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Antagonistic effects of dipfluzine, flunarizine, and cinnarizine on 5-hydroxytryptamine-evoked contraction in pig basilar artery

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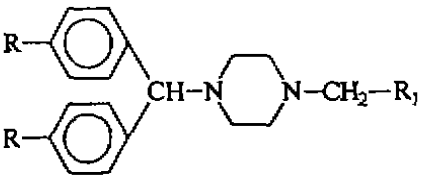
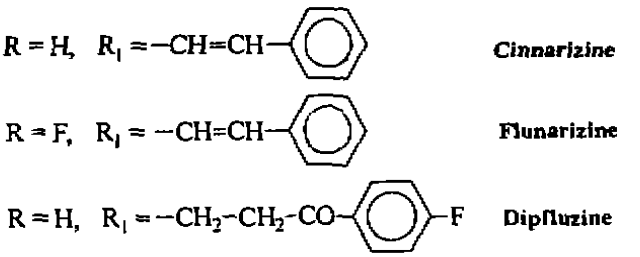
KEY WORDS dipfluzine; flunarizine; cinnarizine; basilar artery; serotonin; vasoconstriction

AIM: *To investigate the effects of dipfluzine* (Dip), a new derivative of cinnarizine (Cin), first developed by China, upon 5-hydroxytryptamine (5-HT)-induced contractions in cerebral arteries. **METHODS**: Compared Dip, flunarizine (Flu), and Cin antagonistic effects and actions on 2-component contractions evoked by 5-HT in isolated pig basilar artery. **RESULTS**: Dip showed a greater concentration-dependent antagonistic effect on 5-HT-evoked contraction than Cin and Flu in pig basilar artery rings. The order of potency (IC_{50}) was Dip (4.0 μmol·L^{-1}) > Flu (15.6 μmol·L^{-1}) > Cin (25.2 μmol·L^{-1}). All the Dip, Flu, and Cin inhibited 2-components of 5-HT-induced contraction. The antagonistic effects of Dip and Cin on the initial fast-phase contraction (FPC) were greater than that on the sustained tonic-phase contraction (STC), but Flu showed no difference between inhibiting effects on 2-component contractions. **CONCLUSION**: Dip was more potent than both of Flu and Cin on cerebrovascular dilation, associated mainly with the inhibition of intracellular calcium release.

5-Hydroxytryptamine (5-HT) released from aggregating platelets is responsible for the genesis of ischemic vasospastic disorders[1]. Calcium antagonists inhibited the cerebral artery contraction evoked by 5-HT[2,3], and reduced neurological damage after subarachnoidal hemorrhage[4]. Dipfluzine (Dip), a novel diphenylpiperazine calcium antagonist first developed by Department of Chemistry, Beijing University, showed a more selective and more potent antagonistic effect than cinnarizine (Cin) on contractions evoked by levaterenol and KCl in isolated cerebral artery[5] and protective effects against ischemic brain edema[6]. In this investigation, we compared the antagonistic effects of Dip, flunarizine (Flu), and Cin on the contraction evoked by 5-HT in pig basilar artery.





MATERIALS AND METHODS

Drugs 5-HT (Fluka) was dissolved in distilled water. Dip (Chemical Department, Beijing University), Flu (Henan Xichuan Pharmaceutical Factory), and Cin (Xi-an Jassen Pharmaceutical Co Ltd) as the stock solution 100 mmol·L^{-1} were made with tartaric acid solution 10 mmol·L^{-1} (solvent), and further diluted with the solvent daily before use. The same amount of the solvent was used as control.

Isolated pig basilar artery rings After slaughter, the pig basilar arteries were immediately separated and cut into 4 - 6 mm rings. The rings were suspended with two wire holders in a 20-mL bath containing K - H solution at 37 ℃ gassed with 95 % O_2 + 5 % CO_2, pH was 7.2 - 7.4. Tension of the rings were measured isometrically with LW-10 force-displacement transducers (Instrument Factory of Shanghai) and recorded on XWT-204 model potentiometric recorders (Shanghai Dahua Appliance Factory). The resting tension was adjusted to 1 g. After a 90-min equilibration, *stimulations of artery with 5-HT (final concentration 50 μmol* ·L^{-1}) was repeated every 20 min in the presence of solvent until a reproducible contractile response (within 10 % deviation) was obtained, which was used as a control standard

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(0 % dilation). Then the ring was incubated in K − H solutions containing a certain concentration of Dip, Flu, or Cin for 15 min.

In "calcium withdrawal" experiments, the ring was incubated in Ca^{2+}-free solution for 15 min, before 5-HT 50 $\mu mol \cdot L^{-1}$ was added. When maximal contraction induced by 5-HT appeared, $CaCl_2$ 2.5 $mmol \cdot L^{-1}$ was restored. After incubation in Ca^{2+}-free solution containning Dip 0, 5, or 15 $\mu mol \cdot L^{-1}$, Flu or Cin 0 or 15 $\mu mol \cdot L^{-1}$ for 15 min, the procedure was repeated. One ring was only used once for solvent and one concentration of drug.

Statistical analysis Comparison of the results were made with t-test.

Fifty percent inhibitory concentrations (IC_{50}) were calculated with Bliss method.

RESULTS

All the Dip, Flu, and Cin induced a concentration-related inhibition on 5-HT-evoked contraction (r = 0.9911, 0.9865, and 0.9955, respectively). The IC_{50} (95 % confidence limits) values of Dip, Flu, and Cin were 4.0 (2.3 − 7.0), 15.7 (9.5 − 25.7), and 25.2 (17.0 − 37.4) $\mu mol \cdot L^{-1}$, respectively (Fig 1).

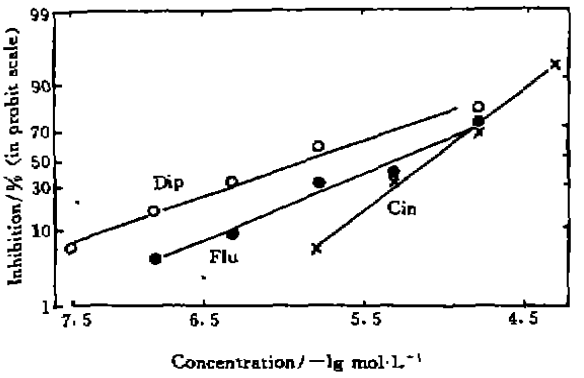

Fig 1. Effects of Dip, Flu, and Cin on 5-HT-induced contraction of pig basilar arterial rings.

5-HT initiated a fast-phase contraction (FPC) in Ca^{2+}-free K − H solution and a further slow sustained tonic contraction (STC) after restoration of $CaCl_2$. Three drugs all inhibited 2 components of contraction and the actions of Dip were more potent than those of Flu and Cin. The inhibitory percentages of Dip for FPC were higher than those for STC, which were qualitatively similar to Cin. Flu showed no significant difference between inhibitory percentages on FPC and STC. Flu effect on FPC was similar to Cin, but Flu effect on STC was greater than Cin (Tab 1).

Tab 1. Effects of Dip, Flu, and Cin on 2 components of 5-HT-evoked contraction. $n = 6$, $\bar{x} \pm s$. $^aP > 0.05$, $^bP < 0.05$, $^cP < 0.01$ *vs* FPC; $^dP > 0.05$, $^eP < 0.05$, $^fP < 0.01$ *vs* Cin; $^gP > 0.05$, $^hP < 0.01$ *vs* Flu; $^kP < 0.05$, $^lP < 0.01$ *vs* 0.

Drugs/ $\mu mol \cdot L^{-1}$		FPC contraction/ g	FPC inhibition/ %	STC contraction/ g	STC inhibition/ %
Cin	0	0.97 ± 0.21		1.29 ± 0.19^b	
	15	0.62 ± 0.25^k	37 ± 8	1.07 ± 0.06^{bk}	21 ± 5^c
Flu	0	1.07 ± 0.17^d		1.40 ± 0.29^{bd}	
	15	0.63 ± 0.09^{dl}	37 ± 9^d	0.87 ± 0.19^{bel}	40 ± 8^{af}
Dip	0	0.96 ± 0.12^{dg}		1.27 ± 0.27^{bdg}	
	5	0.39 ± 0.08^{dgl}	59 ± 10^{fi}	0.71 ± 0.23^{dgl}	40 ± 8^{cfg}
	15	0.00 ± 0.00^{fil}	100 ± 0^{dfi}	0.13 ± 0.03^{dil}	90 ± 8^{cfi}

DISCUSSION

Our results demonstrated that 5-HT-induced contractions in isolated pig basilar artery were depressed by Dip, Flu, or Cin and the relaxing effect of Dip on 5-HT-evoked contraction is more potent than those of Flu and Cin. Considering together with Dip has been known selective to improve cerebral blood flow in ischemic brain, it is suggested that its antagonistic activity on 5-HT-induced contraction may be an important factor in protection against ischemic brain edema.

The contraction evoked by 5-HT in pig basilar artery consists of 2 components, FPC and STC[2], involved in the intracellular release and extracellular influx of Ca^{2+}, respectively. Three drugs could inhibit both components of contraction and the effects of Dip were more potent than those of Cin and Flu. But Dip, like Cin, showed the antagonistic effect on FPC was more marked than that on STC, which indicated the vasodilator effects of Dip and Cin is different from that of Flu, the former may mainly influence intracelluar release of Ca^{2+}.

It was concluded that Dip was more potent than both of Flu and Cin on cerebrovascular dilation, associated mainly with the inhibition of intracellular calcium release.

REFERENCES

1 Vanhoutte PM. 5-Hydroxytryptamine and vascular disease. Fed Proc 1983; 42: 233 - 7.

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 nimodipine. Circ Res 1981; 48: 650 - 7.

3 Li BY, Zhang YC, Li WH. Effects of berbamine on contraction and Ca^{2+} influx of pig basilar artery. Acta Pharmacol Sin 1992; 13: 412 - 6.

4 Allen GS. Role of calcium antagonists in cerebral arterial spasm. Am J Cardiol 1985; 55: 149B - 153B.

5 Wang YL, Li YS, Fu SX, Jin S. Acute toxicity of dipfluzine and its effects on isolated vascular smooth muscle. Acta Pharmacol Sin 1990; 11: 39 - 42.

6 Wang YL, He RR. Protective effect of dipfluzine on experimental brain edema in rats. Acta Pharmacol Sin 1994; 15: 201 - 5

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双苯氟嗪、氟桂利嗪、桂利嗪对5-羟色胺所致离体猪基底动脉收缩的拮抗作用

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关键词 双苯氟嗪；氟桂利嗪；桂利嗪；基底动脉；血清素；血管收缩

A目的：探索双苯氟嗪（Dip，一种我国自行合成的桂利嗪衍生物），对5-羟色胺所致的脑动脉收缩的影响. 方法：比较双苯氟嗪、氟桂利嗪（Flu）、桂利嗪（Cin）对5-羟色胺所致离体猪基底动脉收缩的抑制及两种收缩成分的影响. 结果：三者的拮抗作用强度顺序（IC_{50}）为 Dip 4.0 μmol·L^{-1} > Flu 15.6 μmol·L^{-1} > Cin 25.2 μmo·L^{-1}. 这三种药对5-羟色胺所致离体猪基底动脉的两种收缩成分均有拮抗. Dip 和 Cin 抑制收缩的快速相强于持续相，而 Flu 对二者的作用无显著差异. 结论：在 Dip，Flu 和 Cin 三种药之中，Dip 对脑血管的扩张作用最强，其原因主要与抑制内钙的释放有关.

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Effects of *m*-nisoldipine on aortic calcium accumulation in rats with vascular calcium overload[1]

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KEY WORDS *m*-nisoldipine; verapamil; cholecalciferol; calcium; arteries; electron probe microanalysis

AIM: To study the effects of a novel calcium channel blocker, *m*-nisoldipine, on vascular calcium overload (VCO) at both tissue and cellular levels. **METHODS**: VCO was induced in Wistar rats by treatment with colecalciferol (Col, 400 000 IU·kg^{-1}, *po*) and an aqueous mixture of ethanol and polyethyleneglycol-400 for 3 d. The tissue and subcellular calcium contents of aorta were determined by atomic absorption spectrometer and electron probe microanalysis, respectively. **RESULTS**: Chronic treatment with *m*-nisoldipine (*m*-Nis, 1 - 15 mg·kg^{-1}, *po*, bid) only had mild inhibition on the elevation of total calcium in aorta, and the dose-response relationship of *m*-Nis displayed a bell shape, with inhibition ratio of 24 % only for *m*-Nis 2.5 mg·kg^{-1}. The effect of verapamil (12.5 mg·kg^{-1}, *po*, bid) was a little better than that of *m*-Nis. The intracellular VCO in medial smooth muscle cells of aorta were remarkably inhibited by *m*-Nis (2.5 mg·kg^{-1}), with inhibition ratios of 72 % for cytoplasm and 76 % for mitochondrion. The calcium accumulation in nucleus was reduced to a lesser degree than

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