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Comparative study of elgodipine and nisoldipine on the contractile responses of various isolated blood vessels

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Abstract

The effects of elgodipine, a new dihydropyridine derivative, were compared to those of nisoldipine on contractile responses in various isolated artery rings and on mechanical activity in portal vein segments. Arteries used were: rabbit aorta, mesenteric (fifth branch), femoral and basilar, and sheep coronary arteries. Elgodipine and nisoldipine (10^{-16}–3×10^{-6} M) produced a concentration-dependent inhibition of the contractile responses induced by high K^+ (80 mM), 5-hydroxytryptamine (10^{-5} M) or noradrenaline (10^{-6} M or 10^{-4} M) in all the arteries studied. The inhibitory effect of elgodipine was greater in mesenteric resistance vessels ($IC_{50} = 8.0 \pm 2.1 \times 10^{-12}$ M and $2.0 \pm 0.5 \times 10^{-13}$ M for the depression of high K^+- and agonist-induced contraction, respectively), and in coronary arteries ($IC_{50} = 2.6 \pm 0.3 \times 10^{-10}$ M and $9.0 \pm 1.4 \times 10^{-8}$ M for the inhibition of high K^+-and agonist-induced contraction, respectively). In addition, the action of elgodipine in peripheral resistance vessels and in the coronary artery was more prominent than in aorta or femoral arteries, and this tissue selectivity was more apparent for elgodipine than for nisoldipine. In rat portal vein elgodipine ($IC_{50} = 6.5 \pm 0.9 \times 10^{-8}$ M) and nisoldipine ($IC_{50} = 8.5 \pm 1.3 \times 10^{-8}$ M) reduced in a concentration-dependent manner the development of mechanical activity. Furthermore, contractile responses produced by the addition of Ca^{2+} (1–5 mM) to Ca^{2+}-free high K^+ solution were also concentration dependently inhibited by elgodipine. However, elgodipine did not modify noradrenaline-induced contractions attributed to intracellular Ca^{2+} release. The results of this study indicate that elgodipine has potent vasodilator properties and vascular selectivity. The mechanisms through which elgodipine relaxes vascular smooth fibres seem to be related to its ability to inhibit the entry of extracellular Ca^{2+} into the cell.

Keywords: Elgodipine; Ca^{2+} channel antagonist; Conductance vessel; Resistance vessel; (Rabbit); (Sheep)

1. Introduction

The use of Ca^{2+} channel antagonists in both clinical and research settings is rapidly expanding, with numerous drugs with new indications being discovered and introduced. New Ca^{2+} channel antagonists appear to be useful not only in the therapy of cardiovascular disease, because of their higher vascular selectivity (Greenberg et al., 1987; Dunselman et al., 1989), but also for disease of the central nervous system (Scriabine and Janis, 1990). Many newer 1,4-dihydropyridine Ca^{2+} entry blockers are drugs which not only have selective effects relative to the heart and vascular smooth muscle, but also discriminate among different vascular beds (Ohtsuka et al., 1987).

Elgodipine (isopropyl-(2-(*N*-methyl-*N*-(4-fluorbenzyl)-amine)-ethyl-2,6-dimethyl-4-(2',3'-methylenedioxyphenyl)-1,5-dihydropyridine-3,5-dicarboxylate, monohydrochloride), IQB 875D) is a newly synthesized photoresistant Ca^{2+} channel antagonist of the phenyldihydropyridine class (which resembles oxodipine in its chemical structure). In preliminary experiments it has been found that in rabbit and rat aorta elgodipine exhibits a potent inhibition of high K^+-induced contractions and $^{45}Ca^{2+}$ influx (Tejerina et al., 1989), which indicated that it inhibited Ca^{2+} entry through voltage-operated channels. In vitro the drug proved to

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be 100-fold more potent in causing vasodilatation than in causing negative inotropic and chronotropic effects (Tejerina et al., 1989; Tamargo et al., 1991).

The present experiments were undertaken to further characterize the pharmacological properties of elgodipine in isolated rabbit and sheep vascular smooth muscle. The mode of action and the tissue specificity of elgodipine were evaluated and compared with those of nisoldipine.

2. Materials and methods

2.1. General procedure

New Zealand white rabbits of both sexes weighing 2.5–3 kg were anaesthetized with ethyl ether and killed by exsanguination from the common carotids. The thoracic aorta, femoral, mesenteric arteries (fifth branch) and the brain were rapidly removed, and the basilar artery was isolated as previously described (Whalley et al., 1983).

Thoracic aorta, femoral, and basilar arteries were placed in a modified Krebs-Henseleit solution (Krebs solution) of the following composition (mM): NaCl 118, KCl 4.7, $CaCl_2$ 1.8, $MgSO_4$ 1.2, KH_2PO_4 1.2, $NaHCO_3$ 25 and glucose 11. Adherent fat and surrounding tissue were cleaned off and the arteries were cut into rings approximately 2–3 mm wide. The rings were then suspended between two stainless steel hooks in organ baths containing 10 ml of Krebs solution. The solution was kept at 36 ± 0.5°C and gassed continuously with a 95% O_2-5% CO_2 gas mixture. The aorta, femoral, and basilar arteries were mounted under 2, 1.5, and 0.5 g tension, respectively. Each preparation was allowed to equilibrate for 90–120 min. Contractile responses were measured isometrically by means of force-displacement transducers (Grass FT O3) and were recorded on a Grass polygraph as previously described (Tejerina et al., 1988).

Mesenteric resistance vessels were mounted on a myograph, as described previously (Cauvin et al., 1984). Two 40 μm tungsten wires were passed through the lumen of an isolated cylindrical segment (2 mm long) of the fifth branch (approximately 175 μm inside diameter) of the superior mesenteric arteries. One wire was fastened with screws to a fixed tissue mounter and the other was pulled taut by parallel hooks which were attached to a string-gauge force transducer (U-gauge, Shinko Co.); the positions were adjusted with a micromanipulator. Mesenteric resistance vessels were equilibrated in physiological saline solution with the following composition (mM): NaCl 140, KCl 4.6, MgCl 1, $CaCl_2$ 1.5, glucose 10, Hepes 5. The solution was kept at 36 ± 0.5°C, gassed continuously with O_2, and maintained at an optimal resting tension of 40 mg (Cauvin et al., 1984).

Hearts from sheep of both sexes were obtained from a local abattoir and transferred to the laboratory in oxygenated Krebs solution at 4°C within 20 min. The left anterior descending and circumflex coronary arteries were isolated and placed in warm oxygenated Krebs solution, the adhering tissue was removed and vascular rings approximately 2–4 mm long were cut and mounted in a manner analogous to the aorta rings. Coronary arteries were equilibrated in Krebs solution for 90–120 min and maintained under an optimal resting tension of 1.5–2 g before specific experimental protocols were initiated. Every 10–15 min during equilibration, aerated Krebs solution was replaced and the resting tension readjusted.

After equilibration the following experiments were carried out: (1) each aorta or femoral ring was exposed to single submaximal concentrations of KCl (80 mM) and noradrenaline (10^{-6} M), basilar and coronary rings were exposed to KCl (80 mM) and 5-hydroxytryptamine (5-HT, 10^{-5} M). An initial 10–25 min control contraction was obtained in each experiment with the appropriate stimulating agent. The rings were then washed and rested for a minimum of 45–60 min. Control contractile responses for each agonist were obtained at the beginning of the experiment until two successive responses were almost identical in height. The rings were then exposed to elgodipine or nisoldipine(10^{-12}–10^{-6} M) for 20 min before the addition of KCl, noradrenaline or 5-HT and during exposure to the stimulating agents. The method used to assess the inhibitory effects of elgodipine or nisoldipine on mesenteric resistance vessels was to contract the vessels with a submaximal concentration of KCl (80 mM) or noradrenaline (10^{-4} M) for to 5–7 min, then to wash the activating agent out and repeat the stimulus after 15 min of preincubation of the vessel in physiological saline solution containing elgodipine or nisoldipine (Cauvin et al., 1987). This sequence was repeated with each single vessel for elgodipine or nisoldipine concentrations from 10^{-16} M to 10^{-6} M. Only one agonist was used in each experiment. The results were expressed as a percentage of the maximal control agonist-induced contractile response. (2) To determine whether elgodipine and nisoldipine could relax an existing contraction, aorta rings were contracted by single submaximal concentrations of noradrenaline or KCl. When the contractile response to either agonist was maximal, elgodipine or nisoldipine was added in progressively increasing cumulative concentrations (10^{-14}–10^{-6} M). The rings were allowed to reach a new steady state tension before each successive concentration of the drug was added. (3) To determine if the inhibitory effects of elgodipine were dependent on the Ca^{2+} concentration, aorta rings were incubated in Ca^{2+}-free

Krebs solution for 120 min and then in Ca^{2+}-free high K^+ (80 mM) depolarizing Krebs solution for 10 min. Cumulative concentration-response curves for Ca^{2+} were then obtained by increasing the Ca^{2+} concentration in the bath (1–5 mM) stepwise over the next 45 min. Ca^{2+} was then washed out and the rings were re-incubated in Ca^{2+}-free Krebs solution for 60 min (Tejerina et al., 1993). The high K^+ depolarizing procedure was repeated, but elgodipine or nisoldipine was added to the bath 20 min before the first addition of Ca^{2+}. The results were expressed as a percentage of the maximal contractile response induced by 5 mM $CaCl_2$. (4) In order to evaluate the possible effects of elgodipine on noradrenaline-induced Ca^{2+} release, a similar protocol to that described previously (Hester et al., 1987) was used. Briefly, elgodipine (10^{-6} M) was added 20 min prior to noradrenaline in a Ca^{2+}-free, plus EGTA (10^{-5} M) solution. In another series of experiments using the latter solution, after release was complete (approximately 5 min), the tissues were rinsed free of elgodipine and/or noradrenaline with the same Ca^{2+}-free solution for 20 min and restimulated with noradrenaline.

To study the effects of elgodipine and nisoldipine on the spontaneous phasic myogenic activity in rat portal vein, Sprague-Dawley rats (200–300 g) of both sexes were killed and the veins were removed and mounted in toto. The veins were equilibrated under 1 g tension and treated in a manner analogous to the artery rings.

2.2. Drugs

The following drugs were used: elgodipine (IQB), nisoldipine (Bayer), noradrenaline bitartrate (Sigma), potassium chloride and calcium chloride (Merck), 5-hydroxytryptamine creatine sulphate (Sigma). Stock solutions of elgodipine or nisoldipine (10^{-2} M) were prepared by dissolving elgodipine or nisoldipine powder in 99% ethanol; working solutions were made in Krebs solution or physiological saline solution, since control experiments had demonstrated that the highest ethanol level used (10^{-6} M) had no effect on vascular smooth muscle contraction. The concentrations for each chemical or drug are expressed as final concentrations in the bath in terms of salt. Ascorbic acid was added to each daily prepared solution of noradrenaline. The results are expressed throughout as means ± S.E.M. Concentration-response curves were used to determine the concentration of elgodipine or nisoldipine producing 50% inhibition of the maximal contractile response (IC_{50}), using linear regression analysis over the response range of 20–80% of the maximal inhibition. The data were analyzed using a two way analysis of variance (ANOVA), and Fisher's test. A level of probability $P < 0.05$ was accepted as statistically significant.

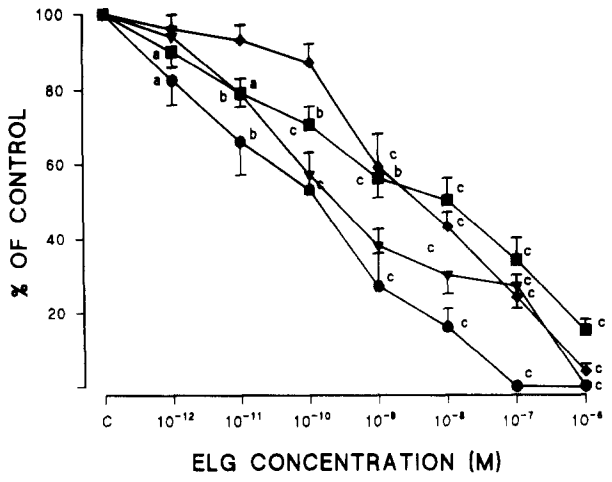

Fig. 1. Inhibitory effect of elgodipine (10^{-12}–10^{-6} M) on the contractile responses induced by high K^+ in (■) rabbit aorta, (♦) rabbit femoral, (▼) sheep coronary and (●) rabbit basilar arteries. Single concentration of K^+ (80 mM) was added in the presence of elgodipine. Each point represents the mean of eight experiments; vertical lines indicate S.E.M. Control contraction values were 4370.0 ± 200.0 mg (aorta), 2290.0 ± 62.6 mg (coronary), 1494 ± 123.0 mg (femoral), 480.2 ± 23.6 mg (basilar). [a] $P < 0.05$; [b] $P < 0.01$; [c] $P < 0.001$.

3. Results

3.1. Effect on contractions induced by KCl, noradrenaline and 5-HT

The inhibitory effects of elgodipine and nisoldipine on the contractile responses induced by high K^+ (80 mM), noradrenaline (10^{-6} or 10^{-4} M) or 5-HT (10^{-5} M) were quantified and compared in rabbit aorta, basilar, femoral, and mesenteric (fifth branch) arteries and in sheep coronary arteries. The control values when the contraction was induced by KCl (80 mM) were 4370.0 ± 200.0 mg (aorta), 2290.0 ± 62.6 mg (coronary), 1494 ± 123.0 mg (femoral), 480.2 ± 23.6 mg (basilar) and 433.7 ± 38.8 mg (mesenteric, fifth branch). When the response was induced by noradrenaline or 5-HT, the correspondig control values were 3526.0 ± 245.0 mg (aorta, noradrenaline-10^{-6} M), 2227.5 ± 42.6 mg (coronary, 5-HT-10^{-5} M), 1806.0 ± 200.0 mg (femoral, noradrenaline-10^{-6} M), 488.3 ± 25.0 mg (basilar, 5-HT-10^{-5} M) and 480.0 ± 5.8 mg (mesenteric, fifth branch, noradrenaline-10^{-4} M).

As shown in Fig. 1, Fig. 2 and Table 1, preincubation with elgodipine at concentrations between 10^{-16}–10^{-6} M produced a concentration-dependent inhibition of the peak contraction induced by the stimulating agents (K^+, noradrenaline, 5-HT) in all arteries. However, elgodipine 10^{-7} M almost suppressed the contractile response induced by KCl (80 mM) in rabbit basilar artery (Fig. 1) and mesenteric arteries (fifth branch) (Fig. 2) but left unaltered more than 30% of the contraction induced in the aorta (Fig. 1). In addi-

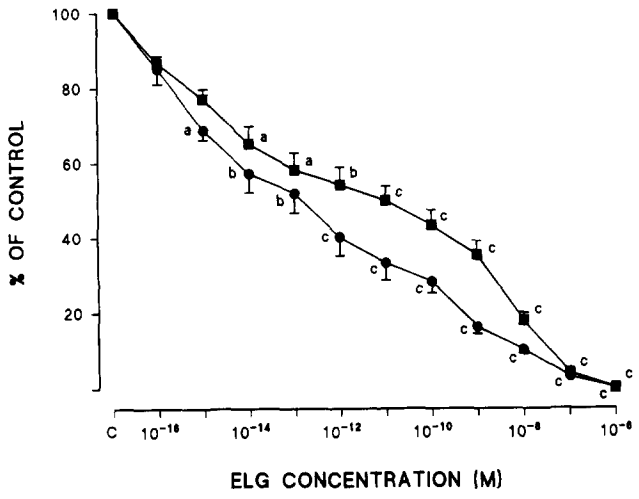

Fig. 2. Inhibitory effect of elgodipine (10^{-16}–10^{-6} M) on the contractile responses induced by (■) high K^+ (80 mM) and (●) noradrenaline (10^{-4} M) in rabbit mesenteric artery (fifth branch). Single concentrations of K^+ or noradrenaline were added in the presence of elgodipine. Each point represents the mean of six to eight experiments; vertical lines indicate S.E.M. Control contractions values were 433.7 ± 38.8 and 480.0 ± 5.8 mg when the responses were induced by high K^+ and noradrenaline, respectively. [a] $P < 0.05$; [b] $P < 0.01$; [c] $P < 0.001$.

tion, elgodipine 10^{-8} M decreased by approximately 90% the response induced by noradrenaline in mesenteric arteries (fifth branch) (Fig. 2) while this concentration left unaltered more than 50% of the contraction induced in the other arteries (data not shown). The concentrations at which elgodipine and nisoldipine inhibited 50% of the maximal contractile response (IC_{50}) and the percentage of resistant contraction (PR) in rabbit and sheep vessels are shown in Table 1.

In another group of experiments, elgodipine or nisoldipine (10^{-12}–10^{-6} M) was cumulatively added to aortic rings maximally contracted with KCl (80 mM) or noradrenaline (10^{-6} M) to test whether these drugs could relax already established contractions. Elgodipine produced a concentration-dependent relaxation of the contractions induced by high K^+ and noradrenaline, reaching significant values ($P < 0.05$) at concentrations equal or higher than 10^{-12} M and 10^{-11} M, respectively (data not shown). In these experiments steady level contractions were measured. The concentrations at which elgodipine or nisoldipine relaxed 50% of the maximal contractile response (IC_{50}) induced by high K^+ were $5.2 \pm 0.5 \times 10^{-11}$ M and $8.0 \pm 1.5 \times 10^{-13}$ M, respectively. The IC_{50} values for elgodipine- and nisoldipine-induced relaxation of noradrenaline-induced maximal response were $2.0 \pm 0.7 \times 10^{-10}$ M and $6.3 \pm 1.2 \times 10^{-8}$ M, respectively.

3.2. Effects on Ca^{2+}-induced contractions

In aortic rings previously depolarized by KCl (80 mM), elgodipine produced a concentration-dependent decrease of the contractions induced by Ca^{2+} and shifted the concentration-response curve downwards (Fig. 3). Elgodipine 10^{-9} M reduced the maximal response to 5 mM Ca^{2+} by 53.3 ± 5.1% ($n = 6$, $P < 0.05$) and at 10^{-7} M by 78.2 ± 3.3% ($n = 6$, $P < 0.01$).

Table 1
Values at which elgodipine and nisoldipine inhibited maximun contraction by 50% (IC_{50}) and the percentage of resistant contractions (PR)

	Elgodipine		Nisoldipine	
	IC_{50}	PR(%)	IC_{50}	PR(%)
Artery				
Rabbit aorta				
KCl (80 mM)	$1.0 \pm 0.4 \times 10^{-8}$	15 ± 3	$1.0 \pm 0.3 \times 10^{-9}$	16 ± 3
Noradrenaline (10^{-6}M)	$> 10^{-6}$	68 ± 4	$1.0 \pm 0.2 \times 10^{-6}$	82 ± 5
Rabbit basilar				
KCl (80 mM)	$1.0 \pm 0.4 \times 10^{-10}$	0 ± 0	$7.0 \pm 1.0 \times 10^{-13}$	0 ± 0
5-HT (10^{-5}M)	$> 10^{-6}$	59 ± 7	$1.2 \pm 0.2 \times 10^{-7}$	48 ± 6
Rabbit mesenteric (fifth branch)				
KCl (80 mM)	$8.0 \pm 2.1 \times 10^{-12}$	0 ± 0	$2.3 \pm 0.1 \times 10^{-10}$	0 ± 0
Noradrenaline (10^{-4}M)	$2.0 \pm 0.5 \times 10^{-13}$	0 ± 0	$5.0 \pm 0.2 \times 10^{-11}$	0 ± 0
Rabbit femoral				
KCl (80 mM)	$3.5 \pm 0.6 \times 10^{-9}$	0 ± 0	$1.3 \pm 1.1 \times 10^{-8}$	4 ± 1
Noradrenaline (10^{-6}M)	$> 10^{-6}$	73 ± 3	$7.5 \pm 8.2 \times 10^{-7}$	65 ± 4
Sheep coronary				
KCl (80 mM)	$2.6 \pm 0.4 \times 10^{-10}$	0 ± 0	$5.0 \pm 0.3 \times 10^{-10}$	22 ± 1
5-HT (10^{-5}M)	$9.0 \pm 1.4 \times 10^{-8}$	32 ± 8	$5.0 \pm 0.4 \times 10^{-7}$	42 ± 3
Vein				
Rat portal				
Spontaneus activity	$6.5 \pm 0.9 \times 10^{-8}$	0 ± 0	$8.5 \pm 1.3 \times 10^{-8}$	0 ± 0

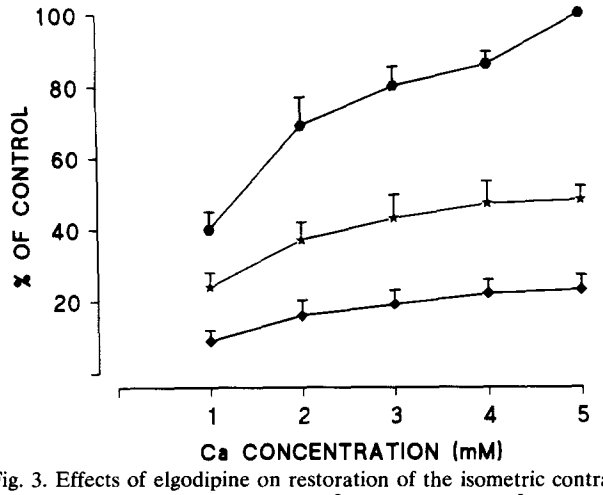

Fig. 3. Effects of elgodipine on restoration of the isometric contractions of aorta rings by addition of Ca^{2+} (1–5 mM) to Ca^{2+}-free high K^+ (80 mM) medium. Ordinate scale: percentage of the maximum control contractions obtained with the highest concentration of Ca^{2+} in each experiment. Each point represents the mean of six to eight experiments; vertical lines indicate S.E.M. (●) control, (★) after elgodipine 10^{-9} M, and (◆) after elgodipine 10^{-7} M.

3.3. Effects on myogenic activity

The effects of elgodipine and nisoldipine were studied on the myogenic activity of rat portal vein. Both drugs (10^{-11}–10^{-5} M) inhibited in a concentration-dependent manner the amplitude of the spontaneous phasic myogenic activity (data not shown). The IC_{50} values for the elgodipine and nisoldipine inhibition of the amplitude of spontaneous activity are shown in Table 1.

3.4. Effect on intracellular Ca^{2+} release

In order to determine whether elgodipine could have an inhibitory effect on noradrenaline-induced contractions attributed to Ca^{2+} release from intracellular stores, we tested the effects of the drug on phasic contractions resulting from the exposure to noradrenaline in the presence of Ca^{2+}-free plus EGTA (10^{-5} M) solution. As shown in Fig. 4 (middle panels) elgodipine (10^{-6} M) did not depress the transient contractions induced by noradrenaline (10^{-6} M) in the aorta (2.4 ± 0.2 g vs. 2.1 ± 0.3 g: control compared to elgodipine-pre-treated arteries). Furthermore, when tissues were rinsed free of elgodipine and/or noradrenaline with the Ca^{2+}-free solution and were re-exposed to the same concentration of noradrenaline, there was only a residual response both in the control aorta and in the aorta previously exposed to elgodipine (0.7 ± 0.2 g vs. 0.6 ± 0.3 g, respectively) (Fig. 4 right panels).

4. Discussion

The results of the present study indicate that elgodipine exhibits on different isolated arteries and rat portal vein segment effects similar to those previously described for nisoldipine (Kazda et al., 1980; Kazda, 1991) and other dihydropyridines (Cauvin et al., 1983; Ljung, 1985). Thus, the mechanism through which elgodipine relaxes vascular smooth muscle fibres seems to be related to its ability to inhibit the influx of extracellular Ca^{2+} through voltage-sensitive channels and receptor-operated channels of the cell membrane.

Different experimental results indicated that elgodipine inhibited Ca^{2+} entry through voltage-sensitive channels. (1) Elgodipine inhibited in a concentration-dependent manner the contractile responses induced by K^+ depolarization or by increases in extracellular Ca^{2+} in K^+-depolarized aortic rings. (2) Elgodipine was as potent as nisoldipine at inhibiting spontaneous

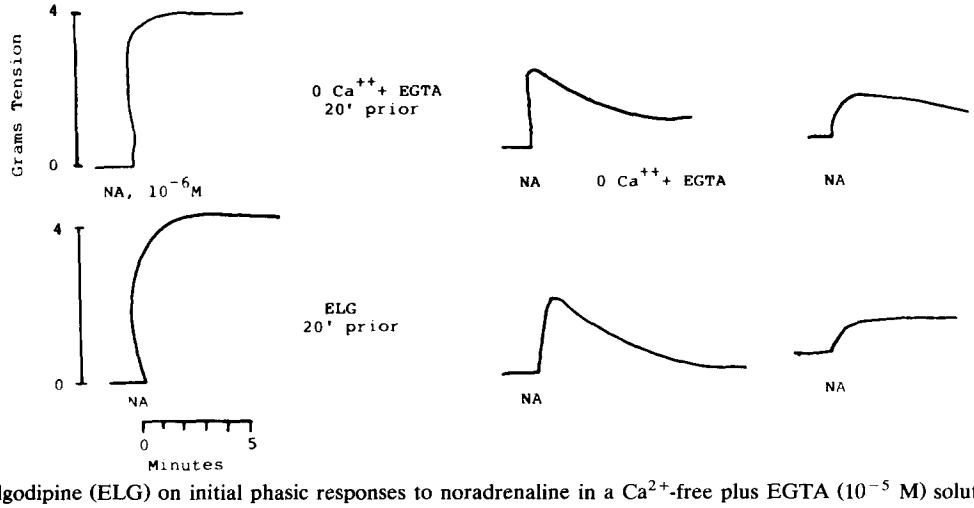

Fig. 4. Effects of elgodipine (ELG) on initial phasic responses to noradrenaline in a Ca^{2+}-free plus EGTA (10^{-5} M) solution (middle tracing, bottom) and 2nd phasic response in the same solution (right tracing, bottom) 20 min after rinsing elgodipine and noradrenaline from the tissues and bath with a similar Ca^{2+}-free plus EGTA solution. Control response to noradrenaline in Krebs solution is also included (left tracings, bottom). Top series represents control responses to noradrenaline in Krebs solution and in Ca^{2+}-free plus EGTA (initial and 2nd phasic responses) in the absence of elgodipine.

myogenic activity in rat portal veins. All these responses are entirely dependent on the entry of extracellular Ca^{2+} via voltage-sensitive Ca^{2+} channels (Sigurdsson et al., 1975; Dacquet et al., 1987), therefore, their inhibition strongly suggests that elgodipine acts by decreasing Ca^{2+} influx through these channels. Furthermore, elgodipine also exerted an inhibitory effect on total contractile responses to noradrenaline and 5-HT, indicating that it may also block Ca^{2+} entry in response to these agonists. It has been proposed that vasoconstrictor agents increase Ca^{2+} entry by opening Ca^{2+}-permeable voltage-independent channels also known as receptor-operated channels (Cauvin et al., 1983; Khalil et al., 1987). Our findings suggest, therefore, that elgodipine may also inhibit Ca^{2+} uptake through these channels. Finally, the phasic contractile response induced in the aorta by noradrenaline in Ca^{2+}-free media was insensitive to elgodipine. This response has been related to the mobilization of Ca^{2+} from intracellular stores (Hudgins and Weiss, 1968; Godfraind and Kaba, 1969). Elgodipine, at the maximum concentration tested (10^{-6} M), which inhibited by more than 30% the noradrenaline-induced contractions in the aorta, had no effect on the phasic contractions elicited in this vessel. These results indicate that at the concentration at which it inhibits Ca^{2+} entry, elgodipine fails to inhibit noradrenaline-induced intracellular Ca^{2+} release. Similar results have been found with other Ca^{2+} channel blockers (Godfraind, 1983; Tamargo and Tejerina, 1989).

During the last years the development of a number of newer Ca^{2+} channel antagonists has been promoted, with the focus largely being on variations in tissue selectivity profile. Elgodipine, in vitro, has been shown to display more prominent effects on vascular smooth muscle than on cardiac muscle (Tejerina et al., 1989; Tamargo et al., 1991). In addition, we found in the present study variations in the sensitivity to elgodipine among the different vessels analyzed. Comparing the vascular beds, we found the most prominent relaxant effects of elgodipine in peripheral resistance vessels (mesenteric artery, 5th branch) and in the coronary artery. The basilar artery was also very sensitive to elgodipine, but only when the contraction was elicited by K^+ depolarization. Moreover, the basilar selectivity of nisoldipine was more pronounced than that of elgodipine, the IC_{50} being more than one and almost three orders of magnitude lower for nisoldipine than for elgodipine (for 5-HT and K^+ responses, respectively). In contrast, for the inhibition of mesenteric resistance vessels (IC_{50}), concentrations of elgodipine around 3 times lower than those of nisoldipine were required (for the depression of both K^+ and noradrenaline responses). Finally, elgodipine and nisoldipine appeared to be almost equipotent in depressing the contractile responses induced in the sheep coronary artery. However, when compared to the rabbit aorta, the effects of elgodipine on the sheep coronary artery were around 10–40 times greater, for agonist and K^+ responses, respectively. Meanwhile, with nisoldipine, the tissue selectivity for sheep coronary artery was only 2 times greater than for the rabbit aorta (when tested on both agonist and K^+ responses). In other words, while both dihydropyridines had higher potency in inhibiting contractions of the rabbit mesenteric and basilar arteries and of the sheep coronary artery than of the rabbit aorta, selectivity of the coronary and the mesenteric vessels was more pronounced with elgodipine while selectivity for the basilar artery was more prominent with nisoldipine. Our findings are in agreement with those previously published in which elgodipine demonstrated potent systemic and coronary dilator properties in anaesthetized and conscious animals (Sassen et al., 1990; Van Woerkens et al., 1991; Drieu la Rochelle et al., 1994).

The precise explanation for the tissue specificity is not yet known, but it may be due to differences in the structure of the channels themselves from vessel to vessel (Quins et al., 1981, Cauvin et al., 1988) and/or to variations in the membrane potentials of the different arteries (Shibata et al., 1991; Tejerina et al., 1992). However, in the case of agonist-induced responses, the different efficacy of the drugs appears to be more related to the source of the Ca^{2+} activator of the contractile process.

Rabbit mesenteric resistance vessels have been shown, in our study and in others, to be extremely sensitive to the inhibitory effects of organic Ca^{2+} blockers (Cauvin et al., 1988; Tejerina et al., 1992). It has been reported that, in these vessels, contractions resulting from receptor activation by noradrenaline are more sensitive to Ca^{2+} antagonistic inhibition than are contractions induced by K^+ depolarization (Cauvin et al., 1984, 1988). In the present study, the IC_{50} value for elgodipine inhibition of noradrenaline-induced contraction was lower (about one order of magnitude) than the IC_{50} value for the inhibition of high K^+-induced contraction. A possible explanation for the greater sensitivity of the noradrenaline-induced contraction in response to elgodipine in resistance vessels is that noradrenaline modulation of voltage-sensitive Ca^{2+} channels may be particularly important in promoting spike activity and Ca^{2+} influx in small resistance vessels where Ca^{2+} stores are relatively poorly developed (Benham and Tsien, 1988). In contrast, in the other arteries (rabbit aorta, femoral, basilar arteries and sheep coronary artery) the inhibitory action of elgodipine and nisoldipine on the K^+-induced contractions was greater than the action on agonist-induced responses. However, the coronary artery was also quite sensitive to the inhibition exerted by elgodipine when the contraction resulted from receptor activation (5-

HT). This higher sensitivity of the coronary artery as compared to the other arteries could be explained, at least partially, by the fact that this vessel appears to be also largely dependent on an extracellular Ca^{2+} source when activated with an agonist (Van Breemen and Siegel, 1980) whereas in the other vessels, Ca^{2+} is supplied to the myofilament from both extra-and intracellular Ca^{2+} sources (Van Breemen and Siegel, 1980; Allen and Banghart, 1979). Thus, the lower degree of sensitivity to elgodipine inhibition of agonist-induced responses, particularly in aorta and femoral arteries, could be due to the contribution of intracellular Ca^{2+} release, a process which was found to be insensitive to the drug (see Fig. 4). It has been proposed that contractility may be regulated not only by the intracellular Ca^{2+} concentration but also by the Ca^{2+} sensitivity of the contractile elements and that the contraction induced by α-adrenoceptor stimulation is the result of an increase in the Ca^{2+} influx and Ca^{2+} release as well as of Ca^{2+} sensitization (Karaki, 1989; Nishimura et al., 1990). Therefore, the lower sensitivity to elgodipine of the agonist-evoked contractions (compared to the high-K^+ responses) could be due to resistance to elgodipine inhibition of not only the agonist-induced Ca^{2+} release but also the agonist-induced Ca^{2+} sensitization. A relatively poor sensitivity to Ca^{2+} blocker depression of agonist-evoked contractions in aorta (Tejerina et al., 1992; Shibata et al., 1991), femoral (Allen and Banghart, 1979; Kazda, 1991) and basilar arteries (Takagi et al., 1983) has been previously reported by different laboratories.

Overall, the results of the present investigation show that elgodipine, a new dihydropyridine derivative, has potent vasodilator properties which appear to be related to its ability to inhibit the entry of extracellular Ca^{2+} into vascular smooth muscle. The inhibitory effects of elgodipine in peripheral resistance vessels and in the coronary artery are more prominent than those in aorta or femoral artery, and this tissue selectivity is more apparent for elgodipine than for nisoldipine.

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