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Antagonistic effects of dihydropyridines and verapamil on $CaCl_2$ and 5–HT–evoked contraction in porcine coronary artery[1]

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ABSTRACT In porcine coronary arterial strips, the antagonistic effects of nifedipine (Nif), nimodipine (Nim), nicardipine (Nic), felodipine (Fel), and verapamil (Ver) to $CaCl_2$–evoked contraction in Ca^{2+}–free, K^+–depolarized solution were greater than that to 5–HT. The order of potency (pD_2') was Nif (9.1) > Fel (8.4) > Nim (7.9) > Nic (7.8) > Ver (7.2) to $CaCl_2$ and Nif (8.3) > Nim (7.5) > Fel (6.8) > Ver (5.6) > Nic (5.3) to 5–HT. Ver inhibited 2 components of 5–HT–evoked contraction in Ca^{2+}–free solution, but Fel inhibited only extracellular Ca^{2+}–dependent contraction, suggesting that their action modes are different.

KEY WORDS nifedipine; nimodipine; nicardipine; felodipine; verapamil; potassium; calcium; coronary vessels

1,4–Dihydropyridines (DHP) are calcium channel blockers which dilate blood vessels and have been widely used to treat hypertention, angina, and cerebral diseases[1]. Their pharmacological actions on vascular smooth muscle were studied mostly in rabbits, rats, and dogs[2–4]. Systematic study of their actions on porcine coronary arteries has not been found in literature. The present study was undertaken to compare the effects of nifedipine (Nif), nimodipine (Nim), nicardipine (Nic), felodipine (Fel) with that of verapamil (Ver) as a representative of phenylalkylamines on 5–hydroxytryptamine (5–HT)– and $CaCl_2$–evoked contractions of porcine coronary arteries and to compare the inhibitory actions of Fel and Ver on 2 components of 5–HT–evoked contraction of porcine coronary artery to explor any differences in their modes of action.

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MATERIALS AND METHODS

Porcine hearts were dissected out immediately after slaughtering, immersed in 4℃ Krebs' solution bubbled with 95% O_2 + 5% CO_2 and the main branch of the anterior descending coronary artery was isolated then. The connective tissue was cleared and the artery was spirally cut into strips about 10 mm × 2 mm. The whole process was completed within 40 min. The strips were mounted in organ baths containing Krebs' solution 5 ml under a resting tension of 3 g. The solution was bubbled with 95% O_2 + 5% CO_2 (pH 7.2–7.4 at 37℃). Isometric tension measured with a force transducer was displayed on a XMT–200 recorder. The strips were allowed to equilibrate for 2 h before the experiment. The volume of drugs added into the organ baths each time was $\leqslant$ 0.1 ml.

Nif, Nim, Nic, Fel (Hebei Medical college) and Ver hydrochloride injection (Changzhou Pharmaceutical Factory) and 5–HT (Fluka, Switzerland). All DHP were dissolved in acetone and stored at 4℃. They were diluted in redistilled water when used. The final acetone concentration was $\leqslant$ 0.2%, which was proved in preliminaty test to show no effect on the strips.

In Ca^{2+}–free Krebs' solution, $CaCl_2$ was omitted from normal Krebs' solution and EGTA 0.1 mmol · L^{-1} was added. High K^+–depolarized solution was prepared from Ca^{2+}–free solution with KCl 80 mmol · L^{-1}.

RESULTS

Action of Nif, Nim, Nic, Fel, and Ver on contraction evoked by 5–HT After the control cumulative concentration response of 5–HT was made, 5–HT was washed out and the strips were restored to their original state, then the strips were inclubated for 20 min in solutions containing different concentrations of Nif, Nim, Nic, Fel, and Ver. Each strip was used only once for one drug. All drugs produced a concentration–related inhibition on the 5–HT–evoked contraction (Fig 1), their pD_2' values were 8.3 (Nif), 7.5 (Nim), 6.8 (Fel), 5.6 (Ver), and 5.3 (Nic).

Action of Nif, Nim, Nic, Fel, and Ver on contraction evoked by $CaCl_2$ The control cumulative concentration response of $CaCl_2$ was made in high K^+–depolarized Ca^{2+}–free solution and the actions were tested in the same manner as described above. All drugs produced concentration–related inhibition of the $CaCl_2$–evoked contraction (Fig 1), their pD_2' values were 9.1 (Nif), 8.4 (Fel), 7.9 (Nim), 7.8 (Nic), and 7.2 (Ver).

Action of Ver and Fel on the components of 5–HT–evoked contraction After equilibration for 2 h in Krebs' solution, the strips were washed with Ca^{2+}–free Krebs' solution for 3 times, then incubated for 30 min in this solution before 5–HT 30 mmol · L^{-1} was added. When maximal contraction appeared, $CaCl_2$ 2.5 mmol · L^{-1} was restored. The experiment was repeated every 20 min. 5–HT 30 mmol · L^{-1} initiated a fast, relatively small and nonsustained contraction, and a further slow and sustained one was seen after restoration of $CaCl_2$. When the incubation times in Ca^{2+}–free Krebs' solution was lenghthened, the 5–HT–induced fast phase decreased and disappeared eventually; whereas $CaCl_2$–induced slow phase increased in such a way that the same maximal tension reached irrespective of the incubation time in Ca^{2+}–free solution

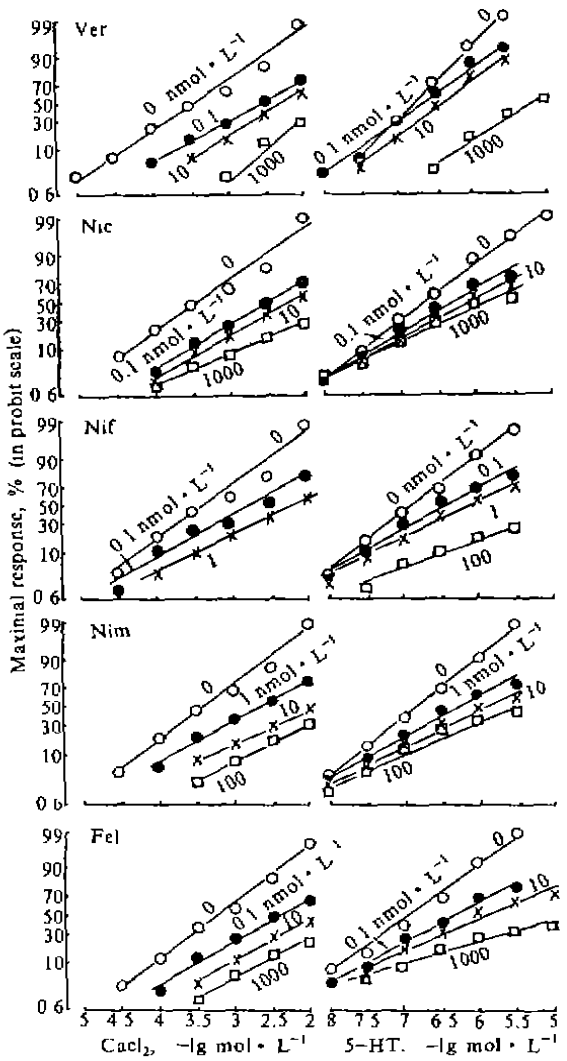

Fig 1. Effects of Ver, Nic, Nif, Nim, Fel on $CaCl_2$– and 5–HT–induced contraction of porcine coronary arterial strips. (○) **Control.** n=5–10 **pigs,** $\bar{x}\pm SD$.

(Fig 2). One strip was used as control, whereas the others from the same coronary artery were used as testing strips which were incubated for 20 min in Ca^{2+}–free Krebs' solution containing either Ver 1 μmol · L^{-1}or Fel 1 μmol · L^{-1}, then 5–HT and $CaCl_2$ were added successively as described above. Ver inhibited significantly the 2 components of

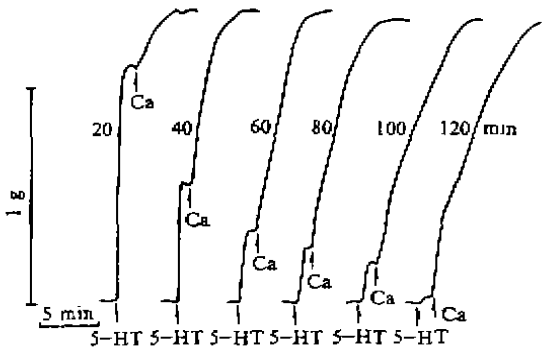

Fig 2. Isometric recordings of contractile response to 5-HT of porcine coronary arterial strip incubated in different periods in Ca^{2+}-free solution; ↑ calcium 2.5 mmol · L^{-1} was restored.

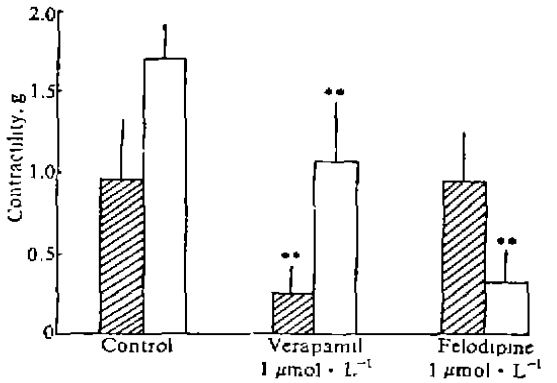

Fig 3. Effects of Ver and Fel on the 2 components of 5-HT-evoked contraction of porcine coronary arterial strips in Ca^{2+}-free solution. (▨) Initial fast phase; (□) Ca^{2+}-dependent late slow phase. $n=6$ pigs, $\bar{x} \pm$ SD. $^{**}P<0.05$ vs control

contraction induced by 5-HT while Fel only inhibited the component of extracellular Ca^{2+} influx (Fig 3).

DISCUSSION

The results showed that Nif was the most potent antagonist on the coronary artery. The potencies we found for Nif and Ver were in good agreement with those reported by Johnson, *et al*[5,6]. But the order of inhibitory potencies for Nif, Fel, and Ver was different from that reported by them. Our results were also different from that of Nyborg, *et al*[7] that the antagonistic effect of Nim in the coronary artery of rats was more potent than Nif, suggesting there may be species differences in actions of DHP. The results that 5-HT initiated a fast small and nonsustained contraction of the coronary arterial strips in Ca^{2+}-free Krebs' solution and that as the incubation time prolonged, the amplitude of this fast contraction decreased and disappeared eventually suggested that 5-HT might evoke contraction by activating 5-HT_2 receptors on membrane[8] resulting in the release of Ca^{2+} from intracellular storage sites[6]. Since the intracellular Ca^{2+} storage was limited, it might be exhausted if not restored. In this experiment, Ver inhibited the 2 components of 5-HT-induced contraction, while Fel inhibited only the contraction evoked by extracellular Ca^{2+} influx. This result was similar to our previous report that Ver inhibited the 2 components of norepinephrine-induced contraction of rat aorta, whereas Nim inhibited only the component caused by extracellular Ca^{2+} influx[9]. This strongly suggests that the site of action of Ver as a representative of phenylalkylamine is different from that of DHP which act mainly on the transmembrane Ca^{2+} influx.

REFERENCES

1 Schwartz A, Matlib MA, Balwierczak J, Lathrop DA. Pharmacology of calcium antagonists. *Am J Cardiol* 1985; **55** : 3C

2 Towart R. The selective inhibition of serotonin-induced contractions of rabbit cerebral vascular smooth muscle by calcium antagonistic dihydropyridines: an investigation of the mechanism of action of nimodipine. *Circ Res* 1981; **48** : 650

3 Ljung B, Kjellstedt A. Functional antagonism of noradrenaling responses by felodipine and other calcium antagonists in vascular smooth muscles. *J Cardiovasc Pharmacol* 1987; **10** (Suppl 1) : S82

4 Nakayama K, Kashiwabara T, Yamada S, Tahaka Y. Assessment in pig coronary artery of

long-lasting and potent calcium antagonistic actions of the novel dihydropyridine derivative meprirodipine hydrochloride. *Arzneimittel-forschung* 1989; **39** : 50

5 *Johnson JD, Fugman DA.* Calcium and calmodulin antagonists binding to calmodulin and relaxation of coronary segments. *J Pharmacol Exp Ther* 1983; **226** : 330

6 Johnson JD, Andrews CT, Khabbaza EJ, Mills JS. The interaction of felodipine with calcium-binding proteins. *J Cardiovasc pharmacol* 1987; **10** (Suppl 1) : S53

7 Nyborg NCB, Mikkelsen EO. Comparison of the inhibitory effects of nifedipine and nimodipine on mechanical responses of isolated rat coronary small arteries. *J Cardiovasc Pharmacol* 1987; **9** : 519

8 Angus JA. 5-HT receptors in the coronary circulation. *Trends Pharmacol Sci* 1989; **10** : 89

9 Dong H, Yang ZC, Wei X. Effect of nimodipine on the contraction and the Ca influx of rat aorta. *Acta Pharmacol Sin* 1990; **11** : 304

二氢吡啶类和维拉帕米对氯化钙及 5-羟色胺所致猪冠状动脉收缩的拮抗作用

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提要 Nif, Nim, Nic, Fel 和 Ver 对无 Ca^{2+}高 K^+ 去极化时 $CaCl_2$ 所致离体猪冠状动脉收缩的拮抗强度依次为：Nif > Fel > Nim > Nic > Ver，pD_2'分别为 9.1, 8.4, 7.9, 7.8, 7.2；对 5-HT 所致收缩的拮抗强度依次为：Nif > Nim > Fel > Ver > Nic，pD_2'分别为 8.3, 7.5, 6.8, 5.6, 5.3，以 Nif 最强. Ver 同时抑制 5-HT 所致的两种收缩成分. Fel 只抑制外 Ca^{2+}内流所致的收缩，提示两类钙拮抗剂的作用不同.

关键词 硝苯啶；尼莫地平；尼卡地平；非洛地平；维拉帕米；钾；钙；冠状血管

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Effects of glibenclamide and tolbutamide on ischemia- and ouabain- induced arrhythmias and membrane potentials of ventricular myocardium from rat and guinea pig

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ABSTRACT Glibenclamide (Gli) 0.3, 1, 3 mg · kg^{-1} and tolbutamide (Tol) 3, 10, 30 mg · kg^{-1} iv 10 min before ischemia or ouabain infusion prevented ventricular fibrillation induced by ischemia in rat and arrhythmias induced by ouabain in guinea pig. Gli 10 μmol · L^{-1} and Tol 1 mmol · L^{-1} increased APD and ERP in rat ventricular muscle. Gli 0.1, 1, 10 μmol · L^{-1} and Tol 0.01, 0.1, 1 mmol · L^{-1} prevented and reversed the shortening of APD and ERP induced by hypoxia in guinea pig ventricular muscle. These effects of Gli and Tol were dose-dependent. The results confirmed that Gli and Tol were effective on arrhythmias induced by ischemia and ouabain by blocking ATP-sensitive potassium channel.

KEY WORDS glibenclamide; tolbutamide; ischemia; ouabain; arrhythmia; membrane potentials; anoxia

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ATP-dependent potassium channels had been demonstrated in cardiac myocytes[1,2]. The channels opened when ATP concentration in myocardial cell was below normal, which may be responsible for K^+ loss and shortening of action potentials and consequently for