Haemodynamic profile of the potassium channel activator EMD 52692 in anaesthetized pigs

L.M.A. Sassen, D.J.G.M. Duncker, B.C.G. Gho, *H.W. Diekmann & ¹P.D. Verdouw

Laboratory for Experimental Cardiology (Thoraxcentre), Erasmus University Rotterdam, Rotterdam, The Netherlands and

*Merck Institute for Experimental Drug Research, Grafting bei München, Federal Republic of Germany

1 The systemic and regional haemodynamic effects of the potassium channel activator EMD 52692 or its solvent were investigated after intravenous and after intracoronary administration in anaesthetized pigs.

2 Consecutive intravenous 10 min infusions of EMD 52692 (0.15, 0.30, 0.60, 1.20 $\mu\text{g kg}^{-1} \text{min}^{-1}$; $n = 7$) dose-dependently decreased mean arterial blood pressure by up to 50%. This was entirely due to peripheral vasodilatation, since cardiac output did not change. Heart rate increased by up to 50%, while left ventricular end diastolic pressure decreased dose-dependently from $6 \pm 1 \text{ mmHg}$ to $3 \pm 1 \text{ mmHg}$ ($P < 0.05$), and stroke volume decreased from $30 \pm 2 \text{ ml}$ to $21 \pm 2 \text{ ml}$ ($P < 0.05$). Left ventricular dP/dt_{max} was not affected.

3 Although cardiac output did not change, EMD 52692 caused a redistribution of blood flow from the arteriovenous anastomoses to the capillary channels. Blood flow to the adrenals, small intestine, stomach, bladder, spleen and brain increased, while renal blood flow decreased and blood flow to several muscle groups and skin were not altered. Vascular conductance was increased dose-dependently in all organs, except for the kidneys, where after the initial increase, vascular conductance returned to baseline with the highest dose. Particularly striking were the effects on the vasculature of the brain. With the highest dose of EMD 52692 blood flow more than doubled, while vascular conductance increased four fold.

4 Transmural myocardial blood flow increased slightly, which was entirely due to an increase in sub-epicardial blood flow. Myocardial O_2 -consumption and segment length shortening were not significantly affected.

5 After consecutive 10 min intracoronary infusions (0.0095, 0.019, 0.0375 and 0.075 $\mu\text{g kg}^{-1} \text{min}^{-1}$; $n = 7$) into the left anterior descending coronary artery (LADCA), mean arterial blood pressure was maintained with the lowest two doses, but decreased by up to 15% with the higher doses, whereas heart rate increased by up to 24%. Blood flow to the LADCA-perfused myocardium doubled with the highest dose, the subepicardium benefitting the most. Coronary venous O_2 -saturation increased dose-dependently from $23 \pm 2\%$ to $60 \pm 4\%$, while myocardial O_2 -consumption of the LADCA-perfused myocardium was not affected by the drug.

6 It is concluded that EMD 52692 is a potent vasodilator, with particularly pronounced effects on vasculature of the brain. Its selectivity for vascular smooth muscle cells exceeds that for the myocytes, since with doses that are much higher than those of potential clinical interest no negative inotropic effects were observed. The compound primarily dilates arteries but some venodilatation may also occur.

Introduction

Diverse pharmacological treatments can have equal hypotensive effects in essential hypertension, despite apparently distinct modes of action (e.g. Ca^{2+} antagonism, angiotensin converting enzyme-inhibition). Recently, a new class of vasodilators, the potassium channel activators, has been introduced. These drugs cause hyperpolarization of the membranes of vascular smooth muscle cells, which reduces the availability of Ca^{2+} and thereby attenuates contraction (Hamilton *et al.*, 1986). Furthermore, the cells become less sensitive to depolarizing stimuli (Clapham & Wilson, 1987; Cook, 1988; 1989). The vasodilator and cardiac effects of drugs like cromakalim (Hamilton *et al.*, 1986; Weir & Weston, 1988; Cook & Hof, 1988) and pinacidil (Bray *et al.*, 1987) and also the cardiac effects of nicorandil (Weir & Weston, 1988) are mediated by opening of adenosine 5'-triphosphate (ATP)-dependent potassium channels. These potassium channels have been shown to be important in mediating the hypoxic dilatation of coronary arteries (Daut *et al.*, 1990).

Cromakalim, at doses causing similar decreases in arterial blood pressure as nifedipine, exhibits a vasodilator profile which is clearly different from that of the calcium antagonists (Buckingham *et al.*, 1986). The aim of the present study was to

investigate the systemic haemodynamics and the distribution of cardiac output (radioactive microsphere technique) in response to intravenous infusions of EMD 52692 in the anaesthetized pig. EMD 52692 (Figure 1), a novel benzopyran derivative, structurally identical to SR 44866 (Findlay *et al.*, 1989), has been shown to hyperpolarize smooth muscle cells, which has been attributed to activation of ATP-dependent potassium channels (Findlay *et al.*, 1989; De Peyer *et al.*, 1989; Gericke *et al.*, 1989). In order to distinguish direct from indirect (secondary to systemic haemodynamic changes)

Figure 1 Chemical structure of EMD 52692.

¹ Author for correspondence at Laboratory for Experimental Cardiology, Thoraxcentre, Erasmus University Rotterdam, P.O. Box 1738, 3000 DR Rotterdam.

effects, we also studied the effects of EMD 52692 on myocardial performance after intracoronary infusion of the compound.

Methods

General

After an overnight fast, cross-bred Landrace $\times$ Yorkshire pigs of either sex (22–28 kg, $n = 29$) were sedated with 120 mg azaperone (Stresnil) i.m., anaesthetized with 150 mg metomidate (Hypnodil) i.v., intubated and connected to a ventilator for intermittent positive pressure ventilation with a mixture of O_2 and N_2O (1:2, v/v). Respiratory rate and tidal volume were set to keep arterial blood gases within the normal range: $7.41 < pH < 7.44$; $42 \text{ mmHg} < PCO_2 < 45 \text{ mmHg}$ and $130 \text{ mmHg} < PO_2 < 170 \text{ mmHg}$. 7F catheters were placed in the superior caval vein for administration of 160 mg kg^{-1} α -chloralose followed by an infusion of a low dose of sodium pentobarbitone ($5 \text{ mg kg}^{-1} \text{ h}^{-1}$); for administration of the muscle relaxant pancuronium bromide (4 mg) prior to thoracotomy; and for administration of haemaccel (Behringwerke A.G., Marburg, F.R.G.) to replace blood loss. Catheters were also positioned in the descending aorta for withdrawal of blood samples and measurement of central aortic blood pressure. A micromanometer-tipped catheter (7F Millar), inserted via the left carotid artery, was used to measure left ventricular pressure and its first derivative (LV dP/dt). After thoracotomy, an electromagnetic flow probe (Skalar, Delft, The Netherlands) was placed around the ascending aorta, while the great cardiac vein was cannulated for collection of blood in which the haemoglobin concentration and O_2 -saturation were determined (OSM2, Radiometer, Copenhagen, Denmark). In 15 of the animals the proximal left anterior descending coronary artery was dissected free for placement of an electromagnetic flow probe around the vessel, while a cannula (outer diameter 0.8 mm) was inserted for intracoronary infusions of EMD 52692.

Regional blood flows

In order to determine regional blood flows, the left atrial appendage was cannulated for injection of a batch of $1\text{--}2 \times 10^6$ carbonized plastic microspheres [$15 \pm 1 \mu\text{m}$ (s.d.) in diameter] labelled with either ^{46}Sc , ^{95}Nb , ^{103}Ru , ^{113}Sn or ^{141}Ce . Fifteen s before the injection of microspheres, blood was withdrawn from a femoral artery at a rate of 10 ml min^{-1} until 60–65 s after completion of the injection of the microspheres. At the end of each experiment in which the animals received intracoronary infusions of either EMD 52692 or its solvent (see below), the area perfused by the left anterior descending coronary artery was identified by intracoronary injection of patent blue violet (Sigma, St. Louis, MO, U.S.A.). All the animals were killed with an overdose of sodium pentobarbitone and from the i.v. treated animals (see below) various organs (adrenals, bladder, stomach, small intestine, brains and kidneys) and tissues (abdominal skin, different muscle groups) were excised, weighed and put into vials. In the animals treated with intracoronary infusions only the heart was excised. The hearts were fixed in formaldehyde (10% v/v) and 48 h later divided into atria and right and left ventricle. The myocardium of the left ventricle was divided into three layers of equal thickness: subepicardium, mesocardium and subendocardium. The kidneys were divided into medulla and three cortical layers of equal thickness. The cerebral hemispheres, cerebellum, diencephalon and brain stem were separated in order to obtain regional blood flow data from the different parts of the brain.

The radioactivity was counted and the amount of blood flow to the various tissues (Q_{tis}) calculated as:

$$Q_{tis}(\text{ml min}^{-1}) = (I_{tis}/I_{art}) \times Q_{art},$$

where I_{tis} and I_{art} are, respectively, the radioactivity (c.p.m.) in a particular tissue and that of the arterial blood sample, while

Q_{art} is the rate of withdrawal of the blood sample. Although the lungs receive microspheres via both peripheral arteriovenous anastomoses and bronchial arteries, the contribution via the latter route appears to be only about 1% of cardiac output (Baile *et al.*, 1982). Thus, the values for the lung blood flow can be used as an index of the arteriovenous anastomotic flow (i.e. non-nutrient part of the cardiac output). The nutrient part of the cardiac output was calculated by subtracting 'lung flow' from the cardiac output. All regional vascular conductances were calculated as the ratio between Q_{tis} and the mean arterial blood pressure. Full details of the procedures and the calculation of flow data using this technique have been described earlier (Saxena *et al.*, 1980).

Myocardial oxygen consumption and contractile function

Myocardial O_2 -consumption ($\dot{M}VO_2$) was calculated as the product of coronary blood flow and the difference in the O_2 contents of the arterial and coronary venous blood.

Regional myocardial segment length shortening was measured with sonomicrometry (Triton Technology Inc., San Diego, CA, U.S.A.) by a pair of ultrasonic crystals implanted approximately 10–15 mm apart in the subepicardial layers (during intracoronary infusions) or the midmyocardial layers (during the intravenous infusions), oriented parallel to the short axis (Freeman *et al.*, 1985). In order to differentiate between the effect of EMD 52692 on the different layers of the myocardium, another pair of crystals was also implanted in the subendocardial layers of the animals which received the intracoronary infusions of EMD 52692. At the end of each experiment the position of the subendocardial crystals was confirmed. From the tracings, segment length shortening (SLS) was calculated as: $\text{SLS (\%)} = 100 \times (\text{EDL} - \text{ESL}) / \text{EDL}$, in which EDL and ESL are the segment length at end-diastole and end-systole, respectively.

Experimental protocols

Four consecutive 10 min intravenous infusions of EMD 52692 ($0.15, 0.3, 0.6$ and $1.2 \mu\text{g kg}^{-1} \text{ min}^{-1}$; total cumulative doses of $1.5, 4.5, 10.5$ and $22.5 \mu\text{g kg}^{-1}$; $n = 7$) or equal volumes of the solvent ($n = 7$) or four consecutive 10 min intracoronary infusions of EMD 52692 ($0.0095, 0.019, 0.0375$ and $0.075 \mu\text{g kg}^{-1} \text{ min}^{-1}$; total cumulative doses of $0.10, 0.29, 0.66$ and $1.41 \mu\text{g kg}^{-1}$; $n = 7$) or equal volumes of the solvent ($n = 8$) were administered. Systemic haemodynamics, regional myocardial function and the distribution of coronary blood flow were determined in all four series of experiments, but the distribution of cardiac output was only determined during the intravenous infusions of EMD 52692 and its solvent.

Determination of EMD 52692 plasma concentrations

At the end of each infusion step blood samples (5 ml) were collected in heparinized tubes for the determination of plasma levels of EMD 52692. The blood samples were centrifuged at $1500g$ at 0°C for 10 min and the plasma (2–3 ml) was stored at -80°C until further analysis. Known amounts of hexadeuterated EMD 52692 were added as internal standard to the plasma samples. EMD 52692 and internal standard were extracted with isopropylether. The extracts were evaporated to dryness under nitrogen, taken up in 20 to $60 \mu\text{l}$ of methanol, and aliquots of $1 \mu\text{l}$ analysed by gas chromatography/mass fragmentography (Hewlett Packard GC 5890 II and autointegrator HP 7673).

Drugs

For the i.v. infusions, EMD 52692 (4-(1,2-dihydro-2-oxo-1-pyridyl)-2,2-dimethyl-2H-1-benzopyran-6-carbonitrile; courtesy Dr P. Schelling, Merck Darmstadt, F.R.G.) was dissolved in 4% (v/v) polyethylene glycol for the two highest (0.60 and $1.20 \mu\text{g kg}^{-1} \text{ min}^{-1}$) doses and subsequently further diluted

with NaCl (0.9%) to give the two lower (0.15 and $0.30 \mu\text{g kg}^{-1} \text{min}^{-1}$) doses. The infusion rate was 1 ml min^{-1} for the lowest and the third, and 2 ml min^{-1} for the second and the highest dose. For the intracoronary infusions EMD 52692 was dissolved in 1% (v/v) polyethylene glycol and the required doses were reached by adjusting the infusion rate (from 0.25 ml min^{-1} to 2 ml min^{-1}).

Statistical evaluation

All data are presented as the arithmetic mean $\pm$ s.e.mean. The significance of the effects of the solvent or EMD 52692 on the different variables was evaluated by Duncan's new-multiple range-test once an analysis of variance (randomized block design) had revealed that the samples represented different populations. Statistical significance was defined as $P < 0.05$ (two-tailed).

Results

Intravenous infusions of EMD 52692

Plasma concentrations of EMD 52692 As the infusion rates (0.15, 0.30, 0.75 and $1.5 \mu\text{g kg}^{-1} \text{min}^{-1}$) of EMD 52692 were increased, the arterial plasma concentrations reached levels of 5.9 ± 0.5 , 14.6 ± 1.1 , 30.8 ± 1.1 and $62.5 \pm 2.3 \mu\text{g l}^{-1}$, respectively.

Systemic haemodynamics No significant changes occurred during infusion of the solvent in any of the systemic haemodynamic variables (Table 1). EMD 52692 lowered the arterial blood pressure dose-dependently (by 51% with the highest dose). This fall in arterial blood pressure was caused by systemic vasodilatation (fall in systemic vascular resistance of 52%) as cardiac output was not affected. Cardiac output was maintained, but stroke volume decreased (by up to 30%, $P < 0.05$), most likely secondary to the fall in left ventricular filling pressure (from $6 \pm 1 \text{ mmHg}$ to $3 \pm 1 \text{ mmHg}$, $P < 0.05$). Heart rate increased dose-dependently from $103 \pm 7 \text{ beats min}^{-1}$ to $151 \pm 13 \text{ beats min}^{-1}$ ($P < 0.05$) with the highest dose. $\text{LVdP/dt}_{\text{max}}$ was not affected.

Left ventricular blood flow and performance The solvent had no effect on the transmural blood flow and its distribution over the subendocardial and the subepicardial layers or on the

Figure 2 Effect of cumulative 10 min intravenous infusions (total dose 0, 1.5, 4.5, 10.5 and $22.5 \mu\text{g kg}^{-1}$) of EMD 52692 (hatched columns, $n = 7$) or equal volumes of its solvent (open columns, $n = 7$) on transmural (a), subendocardial (b) and subepicardial (c) left ventricular blood flows and conductances in anaesthetized pigs. Data are presented as mean and the bars show s.e.mean. * Indicates that the drug-induced changes from baseline are significantly different ($P < 0.05$) from the solvent-induced changes from baseline.

coronary vascular conductance (Figure 2). Infusions of EMD 52692 had no significant effect on transmural left ventricular blood flow, but with the highest two doses there was a redistribution in favour of the subepicardium, as the flow to the subepicardial layers increased dose-dependently from $154 \pm 14 \text{ ml min}^{-1} 100 \text{ g}^{-1}$ to $257 \pm 18 \text{ ml min}^{-1} 100 \text{ g}^{-1}$ ($P < 0.05$), and flow to the subendocardium did not change. Vasodilatation occurred in all layers of the myocardium as indicated by an increase in vascular conductance. The increase in the subepicardium (by up to $268 \pm 41\%$) was more pronounced than the increase (by up to $65 \pm 16\%$) in the subendocardium (Figure 2).

In the solvent-treated animals there was a minor narrowing

Table 1 Systemic haemodynamics after cumulative intravenous 10 min infusions of EMD 52692 or its solvent in anaesthetized pigs

		EMD 52692 ($\mu\text{g kg}^{-1}$) or equal volumes of its solvent				
		0	1.5	4.5	10.5	22.5
MAP	Solvent	89 ± 5	88 ± 4	88 ± 4	90 ± 4	88 ± 4
	EMD 52692	90 ± 3	$82 \pm 2^*$	$65 \pm 2^*$	$51 \pm 4^*$	$44 \pm 4^*$
APP	Solvent	43 ± 2	46 ± 2	45 ± 2	46 ± 2	45 ± 2
	EMD 52692	42 ± 3	39 ± 3	39 ± 3	42 ± 3	44 ± 4
CO	Solvent	2.8 ± 0.2	2.8 ± 0.2	2.8 ± 0.2	2.7 ± 0.2	2.7 ± 0.2
	EMD 52692	3.0 ± 0.2	3.1 ± 0.2	3.0 ± 0.2	3.0 ± 0.3	3.1 ± 0.2
SVR	Solvent	34 ± 4	33 ± 4	33 ± 3	35 ± 3	34 ± 3
	EMD 52692	31 ± 3	28 ± 2	$22 \pm 2^*$	$18 \pm 2^*$	$15 \pm 2^*$
HR	Solvent	99 ± 6	96 ± 6	99 ± 8	98 ± 6	99 ± 6
	EMD 52692	103 ± 7	$111 \pm 8^*$	$129 \pm 7^*$	$141 \pm 12^*$	$151 \pm 13^*$
$\text{LVdP/dt}_{\text{max}}$	Solvent	2670 ± 170	2610 ± 130	2650 ± 130	2540 ± 150	2530 ± 140
	EMD 52692	2710 ± 120	2630 ± 120	2880 ± 130	2730 ± 280	2800 ± 300
LVEDP	Solvent	6 ± 1	6 ± 1	6 ± 1	6 ± 1	7 ± 1
	EMD 52692	6 ± 1	$5 \pm 1^*$	$4 \pm 1^*$	$3 \pm 1^*$	$3 \pm 1^*$
SV	Solvent	28 ± 1	29 ± 1	29 ± 2	27 ± 1	27 ± 1
	EMD 52692	30 ± 2	$28 \pm 2^*$	$23 \pm 2^*$	$22 \pm 2^*$	$21 \pm 2^*$

Data are mean $\pm$ s.e.mean; $n = 7$ for both groups. MAP = mean arterial blood pressure (mmHg); APP = arterial pulse pressure (mmHg); CO = cardiac output (l min^{-1}); SVR = systemic vascular resistance (mmHg l min^{-1}); HR = heart rate (beats min^{-1}); $\text{LVdP/dt}_{\text{max}}$ = maximal rate of rise of left ventricular pressure (mmHg s^{-1}); LVEDP = left ventricular end diastolic pressure (mmHg); SV = stroke volume (ml).

* Changes versus baseline in the EMD 52692-treated animals are significantly ($P < 0.05$) different from those in the solvent-treated animals.

Figure 3 Effect of cumulative 10 min intravenous infusions of EMD 52692 (hatched columns, total dose 0, 1.5, 4.5, 10.5 and 22.5 µg kg⁻¹, *n* = 7) or equal volumes of its solvent (open columns, *n* = 7) on (a) coronary venous O₂-saturation (cvO₂-sat) and (b) left ventricular O₂-consumption (MVO₂). Data are presented as mean and the bars show s.e.mean. * Indicates that the drug-induced changes from baseline are significantly different (*P* < 0.05) from the solvent-induced changes from baseline.

of the difference in the O₂ contents of the arterial and coronary venous blood (5%, *P* < 0.05), reflected by the small increase (from 20 ± 1% to 24 ± 2%) in the O₂-saturation in the great cardiac vein (Figure 3). With EMD 52692, the decrease (35% after the highest infusion rate) in O₂-extraction was considerably large as the coronary venous O₂-saturation almost tripled (from 17 ± 1% to 45 ± 5% with the highest

Figure 4 Fractionation of cardiac output in arteriovenous anastomotic (open columns) and capillary (hatched columns) blood flow during cumulative 10 min intravenous infusions of EMD 52692 (total dose 0, 1.5, 4.5, 10.5 and 22.5 µg kg⁻¹, *n* = 7) or equal volumes of its solvent (*n* = 7) in anaesthetized pigs. Data are presented as mean and the bars show s.e.mean. * Indicates that the drug-induced changes from baseline are significantly different (*P* < 0.05) from the solvent-induced changes from baseline.

dose, *P* < 0.05). Myocardial O₂-consumption was not affected by the solvent or by the drug. Data are presented in Fig. 3.

Myocardial segment length shortening, determined from the crystals placed in the midmyocardium, did not change either

Figure 5 Effect of cumulative 10 min intravenous infusions (total dose 0, 1.5, 4.5, 10.5 and 22.5 µg kg⁻¹) of EMD 52692 (hatched columns, *n* = 7) and equal volumes of its solvent (open columns, *n* = 7) on atrial (a) and right ventricular blood flow (b) and conductance in anaesthetized pigs. Data are presented as mean and the bars show s.e.mean. * Indicates that the drug-induced changes from baseline are significantly different (*P* < 0.05) from the solvent-induced changes from baseline.

during infusion of the solvent ($19 \pm 1\%$, $18 \pm 1\%$, $19 \pm 1\%$, $19 \pm 1\%$ and $20 \pm 2\%$ at baseline and after 10, 20, 30 and 40 min, respectively) or during infusion of EMD 52692 ($20 \pm 1\%$, $19 \pm 1\%$, $18 \pm 2\%$ and $18 \pm 1\%$ at baseline, and after cumulative infusions of 0.15, 0.30, 0.60 and $1.2 \mu\text{g kg}^{-1} \text{min}^{-1}$ of EMD 52692, respectively).

Distribution of cardiac output

Fractionation of cardiac output into arteriovenous anastomotic and capillary flows In anaesthetized animals a large fraction of the cardiac output is shunted through arteriovenous anastomoses (Kaiharu *et al.*, 1968; Van Woerkens *et al.*, 1990) and without affecting total cardiac output, antihypertensive drugs may affect this non-nutritional flow differently (Hof & Hof, 1989). Figure 4 illustrates that, under baseline conditions, in both groups approximately 40% of the cardiac output was shunted through arteriovenous anastomoses. This fraction did not change during infusion of the solvent. Although EMD 52692 did not affect cardiac output, there was a redistribution in favour of the capillary flow, which increased to 85% of cardiac output after the highest dose.

Regional blood flows Blood flow to the atria and the right ventricle increased dose-dependently during the EMD 52692 infusions (Figure 5). Taking into account the decrease in perfusion pressure it is clear that the increments in vascular conductance were even more pronounced (Figure 5). EMD 52692 caused pronounced increases in total cerebral blood flow (32 ± 3 , 34 ± 2 , 43 ± 3 , 66 ± 7 and $69 \pm 7 \text{ ml min}^{-1} 100 \text{ g}^{-1}$ at baseline and after 1.5, 4.5, 10.5 and $22.5 \mu\text{g kg}^{-1}$, respectively). Although flow to all parts of the brain increased (Figure 6), the increase was more pronounced in the brain stem ($216 \pm 24\%$) and diencephalon ($178 \pm 40\%$) than in the cerebellum ($113 \pm 14\%$) and the hemispheres ($100 \pm 10\%$). The increases in regional conductances were even more impressive than the increases in flow (Figure 6).

Total renal blood flow remained constant with the two lowest doses, but decreased by $43 \pm 7\%$ after infusion of the highest dose (Table 2). Further analysis revealed that this was predominantly due to a dose-dependent decrease in glomerular blood flow of the outer cortex, as the glomerular blood flow of the inner cortex decreased only slightly and flow to the medulla even increased (Table 2). The decreases in flow were secondary to the fall in arterial blood pressure, as total renal conductance increased significantly by as much as

Figure 6 Effect of cumulative 10 min infusions of EMD 52692 (hatched columns, total dose 0, 1.5, 4.5, 10.5 and $22.5 \mu\text{g kg}^{-1}$, $n = 7$) or equal volumes of its solvent (open columns; $n = 7$) on blood flow and conductance of the cerebral hemispheres (a), diencephalon (b), cerebellum (c) and brain stem (d) in anaesthetized pigs. Data are presented as mean and the bars show s.e.mean. * Indicates that the drug-induced changes from baseline are significantly different ($P < 0.05$) from the solvent-induced changes from baseline.

$110 \pm 50\%$ after the third dose. With the highest dose renal vascular conductance started to return to baseline values. The increases in vascular conductance, observed in the inner

Table 2 Effect of continuous intravenous 10 min infusions of EMD 52692 or its solvent on renal blood flow and conductance in anaesthetized pigs

		EMD 52692 ($\mu\text{g kg}^{-1}$) or equal volumes of its solvent				
Blood flow ($\text{ml min}^{-1} 100 \text{ g}^{-1}$)		0	1.5	4.5	10.5	22.5
Kidneys	Solvent	252 $\pm$ 22	267 $\pm$ 35	253 $\pm$ 24	249 $\pm$ 24	236 $\pm$ 15
	EMD 52692	247 $\pm$ 19	225 $\pm$ 14	228 $\pm$ 16	191 $\pm$ 11*	137 $\pm$ 17*
Glomeruli of the inner cortex	Solvent	210 $\pm$ 18	210 $\pm$ 19	202 $\pm$ 16	190 $\pm$ 13	183 $\pm$ 11
	EMD 52692	163 $\pm$ 10	161 $\pm$ 10	178 $\pm$ 9	155 $\pm$ 9	115 $\pm$ 13
Glomeruli of the outer cortex	Solvent	393 $\pm$ 25	407 $\pm$ 32	405 $\pm$ 32	412 $\pm$ 33	387 $\pm$ 26
	EMD 52692	386 $\pm$ 31	333 $\pm$ 19	316 $\pm$ 25*	258 $\pm$ 18*	179 $\pm$ 23*
Medulla	Solvent	58 $\pm$ 7	59 $\pm$ 6	53 $\pm$ 5	46 $\pm$ 5	46 $\pm$ 5
	EMD 52692	42 $\pm$ 5	44 $\pm$ 5	55 $\pm$ 7*	58 $\pm$ 5*	55 $\pm$ 6*
Conductance ($\text{ml min}^{-1} 100 \text{ g}^{-1} \text{ mmHg}^{-1}$)						
Kidneys	Solvent	2.89 $\pm$ 0.31	3.14 $\pm$ 0.25	2.95 $\pm$ 0.34	2.79 $\pm$ 0.31	2.67 $\pm$ 0.17
	EMD 52692	2.78 $\pm$ 0.23	2.74 $\pm$ 0.17	3.35 $\pm$ 0.33*	3.88 $\pm$ 0.33*	3.19 $\pm$ 0.34
Glomeruli of the inner cortex	Solvent	2.40 $\pm$ 0.25	2.54 $\pm$ 0.22	2.33 $\pm$ 0.22	2.12 $\pm$ 0.18	2.08 $\pm$ 0.14
	EMD 52692	1.84 $\pm$ 0.13	1.96 $\pm$ 0.12	2.79 $\pm$ 0.18*	3.18 $\pm$ 0.35*	2.73 $\pm$ 0.36*
Glomeruli of the outer cortex	Solvent	4.43 $\pm$ 0.34	4.89 $\pm$ 0.32	4.62 $\pm$ 0.41	4.62 $\pm$ 0.42	4.37 $\pm$ 0.20
	EMD 52692	4.32 $\pm$ 0.35	4.05 $\pm$ 0.19	4.95 $\pm$ 0.48	5.22 $\pm$ 0.43	4.14 $\pm$ 0.45
Medulla	Solvent	0.67 $\pm$ 0.08	0.68 $\pm$ 0.07	0.62 $\pm$ 0.07	0.51 $\pm$ 0.06	0.53 $\pm$ 0.06
	EMD 52692	0.48 $\pm$ 0.05	0.53 $\pm$ 0.06	0.84 $\pm$ 0.09*	1.16 $\pm$ 0.11*	1.27 $\pm$ 0.11*

Data are mean $\pm$ s.e.mean; $n = 7$ for both groups. * Changes versus baseline in the EMD 52692-treated animals are significantly ($P < 0.05$) different from those in the solvent-treated animals.

Table 3 Effect of continuous intravenous 10 min infusions of EMD 52692 or its solvent on regional blood flow and conductance in anaesthetized pigs

Blood flow (ml min ⁻¹ 100 g ⁻¹)		EMD 52692 (µg kg ⁻¹) or equal volumes of its solvent				
		0	1.5	4.5	10.5	22.5
Spleen	Solvent	271 ± 46	317 ± 52	273 ± 43	254 ± 55	267 ± 56
	EMD 52692	255 ± 23	311 ± 31	453 ± 36	375 ± 42	272 ± 31
Adrenals	Solvent	131 ± 16	139 ± 14	142 ± 7	144 ± 12	149 ± 22
	EMD 52692	180 ± 27	220 ± 23	332 ± 20*	291 ± 19*	231 ± 14*
Stomach	Solvent	21 ± 1	20 ± 1	23 ± 2	21 ± 2	20 ± 2
	EMD 52692	21 ± 2	20 ± 1	19 ± 2	21 ± 3	29 ± 5
Small Intestine	Solvent	38 ± 3	37 ± 2	36 ± 2	39 ± 2	36 ± 2
	EMD 52692	44 ± 5	46 ± 8	51 ± 9	62 ± 9	77 ± 7*
Skin	Solvent	1.9 ± 0.5	1.9 ± 0.9	1.5 ± 0.4	2.0 ± 0.7	2.3 ± 1.0
	EMD 52692	2.4 ± 0.4	2.7 ± 0.5	2.2 ± 0.5	2.6 ± 0.4	2.5 ± 0.3
Bladder	Solvent	3.5 ± 0.3	4.3 ± 0.4	4.0 ± 0.4	3.9 ± 0.3	3.9 ± 0.2
	EMD 52692	5.0 ± 0.5	4.9 ± 0.5	4.5 ± 0.6	4.5 ± 0.5	7.6 ± 0.9
Conductance (ml min ⁻¹ 100 g ⁻¹ mmHg ⁻¹)						
Spleen	Solvent	3.13 ± 0.60	3.70 ± 0.60	3.16 ± 0.51	2.84 ± 0.65	3.05 ± 0.64
	EMD 52692	2.83 ± 0.23	3.79 ± 0.35	7.00 ± 0.38*	7.30 ± 0.54*	6.18 ± 0.39*
Adrenals	Solvent	1.53 ± 0.22	1.72 ± 0.14	1.64 ± 0.11	1.60 ± 0.14	1.67 ± 0.19
	EMD 52692	2.04 ± 0.32	2.70 ± 0.30	5.16 ± 0.36*	5.83 ± 0.42*	5.45 ± 0.44*
Stomach	Solvent	0.24 ± 0.02	0.25 ± 0.02	0.26 ± 0.02	0.23 ± 0.02	0.23 ± 0.03
	EMD 52692	0.24 ± 0.03	0.25 ± 0.02	0.31 ± 0.03	0.42 ± 0.06*	0.67 ± 0.11*
Small Intestine	Solvent	0.44 ± 0.05	0.44 ± 0.03	0.42 ± 0.030	0.44 ± 0.03	0.42 ± 0.03
	EMD 52692	0.49 ± 0.06	0.56 ± 0.09	0.81 ± 0.017	1.23 ± 0.16*	1.79 ± 0.18*
Skin	Solvent	0.022 ± 0.007	0.022 ± 0.012	0.018 ± 0.005	0.023 ± 0.008	0.028 ± 0.013
	EMD 52692	0.027 ± 0.005	0.033 ± 0.007	0.036 ± 0.009	0.051 ± 0.007*	0.058 ± 0.006*
Bladder	Solvent	0.041 ± 0.004	0.066 ± 0.006	0.046 ± 0.002	0.044 ± 0.004	0.044 ± 0.003
	EMD 52692	0.056 ± 0.006	0.059 ± 0.006	0.072 ± 0.013	0.090 ± 0.011*	0.178 ± 0.022*

Data are mean ± s.e.mean; *n* = 7 for both groups. Data are in ml min⁻¹ 100 g⁻¹ mmHg⁻¹; * changes versus baseline in the EMD 52692-treated animals are significantly different (*P* < 0.05) from those in the solvent-treated animals.

cortex and the medulla were not observed in the outer cortex (Table 2).

Flow to a number of other organs was well maintained (Table 3). Increases were observed in flow to the small intestine (by 81 ± 19% with the highest dose, *P* < 0.05) and the adrenals (by up to 38 ± 15%, *P* < 0.05). Since the fall in arterial perfusion pressure outweighed the decrease in flow, vascular conductance was increased in all these organs (Table 3).

The blood flow to eight different muscle groups was measured, of which only flow to the M. sternocleidomastoideus increased from 5.6 ± 0.7 ml min⁻¹ 100 g⁻¹ at baseline to 12.4 ± 2.7 ml min⁻¹ (*P* < 0.05), with the highest dose. Flow to all the other muscles (M. iliopsoas, pars costalis diaphragmatis, M. pectoralis, M. erector spinae, M. gluteus, M. masseter and the M. rectus abdominis) remained constant (baseline values between 2 and 6 ml min⁻¹ 100⁻¹). Vasodilatation

occurred in all muscle groups as vascular conductance had increased 2 to 5 fold after infusion of the highest dose.

Intracoronary infusions of EMD 52692

Systemic haemodynamics No arrhythmias were observed during infusions of either the solvent or EMD 52692. Infusion of the solvent had no effect on systemic haemodynamics (Table 4). During infusion of the lowest two doses of EMD 52692 the only change was a 5% increase in heart rate. With the higher infusion rates there was a further increase in heart rate (by up to 22 ± 4%) and a 16 ± 3% decrease in mean arterial blood pressure, due to peripheral vasodilatation (decrease in systemic vascular resistance of 16 ± 4%). Cardiac output and left ventricular filling pressure did not change, while LV dP/dt_{max} showed an increase (24 ± 8%, *P* < 0.05) after infusion of the highest dose.

Table 4 Systemic haemodynamics after continuous intracoronary 10 min infusions of EMD 52692 or its solvent in anaesthetized pigs

		EMD 52692 (µg kg ⁻¹) or equal volumes of its solvent				
		0	0.10	0.29	0.66	1.41
HR	Solvent	111 ± 6	111 ± 6	111 ± 5	113 ± 5	114 ± 6
	EMD 52692	116 ± 5	117 ± 5	123 ± 6*	130 ± 8*	144 ± 10*
MAP	Solvent	87 ± 4	85 ± 3	84 ± 3	85 ± 4	84 ± 5
	EMD 52692	93 ± 4	94 ± 4	89 ± 5	86 ± 4*	79 ± 5*
CO	Solvent	2.6 ± 0.3	2.6 ± 0.2	2.6 ± 0.3	2.6 ± 0.3	2.5 ± 0.3
	EMD 52692	2.3 ± 0.1	2.4 ± 0.2	2.3 ± 0.2	2.3 ± 0.1	2.3 ± 0.1
SVR	Solvent	35 ± 3	35 ± 3	34 ± 3	35 ± 3	35 ± 3
	EMD 52692	42 ± 3	41 ± 3	39 ± 3	38 ± 2*	35 ± 1*
LV dP/dt_{max}	Solvent	2490 ± 290	2460 ± 280	2460 ± 290	2610 ± 370	2580 ± 370
	EMD 52692	2270 ± 160	2330 ± 190	2500 ± 250	2680 ± 300	2830 ± 320*
LVEDP	Solvent	7 ± 1	6 ± 1	6 ± 1	6 ± 1	6 ± 1
	EMD 52692	7 ± 1	7 ± 1	7 ± 1	7 ± 1	6 ± 1

Data are mean ± s.e.mean; *n* = 8 for the solvent-treated and *n* = 7 for the EMD 52692-treated animals. HR = heart rate (beats min⁻¹); MAP = mean arterial blood pressure (mmHg); CO = cardiac output (l min⁻¹); SVR = systemic vascular resistance (mmHg min l⁻¹); LV dP/dt_{max} = maximal rate of rise of left ventricular pressure (mmHg s⁻¹); LVEDP = left ventricular end-diastolic pressure (mmHg).

* Changes versus baseline in the EMD 52692-treated are significantly different (*P* < 0.05) from those in the solvent-treated animals.

Figure 7 Transmural (a), subendocardial (b) and subepicardial (c) blood flow after 10 min cumulative intracoronary infusions of EMD 52692 (hatched columns, total doses 0, 0.10, 0.29, 0.66 and 1.41 $\mu\text{g kg}^{-1}$, $n = 7$) or its solvent (open columns, $n = 8$) directly into the left anterior descending coronary artery (LADCA). Non-LADCA = area not perfused by the LADCA (posterior wall of the left ventricle). Data are presented as mean and the bars show s.e.mean. * Indicates that the EMD 52692-induced changes from baseline are significantly different ($P < 0.05$) from solvent-induced changes from baseline.

Myocardial performance Intracoronary infusions of the solvent had no effect on myocardial perfusion (Figure 7). A representative example of the effects of EMD 52692 is presented in Figure 8, which clearly shows the EMD 52692-induced increases in coronary blood flow. EMD 52692 dose-dependently increased transmural blood flow of the myocard-

ium perfused by the left anterior descending coronary artery by up to 128% (Figure 7). Although all layers benefitted from the increase in transmural flow, the increments in the subepicardium ($242 \pm 55 \text{ ml min}^{-1} 100 \text{ g}^{-1}$) were considerably larger than in the subendocardium ($140 \pm 46 \text{ ml min}^{-1} 100 \text{ g}^{-1}$). There were no significant changes in myocardial blood flow (Figure 7) or vascular conductance (not shown) of the posterior wall of the left ventricle (the myocardium not perfused by the left anterior descending coronary artery).

The solvent had no effect on myocardial O_2 -consumption (499 ± 39 , 486 ± 41 , 593 ± 29 , 529 ± 52 , $523 \pm 59 \mu\text{mol min}^{-1} 100 \text{ g}^{-1}$ at baseline and after 10, 20, 30 and 40 min of solvent infusion, respectively) or on regional subendo- and subepicardial segment length shortening (Table 5). Subendocardial segment length shortening decreased from $18.2 \pm 1.6\%$ to $16.8 \pm 1.0\%$ with the highest dose of EMD 52692 ($P < 0.05$). Subepicardial segment length shortening tended to increase, but the increments did not reach levels of significance (Table 5).

Myocardial O_2 -consumption of the LADCA-perfused myocardium was not affected by the drug (542 ± 43 , 522 ± 26 , 560 ± 33 , 618 ± 17 and $521 \pm 46 \mu\text{mol min}^{-1} 100 \text{ g}^{-1}$ at baseline and after 0.095, 0.29, 0.66 and 1.41 $\mu\text{g kg}^{-1}$, respectively).

Discussion

In the present study the plasma levels of EMD 52692 ranged from 5.1 to 62 $\mu\text{g l}^{-1}$. This range compares well with the effective *in vitro* concentrations described by De Peyer and colleagues (1989). In this last study the EC_{50} of EMD 52692 to hyperpolarize the membrane of the vascular smooth muscle cells of porcine coronary arteries was $0.6-1 \times 10^{-7} \text{ M}$, which corresponds with a concentration of 16–26 $\mu\text{g l}^{-1}$.

EMD 52692 is a potent systemic vasodilator. Its potency exceeds that of nicorandil, tested in the same model by a factor of 100 (Verdouw *et al.*, 1987). Furthermore EMD 52692 was shown to be 5 and 50 times more potent in anaesthetized

Figure 8 Representative tracing of the effects of intracoronary (left anterior descending coronary artery) infusions of EMD 52692 on systemic haemodynamics and regional myocardial blood flow and function in an anaesthetized pig. AP, LVP and LAP are central aortic, left ventricular and left atrial blood pressure, respectively; LVdP/dt = first derivative of LVP; ABF = ascending aortic blood flow; LADCABF = left anterior descending coronary artery blood flow; SLS(Endo) = subendocardial segment length; SLS(Epi) = subepicardial segment length. The infusion rates of EMD 52692 (total dose, $\mu\text{g kg}^{-1}$) are given at the bottom of the figure.

Table 5 Effect of continuous intracoronary 10 min infusions of EMD 52692 or its solvent on subendocardial and subepicardial segment length shortening in anaesthetized pigs

		EMD 52692 ($\mu\text{g kg}^{-1}$) or equal volumes of its solvent				
		0	0.10	0.29	0.66	1.41
Subendocardium	Solvent	17.3 $\pm$ 1.4	17.7 $\pm$ 1.5	16.8 $\pm$ 1.5	17.2 $\pm$ 1.7	17.5 $\pm$ 1.5
	EMD 52692	18.2 $\pm$ 1.6	18.3 $\pm$ 1.2	17.8 $\pm$ 1.8	17.7 $\pm$ 1.2	16.8 $\pm$ 1.0*
Subepicardium	Solvent	15.1 $\pm$ 1.1	14.9 $\pm$ 1.4	14.6 $\pm$ 1.4	15.1 $\pm$ 1.6	15.0 $\pm$ 1.3
	EMD 52692	16.7 $\pm$ 1.7	17.1 $\pm$ 1.7	17.4 $\pm$ 1.4	17.9 $\pm$ 1.4	17.6 $\pm$ 1.4

Data are mean $\pm$ s.e.mean; $n = 8$ for the solvent-treated animals and $n = 7$ for the EMD 52692-treated animals. Data are expressed as %. * Changes versus baseline in the EMD 52692-treated animals are significantly different from those in the solvent-treated animals.

dogs than cromakalim and pinacidil, respectively (Schliep *et al.*, 1989), two other compounds of which the pharmacological actions have been ascribed to potassium channel opening (Hamilton *et al.*, 1986; Bray *et al.*, 1987; Weir & Weston, 1988). The potency of EMD 52692 to dilate the systemic vascular bed, also compares favourably with that of the most potent representatives of the dihydropyridine calcium antagonists such as nisoldipine (Duncker *et al.*, 1986) and elgodipine (Sassen *et al.*, 1990).

Contraction of vascular smooth muscle cells in capacitance vessels depends on a rise in intracellular free calcium, mediated by activation of receptor-operated rather than voltage-operated events (Bolton, 1979). Webb and colleagues (1989) found that cromakalim produced dose-dependent dilatation of forearm resistance vessels, but did not dilate dorsal hand veins precontracted with noradrenaline and concluded that this potassium channel activator was arterioselective. However, we observed at similar increments in heart rate (e.g. compare the second infusion rate of the intravenous and the highest infusion rate of the intracoronary experiments), that left ventricular end-diastolic pressure decreased after the intravenous, but not after the intracoronary infusions. The dose-dependent decrease in left ventricular filling pressure during the intravenous infusions can therefore not solely be attributed to the tachycardia and EMD 52692 most likely caused some venodilatation during the intravenous infusions.

The increase in heart rate, which has also been demonstrated for cromakalim (Buckingham *et al.*, 1986), was most likely a reflex-mediated response, and is a common feature of acute administration of vasodilators. Despite the tachycardia, cardiac output did not increase. We have shown that in the same model vasodilators such as nisoldipine (Duncker *et al.*, 1986) and nicorandil (Verdouw *et al.*, 1987) elicited similar responses. In conscious pigs these compounds caused marked increases in heart rate, left ventricular $dP/dt_{\max}$ and cardiac output (Verdouw *et al.*, 1987; Duncker *et al.*, 1988), indicating that the anaesthesia may have attenuated the reflex-mediated responses.

Potassium channel activators have potentially negative inotropic properties (Yanagisawa *et al.*, 1988), although the selectivity for vascular smooth muscle outweighs that for the myocardium (Cohen & Colbert, 1986; Longman *et al.*, 1988; Gotanda *et al.*, 1988). SR 44866, a compound which is structurally identical to EMD 52692 has been shown to have inhibitory effects on the electrical and mechanical activity of cardiac muscle (Findlay *et al.*, 1989), but the required dose is much higher for cardiac muscle than for smooth muscle. We did not see any changes in $LVdP/dt_{\max}$, a frequently used measure of myocardial contractility. This index is, however, also influenced by changes in heart rate and pre- and afterload, all of which were affected considerably by EMD 52692. However, the two lowest intracoronary infusion rates (where heart rate, pre- and afterload were only slightly affected, with no changes in segment length shortening) demonstrated that at comparable intravenous infusions EMD 52692 exhibits no negative inotropic action. With the highest intracoronary dose subendocardial layers segment length shortening decreased significantly, while subepicardial segment length shortening was not affected. At present these results are difficult to inter-

pret. However, it is unlikely that these findings would have clinical implications since the dose used ($0.075 \mu\text{g kg}^{-1} \text{ min}^{-1}$, into the coronary artery) exceeds the doses that can be used in the clinical situation.

In anaesthetized animals a substantial fraction of the cardiac output is shunted through arteriovenous anastomoses (Kaiharu *et al.*, 1968; Saxena & Verdouw, 1985), which is reflected by the large entrapment of the microspheres in the lungs after induction of anaesthesia (Van Woerkens *et al.*, 1990). Figure 4 shows that although cardiac output did not change, the nutritional fraction increased with increasing doses of EMD 52692. This increase was most prominent in the brain, adrenals and small intestine, but absent in skeletal muscle. This study therefore confirms earlier investigations that potassium channel activators at equihypotensive dosages have a different vasodilator profile than for instance the calcium antagonists (Duncker *et al.*, 1986; Buckingham *et al.*, 1986; Sassen *et al.*, 1990). When comparing the vasodilator profile of EMD 52692 with two compounds that were tested in the same model, nicorandil (Verdouw *et al.*, 1987) and elgodipine (Sassen *et al.*, 1990), a calcium antagonist very similar to nisoldipine (Duncker *et al.*, 1986), it appears that EMD 52692 closely resembles nicorandil; similar increases in adrenal, cerebral, left ventricular and intestinal blood flow occurred while no effect on skeletal muscle blood flow at comparable increases in systemic vascular conductance was observed. Furthermore, it can be concluded that EMD 52692 dilates cerebral and adrenal vascular beds much more potently than elgodipine, but that the increase in vascular conductance of skeletal muscle by the potassium channel activator is less than by the calcium antagonist. Of interest is that the initial vasodilatation in the kidneys, as observed with elgodipine and EMD 52692 was followed by a decrease with the highest dose although systemic vascular conductance still increased. These vascular responses in the kidneys differed markedly in the inner and outer cortex and the medulla, as a result of which blood flow was redistributed away from the outer to the inner layers. A similar redistribution of renal flow during hypotension has been obtained by other investigators (see Carrière, 1975) and may thus be a perfusion pressure-dependent response rather than a direct drug action. The reduction in cortical glomerular blood flow might serve to enhance fluid retention to counterbalance the severe hypotension (Wright & Briggs, 1979).

Only a few studies have described the vasodilator profiles of other potassium channel activators (Arrigoni-Martelli *et al.*, 1980; Buckingham *et al.*, 1986; Cook & Hof, 1988). Cook & Hof (1988) studied the effects of cromakalim on regional blood flows in anaesthetized rabbits, and showed at doses inducing a blood pressure drop of up to 20% that renal blood flow decreased, skeletal muscle flow was maintained and blood flow to the stomach and small intestine were enhanced. These responses correspond well with our experiments. At variance with our findings, cerebral blood flow did not change significantly in their study. Arrigoni-Martelli and colleagues (1980) showed that pinacidil also did not affect blood flow through the femoral artery (representing muscle blood flow).

Myocardial O_2 -consumption did not change after both the intravenous and the intracoronary infusions. In both experi-

ments the increase in O_2 -demand caused by the increase in heart rate was fully compensated for by the decrease in arterial blood pressure. The increase in flow has therefore no metabolic origin and indicates that the coronary blood flow response is the result of a direct action of the drug on the coronary vascular bed. The finding that with the intravenous infusions the increase in flow was almost exclusively confined to the subepicardial layers is not surprising in view of the tachycardia and hypotension (Domenech & Goich, 1976); this has also been demonstrated for nicorandil (Verdouw *et al.*, 1987). Chronic treatment often leads to a reduction of the reflex tachycardia (Kiowski *et al.*, 1983). If the coronary vasodilator action were to be sustained with chronic treatment, the lower heart rate would most likely not only lower myocardial O_2 -demand but also shift coronary blood flow in favour of the subendocardial layers.

References

- ARRIGONI-MARTELLI, E., KAERGAARD NIELSEN, C., BANG OLSEN, U. & PETERSEN, J.H. (1980). N'-cyano-N-4-pyridyl-N'-1,2,2-trimethyl-propylguanidine, monohydrate (P 1134): a new, potent vasodilator. *Experientia*, **36**, 445-447.
- BAILE, E.M., NELEMS, J.M.B., SCHULZER, M. & PARÉ, P.D. (1982). Measurement of regional bronchial arterial flow and bronchovascular resistance in dogs. *J. Appl. Physiol. Respir. Environ. Exer. Physiol.*, **53**, 1004-1049.
- BOLTON, T.B. (1979). Mechanism of actions of transmitters and other substances on smooth muscle. *Physiol. Rev.*, **59**, 606-718.
- BRAY, K.M., NEWGREEN, D.T., SMALL, R.C., SOUTHERTON, J.S., TAYLOR, S.G., WEIR, S.W. & WESTON, A.H. (1987). Evidence that the mechanism of the inhibitory action of pinacidil in rat and guinea-pig smooth muscle differs from that of glyceryl trinitrate. *Br. J. Pharmacol.*, **91**, 421-429.
- BUCKINGHAM, R.E., CLAPHAM, J.C., HAMILTON, T.C., LONGMAN, S.D., NORTON, J. & POYSER, R.H. (1986). BRL 34915, a novel antihypertensive agent: comparison of effects on blood pressure and other hemodynamic parameters with those of nifedipine in animal models. *J. Cardiovasc. Pharmacol.*, **8**, 798-804.
- CARRIERE, S. (1975). Factors affecting renal cortical blood flow. A review. *Can. J. Physiol. Pharmacol.*, **53**, 1-20.
- CLAPHAM, J.C. & WILSON, C. (1987). Anti-spasmodic and spasmolytic effects of BRL 34915: a comparison with nifedipine and nicorandil. *J. Auton. Pharmacol.*, **7**, 233-242.
- COHEN, M.L. & COLBERT, W.E. (1986). Comparison of the effects of pinacidil and its metabolite, pinacidil-N-oxide, in isolated smooth and cardiac muscle. *Drug Develop. Res.*, **7**, 111-124.
- COOK, N.S. (1988). The pharmacology of potassium channels and their therapeutic potential. *Trends Pharmacol. Sci.*, **9**, 21-28.
- COOK, N.S. (1989). Effect of some potassium channel blockers on contractile responses of the rabbit aorta. *J. Cardiovasc. Pharmacol.*, **13**, 299-306.
- COOK, N.S. & HOF, R.P. (1988). Cardiovascular effects of apamin and BRL 34915 in rats and rabbits. *Br. J. Pharmacol.*, **93**, 121-131.
- DAUT, J., MAIER-RUDOLPH, W., VON BECKERATH, N., MEHRKE, G., GÜNTHER, K. & GOEDEL-MEINEN, L. (1990). Hypoxic dilation of coronary arteries is mediated by ATP-sensitive potassium channels. *Science*, **247**, 1341-1344.
- DE PEYER, J.E., LUES, I., GERICKE, R. & HÄUSLER, G. (1989). Characterization of a novel K^+ channel activator, EMD 52692, in electrophysiological and pharmacological experiments. *Pflügers Arch.*, **414** (Suppl. 1), S191.
- DOMENECH, R.J. & GOICH, J. (1976). Effect of heart rate on regional coronary blood flow. *Cardiovasc. Res.*, **10**, 224-231.
- DUNCKER, D.J., HARTOG, J.M., HUGENHOLTZ, P.G., SAXENA, P.R. & VERDOUW, P.D. (1986). The effects of nisoldipine (Bay K 5552) on cardiovascular performance and regional blood flow in pentobarbital-anaesthetized pigs with or without β -adrenoceptor blockade. *Br. J. Pharmacol.*, **88**, 9-18.
- DUNCKER, D.J., SAXENA, P.R. & VERDOUW, P.D. (1988). Systemic haemodynamics of dihydropyridine derivatives in conscious pigs with or without propranolol. *Eur. J. Pharmacol.*, **156**, 401-409.
- FINDLAY, I., DEROUBAIX, E., GUIRAUDOU, P. & CORABOEUF, E. (1989). Effects of activation of ATP-sensitive K^+ channels in mammalian ventricular myocytes. *Am. J. Physiol.*, **257**, H1551-H1559.
- FREEMAN, G.L., LE WINTER, M.M., ENGLER, R.L. & COVELL, J.W. (1985). Relationship between myocardial fiber direction and segment shortening in the midwall of the canine left ventricle. *Circ. Res.*, **56**, 31-39.
- GERICKE, R., LUES, I., DE PEYER, J.E. & HÄUSLER, G. (1989). Electrophysiological and pharmacological characterization of EMD 52692, a new potassium channel activator. *Naunyn-Schmiedeberg's Arch. Pharmacol.*, **339** (Suppl.), R 62.
- GOTANDA, K., SATOH, K. & TAIRA, N. (1988). Is the cardiovascular profile of BRL 34915 characteristic of potassium channel activators? *J. Cardiovasc. Pharmacol.*, **12**, 239-246.
- HAMILTON, T.C., WEIR, S.W. & WESTON, A.H. (1986). Comparison of the effects of BRL 34915 and verapamil on electrical and mechanical activity in rat portal vein. *Br. J. Pharmacol.*, **88**, 103-111.
- HOF, R.P. & HOF, A. (1989). Differential effects of antihypertensive drugs on nutritive and nonnutritive blood flow in anaesthetized rabbits. *J. Cardiovasc. Pharmacol.*, **13**, 565-571.
- KAIHARA, S., VAN HEERDEN, P.D., MIGITA, T. & WAGNER, H.N. (1968). Measurement of distribution of cardiac output. *J. Appl. Physiol.*, **25**, 696-700.
- KIOWSKI, W., BERTEL, O., ERNE, P., BOLLI, P., HULTHÉN, U.L., RITZ, R. & BÜHLER, F.R. (1983). Hemodynamic and reflex responses to acute and chronic antihypertensive therapy with the calcium entry blocker nifedipine. *Hypertension*, **5** (Suppl. 1), 170-174.
- LONGMAN, S.D., CLAPHAM, J.C., WILSON, C. & HAMILTON, T.C. (1988). Cromakalim, a potassium channel activator: a comparison of its cardiovascular hemodynamic profile and tissue specificity with those of pinacidil and nicorandil. *J. Cardiovasc. Pharmacol.*, **12**, 535-542.
- SASSEN, L.M.A., SOEI, L.K., KONING, M.M.G. & VERDOUW, P.D. (1990). The central and regional cardiovascular responses to intravenous and intracoronary administration of the phenylhydripyridine elgodipine in anaesthetized pigs. *Br. J. Pharmacol.*, **99**, 355-363.
- SAXENA, P.R., SCHAMHARDT, H.C., FORSYTH, R.P. & LOEVE, J. (1980). Computer programs for the radioactive microsphere technique. Determination of regional blood flows and other haemodynamic variable in different experimental circumstances. *Comp. Progr. Biomed.*, **12**, 63-84.
- SAXENA, P.R. & VERDOUW, P.D. (1985). Tissue blood flow and localization of arteriovenous anastomoses in pigs with microspheres of four different sizes. *Pflügers Arch.*, **403**, 128-135.
- SCHLIEP, H.J., BECKER, K.H., BERGMANN, R., HAASE, A.F., SCHELLING, P. & SCHULZE, E. (1989). Cardiovascular effects of EMD 52692, a novel potassium channel activator. *Naunyn-Schmiedeberg's Arch. Pharmacol.*, **339** (Suppl.), R 62.
- VAN WOERKENS, L.J., HUIGEN, R.J., DUNCKER, D.J., VAN DER GIESSEN, W.J. & VERDOUW, P.D. (1990). Redistribution of cardiac output with the emphasis on the opening of arteriovenous anastomoses by the combination of the neuroleptic azaperone and the hypnotic metomidate in pigs. *Br. J. Anaest.*, (in press).
- VERDOUW, P.D., SASSEN, L.M.A., DUNCKER, D.J., SCHMEETS, I.O.L., RENSEN, R.J. & SAXENA, P.R. (1987). Nicorandil-induced changes in the distribution of cardiac output and coronary blood flow in pigs. *Naunyn-Schmiedeberg's Arch. Pharmacol.*, **336**, 352-358.
- WEBB, D.J., BENJAMIN, N. & VALLANCE, P. (1989). The potassium channel opening drug cromakalim produced arterioselective vasodilatation in the upper limbs of healthy volunteers. *Br. J. Clin. Pharmacol.*, **27**, 757-761.
- WEIR, S.W. & WESTON, A.H. (1988). The effects of BRL 34915 and

The authors gratefully acknowledge Miss Marjo van Ee for her assistance in the preparation of this manuscript.

- nicorandil on electrical and mechanical activity and on ^{86}Rb efflux in rat blood vessels. *Br. J. Pharmacol.*, **88**, 121–128.
- WRIGHT, F.S. & BRIGGS, J.P. (1979). Feedback control of glomerular blood flow, pressure, and filtration rate. *Physiol. Rev.*, **59**, 858–1006.
- YANAGISAWA, T., HASHIMOTO, H. & TAIRA, N. (1988). The negative inotropic effect of nicorandil is independent of cyclic GMP changes: a comparison with pinacidil and cromakalim in canine atrial muscle. *Br. J. Pharmacol.*, **95**, 393–398.

(Received April 24, 1990

Revised June 21, 1990

Accepted June 25, 1990)