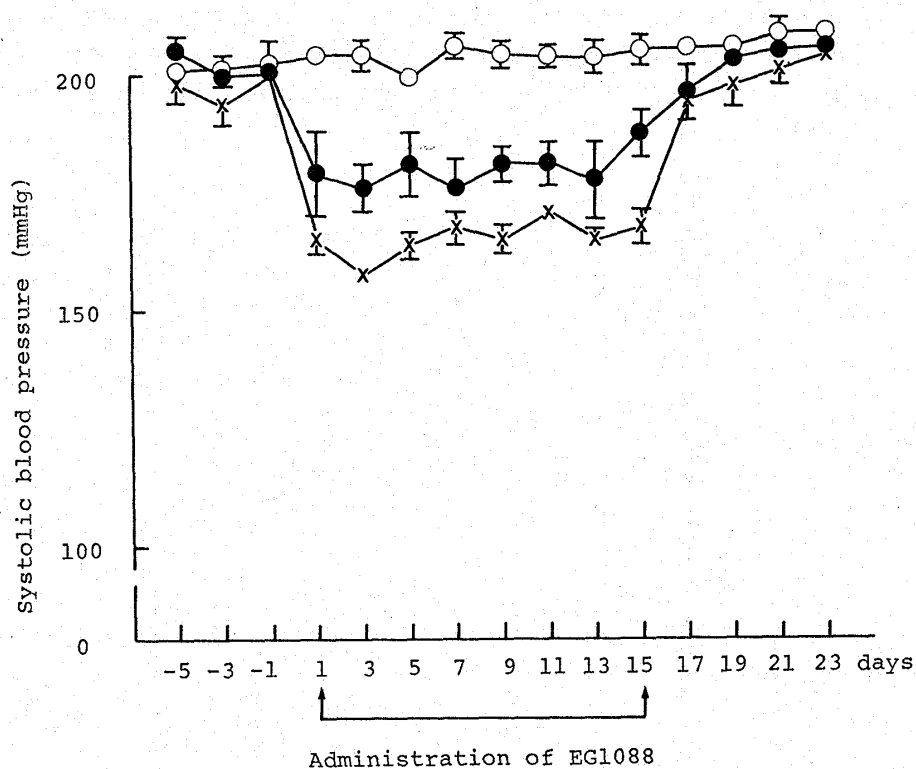


Fig. 2. Anti-hypertensive Effect of EG1088 during 15 Days of Successive Administration to Groups of 6 Male SHR

EG1088 in doses of 3 (●—●) and 10 (x—x) mg/kg was administered p.o. once daily for 15 days. Vehicle (0.5% sodium carboxymethyl cellulose) was given to the control group (o—o). The systolic blood pressure was measured every other day 4 h after administration of the drug or vehicle. Vertical bars represent S.E.

The hypotensive action of la was approximately 10 times less potent than that of lb and lc. Experiments to clarify the mechanisms of the hypotensive action of lb and lc compared with the prototype compound, EG1088 (la), are illustrated below. The hypotensive effect of la was slightly but significantly ($p < 0.05$) reduced in anesthetized rabbits after transection of the spinal cord at the C_1 level and the bilateral cervical vagal nerves. In contrast, the hypotensive effect of diltiazem remained unchanged and the effect of clonidine was abolished and converted to a significant ($p < 0.01$) pressor response after the transection (Fig. 3). Thus, it seems likely that the hypotensive effect of la is partly due to its action on the CNS. On the other hand, the observation that the closed arterial injection of la (0.1 to 100 μ g) produced a dose-dependent decrease in the perfusion pressure in the hind limb of a rabbit (perfused at a constant flow rate with peristaltic pump) indicate that peripheral vasodilation may contribute to the hypotensive effect of la. The hypotensive effect of la was not altered by atropine or propranolol in doses sufficient to antagonize the depressor responses to acetylcholine and isoproterenol. This suggests that the effect of la is not mediated through excitation of adrenergic beta-receptors or muscarinic receptors.

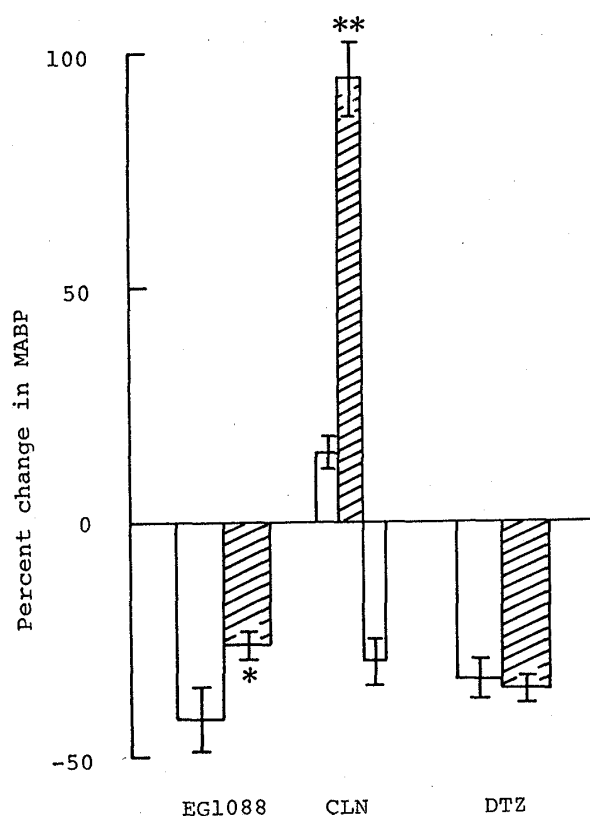


Fig. 3. Effect of Transection of the Spinal Cord and Bilateral Cervical Vagal Nerves on the Hypotensive Effects of EG1088, Clonidine (CLN) and Diltiazem (DTZ) in Anesthetized Rabbit

Mean arterial blood pressure (MABP) was calculated as percentages of the values obtained before the drug administration. The doses of EG1088, clonidine and diltiazem were 10, 10 and 300 μ g/kg, i.v., respectively. $\square$: Intact, hatched : Transection of the spinal cord at C_1 level and the bilateral cervical vagal nerves. *: $p < 0.05$, **: $p < 0.01$.

Compound la produced a concentration-dependent relaxation of rabbit arterial strips contracted with 30 mM KCl. The relaxing activity of la was most potent in the basilar artery ($IC_{50} = 4.4 \times 10^{-6}$ M) and least in the thoracic aorta ($IC_{50} = 4.1 \times 10^{-5}$ M). The concentration-response curve for KCl in the thoracic aorta was shifted to the right

and downward by 1a and the pD_2' value was calculated to be 4.3. However, the agent did not affect the concentration-response curve for norepinephrine in the thoracic aorta. The calcium contraction in Ca^{++} -free and K^+ -depolarized thoracic aorta was noncompetitively inhibited by 1a and a pD_2' value of 4.3 was obtained. Compound 1a inhibited the contractile responses of the thoracic aorta to 10^{-4} M nicotine and 10^{-4} M tyramine, but had no effect on the contraction induced by norepinephrine. The IC_{50} values to nicotine and tyramine were 3.3×10^{-6} M and 6.1×10^{-5} M, respectively. Adenosine-induced relaxation in the thoracic aorta contracted with norepinephrine was significantly potentiated by 1a (pC_{50} : Concentration which potentiates adenosine relaxation by 50% = 4.9×10^{-6} M). It is therefore speculated that 1a possesses calcium antagonistic action and inhibitory effect on norepinephrine release and/or nicotinic blocking action, but lacks adrenergic alpha-receptor blocking action.

Compound 1a in a concentration of 10^{-5} M or less had little effect on spontaneous movement of the right atrium isolated from rabbits. In a higher concentration of 10^{-4} M, however, the agent reduced the heart rate and contractile force by approximately 20%. A positive chronotropic response induced by 10^{-4} M tyramine and a negative chronotropic response followed by a positive one induced by 10^{-4} M nicotine were significantly inhibited by 1a and the IC_{50} values were calculated to be 1.4×10^{-5} M and 3.1×10^{-5} M, respectively. The concentration-chronotropic response curve for isoproterenol was shifted to the right and downward by pretreatment with 1a and a pD_2' value of 5.3 was obtained. Thus, 1a probably possesses the functional beta-blocking action seen with calcium antagonists.⁵⁾

In conclusion, the hypotensive effect of 1a-c may be at least partly due to a calcium antagonistic action, inhibition of norepinephrine release, potentiation of adenosine response, effects on CNS, nicotinic blocking action and a functional beta-blocking action, but the hypotensive effect is not mediated through excitation of adrenergic beta-receptors or muscarinic receptors. The detailed mechanism of action and preclinical examination of these new agents will be reported elsewhere.

ACKNOWLEDGEMENT

The authors are grateful to M. Ohara for comments on the manuscript.

REFERENCES AND NOTES

- 1) This work was presented at the 3rd Symposium on Medicinal Chemistry, Osaka November 1981.
- 2) M. Ishikawa and Y. Eguchi, *Heterocycles*, **16**, 31 (1981).
- 3) H. W. Grimmel, A. Guenther and J. F. Morgan, *J. Am. Chem. Soc.*, **68**, 542 (1964).
- 4) H. Ishii, K. Itoh and T. Nose, *Europ. J. Pharmacol.*, **64**, 21 (1980).
- 5) a) M. Briley, I. Caverio, S. Z. Langer and A. G. Roach, *Brit. J. Pharmacol.*, **69**, 669 (1980); b) M. Wehling, K. R. Whitmer, A. Schwartz, G. Grupp, I. Grupp and E. T. Wallick, *Fedn. Proc.*, **39**, 626 (1980).

(Received October 21, 1981)