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INFLUENCE OF DOPAMINE AND NORADRENALINE ON ISOLATED CEREBRAL ARTERIES OF THE DOG

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1 Effects of dopamine and noradrenaline were compared in helically-cut strips of canine cerebral arteries.

2 Dopamine caused a greater maximal contraction than noradrenaline, although the ED_{50} for noradrenaline was appreciably less. The contraction induced by these amines was reversed to a relaxation by treatment with phenoxybenzamine.

3 Relaxation induced by dopamine in phenoxybenzamine-treated and prostaglandin-contracted cerebral arteries was not influenced by 1 μM propranolol, while relaxation induced by noradrenaline at low concentrations (2 μM and 10 μM) was significantly attenuated. Neither aminophylline nor atropine affected the relaxant effect of dopamine.

4 A mechanism other than β-adrenergic, cholinergic or adenosine-related appears to be involved in the relaxation elicited by dopamine in cerebral arterial strips.

Introduction

Specific dopamine receptors have been postulated to exist in renal, mesenteric and coronary vessels from the following findings; in anaesthetized dogs, dopamine causes vasodilatation which is not blocked by β-adrenoceptor blocking agents but by haloperidol, bulbocapnine, phenothiazines and apomorphine (reviewed by Goldberg, 1972) and this specific action of dopamine is observed only in renal (McDonald, Goldberg, McNay & Tuttle, 1963; McNay, McDonald & Goldberg, 1965), mesenteric (Eble, 1964) and coronary (Schuelke, Mark, Schmid & Eckstein, 1971) but not in femoral and carotid vasculatures. Recent studies in our laboratory have demonstrated that dopamine causes a dose-related relaxation in canine isolated renal, mesenteric and coronary arteries treated with phenoxybenzamine and this relaxation is not influenced by propranolol (Toda & Goldberg, 1973; 1975; Goldberg & Toda, 1975).

It has been reported that continuous infusion of dopamine via the femoral vein increases the cerebral blood flow of anaesthetized dogs after treatment with α-adrenoceptor blocking agents; the increase is not inhibited by propranolol but by pimozide (von Essen, 1974), a dopamine antagonist (Andén, Butcher, Corrodi, Fuxe & Ungerstedt, 1970). However, when such experiments are done *in situ*, the possible involvement of regulatory factors in cerebral circulation and the interaction of these factors with actions of dopamine cannot be completely excluded.

The present study was therefore undertaken to elucidate the direct action of dopamine on cerebral arteries isolated from dogs, and to make a comparison with the action of noradrenaline.

Methods

Mongrel dogs of either sex, weighing 7 to 15 kg, were anaesthetized with intraperitoneal injections of sodium pentobarbitone (50 mg/kg) and killed by bleeding from the common carotid arteries. The brain was isolated, and basilar and middle cerebral arteries (0.4 to 0.8 mm outside diameter) were rapidly removed. The arteries were cut helically at an angle of approximately 45° into strips 20–25 mm in length. The helical strips were fixed vertically between hooks in a 20 ml muscle bath containing the nutrient solution. Hooks anchoring the upper end of the strips were connected to the lever of a force-displacement transducer (Nihonkoden Kogyo Co., Tokyo, Japan), and the resting tension was adjusted to 1.5 g which has been found to be optimal for producing the maximum contraction (Toda, Hayashi & Hatano, 1976). The bathing medium was maintained at 37 ± 0.5°C and aerated with a mixture of 95% O_2 and 5% CO_2. The composition of the solution was as follows (mM): Na^+, 162.1; K^+, 5.4; Ca^{++}, 2.2; Cl^-, 157.0; HCO_3^-, 14.9; and dextrose, 5.6. Before the start of experiments, the

preparations were allowed to equilibrate for 90 to 120 min in the bathing medium, during which time the fluid was replaced every 15 to 20 minutes.

The contractile response to 30 mM K^+ was first obtained and then the preparations were repeatedly washed. The dose-contraction relationship to dopamine or noradrenaline was then obtained and the values of contractions induced by these amines relative to the K^+-induced contraction are presented in the text and figures. In order to minimize the response to stimulation of α-adrenoceptors, preparations were treated for 60 min with 10 μM phenoxybenzamine. The phenoxybenzamine-containing solution was then replaced with normal solution in which the preparations were allowed to equilibrate for 30 to 40 minutes. Arterial strips were always contracted with prostaglandin $F_{2\alpha}$ for demonstration of the relaxant effects. After the dose-relaxation relationship of the amines had been obtained, papaverine in a concentration of 100 μM was added to obtain maximum relaxation (Toda, 1974). Values of the relaxation induced by the amines relative to the papaverine-induced relaxation are presented in the text and figures. Dopamine or noradrenaline was added directly to the bathing medium in cumulative concentrations. Preparations were treated for 20 min with blocking agents before the addition of dopamine or noradrenaline. Contractions and relaxations were displayed on an ink-writing oscillograph (Sanei Sokki Co., Tokyo, Japan). Results shown in the text, figures and table are expressed as mean values ± standard errors of the means. Statistical analyses were made using Student's *t* test.

Drugs used were dopamine hydrochloride, (±)-noradrenaline hydrochloride, phenoxybenzamine hydrochloride, phentolamine mesylate, (±)-propranolol hydrochloride, sotalol hydrochloride, aminophylline, atropine sulphate, adenosine, papaverine and prostaglandin $F_{2\alpha}$ (Ono Co.).

Results

Contraction induced by dopamine and noradrenaline

The addition of dopamine in concentrations ranging from 0.2 μM to 500 μM caused a dose-related contraction in canine isolated cerebral arteries. The maximum contraction induced by dopamine (458 ± 69 mg) was approximately double the contraction induced by 50 μM noradrenaline (270 ± 59 mg) (Figure 1). In all six strips in which the effect of dopamine and noradrenaline was compared, the dopamine-induced contraction was always greater. The addition of noradrenaline in a concentration of 50 μM elicited a slight, transient contraction followed by a sustained relaxation, and when the concentration was increased to 200 μM, a marked relaxation always occurred. The average ED_{50} of dopamine was approximately 100 times the value of noradrenaline (Table 1).

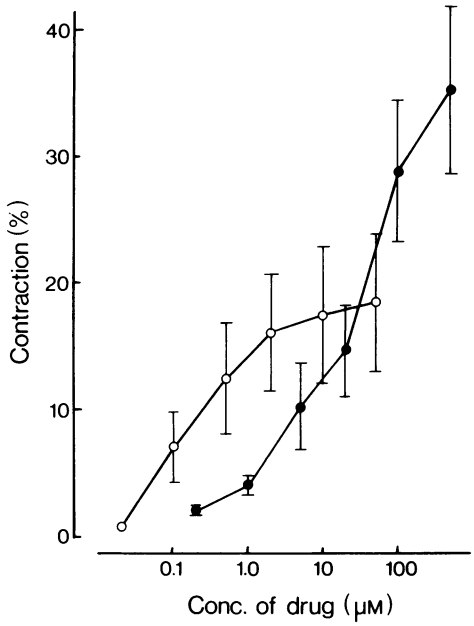

Figure 1 Dose-response curve to dopamine (●) and noradrenaline (○) in canine isolated cerebral arteries. Ordinate scale, mean values of contractions relative to the K^+ (30 mM)-induced contraction; mean absolute contractions induced by K^+ in experiments with dopamine and noradrenaline were 1424 ± 142 mg and 1382 ± 197 mg, respectively. Vertical bars represent s.e. means. The dopamine responses are the mean results from 22 preparations; those for noradrenaline are the mean results from 10 preparations.

The contractile response of cerebral arterial strips to dopamine and noradrenaline was reversed to a relaxation following treatment with phenoxybenzamine or phentolamine. Typical recordings of the effects of dopamine and noradrenaline before and

Table 1 Median effective concentrations of dopamine and noradrenaline in isolated cerebral arteries of the dog

Response	*Median effective concentrations (μM)*	
	Dopamine	*Noradrenaline*
Contraction	51 ± 7.4 (19)	0.42 ± 0.1 (10)
Relaxation	31 ± 9.1 (20)	79 ± 4.4 (10)

Figures in parentheses indicate the number of preparations used.

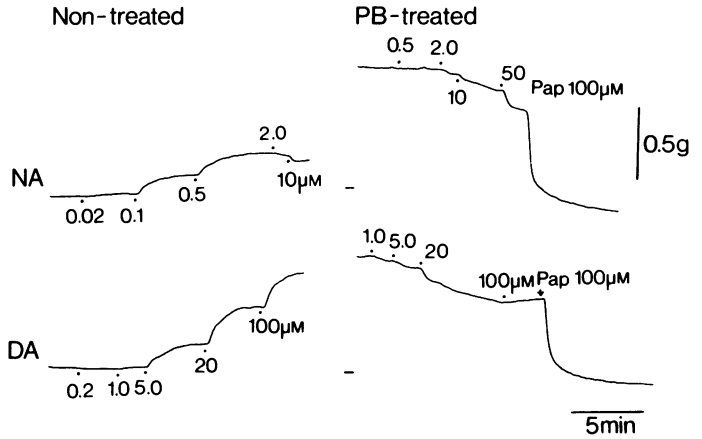

Figure 2 Responses of a basilar arterial strip to dopamine and noradrenaline. After the response to 30 mM K^+ (2.32 g contraction) was obtained and the preparation was repeatedly washed, dose-response relationships to noradrenaline and dopamine were obtained. The preparation was then treated for 60 min with 10 μM phenoxybenzamine (PB), washed and equilibrated in normal solution. When the prostaglandin (1 μM)-induced contraction levelled off, the dose-response relationship for noradrenaline was obtained and (100 μM) papaverine (Pap) was added. After repeated washing, the preparation was again treated for 20 min with 10 μM phenoxybenzamine and washed. The strip was contracted with 1 μM prostaglandin $F_{2\alpha}$ and the dose-relationship for dopamine was obtained. Horizontal lines just left of the right tracings represent the level prior to the addition of prostaglandin. Drug concentrations = μM.

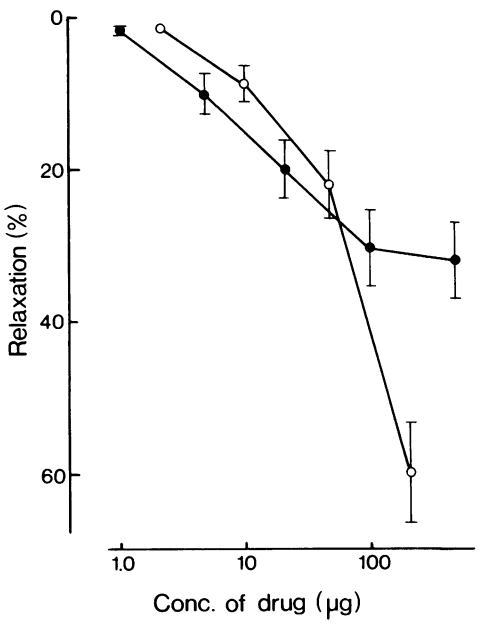

Figure 3 Dose-response curves to dopamine (●) and noradrenaline (○) in isolated cerebral arteries. Ordinate scale, mean values of relaxations relative to the papaverine (100 μM)-induced relaxation; mean absolute relaxations by papaverine in experiments with dopamine and noradrenaline were 694 ± 53 mg and 518 ± 94 mg, respectively. Average contractions induced by prostaglandin in experiments with dopamine and noradrenaline were 398 ± 38 mg and 322 ± 56 mg, respectively. Dopamine responses are mean results from 22 preparations, those for noradrenaline are the mean results from 14 preparations.

after treatment with phenoxybenzamine in a canine basilar artery are presented in Figure 2.

Relaxation induced by dopamine and noradrenaline

After treatment with phenoxybenzamine, the preparations were contracted with prostaglandin $F_{2\alpha}$ (0.1 to 3 μM) for the determination of dose-relaxation relationships. The results with dopamine and noradrenaline are summarized in Figure 3. The maximum relaxation attained with 200 μM noradrenaline was considerably greater than that with 500 μM dopamine. The mean value of the ED_{50} for dopamine was approximately one third the value for noradrenaline (Table 1).

In nine cerebral arterial strips treated with phenoxybenzamine and then washed, the dose-response curve to dopamine was not significantly altered by treatement for 20 min with 1 μM propranolol (Figure 4) or by 10 μM sotalol. On the other hand, the relaxation induced at low concentrations of noradrenaline (2 μM and 10 μM) was

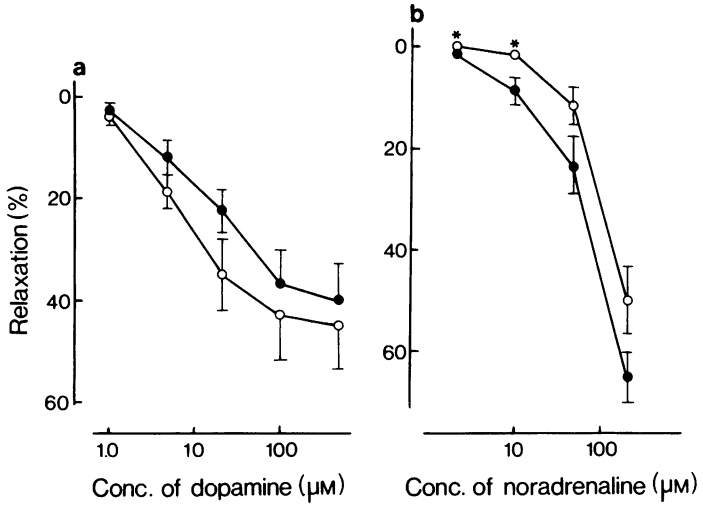

Figure 4 Modification by propranolol (●) of the relaxant effect of (a) dopamine and (b) noradrenaline in cerebral arterial strips. Ordinate scales, mean values of relaxations relative to papaverine (100 μM)-induced relaxation; mean absolute relaxations induced by papaverine in control (●) and propranolol-treated (○) preparations were 792 ± 80 mg and 742 ± 124 mg, respectively (a), and 390 ± 71 mg and 374 ± 75 mg, respectively (b). Average contractions induced by prostaglandin in control and propranolol-treated preparations were 408 ± 55 mg and 422 ± 52 mg, respectively (a) and 272 ± 60 mg and 278 ± 54 mg, respectively (b). In (a) 9 preparations were used for both control and propranolol-treated results; in (b) 10 preparations were used in each case. *, Significantly different from respective controls, $P < 0.05$.

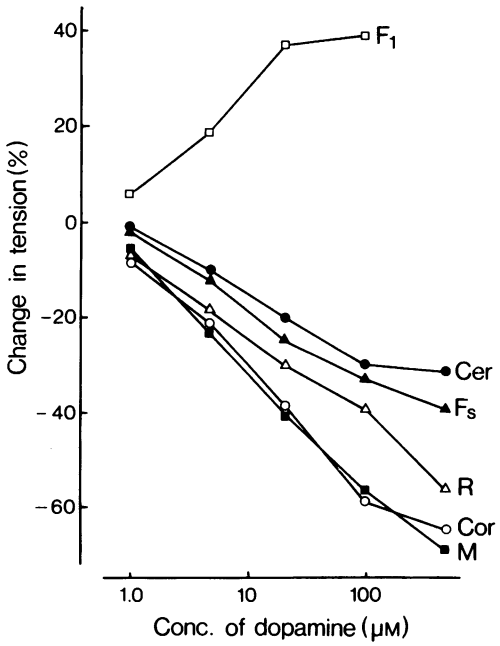

Figure 5 Comparisons of the relaxant effect of dopamine in different arteries from dogs. Ordinate scale, mean values of relaxations relative to the papaverine-induced relaxation except in large femoral arteries (F_1) in which the mean contraction relative to the contraction induced by prostaglandin added to contract the arteries prior to dopamine are presented. Data for mesenteric (M), renal (R), small femoral (F_s) and large femoral arteries (Goldberg & Toda, 1975) and those for coronary arteries (Cor) (Toda & Goldberg, 1975) have been published elsewhere. Cer, cerebral arteries. Preparations were treated with 10 μM phenoxybenzamine.

significantly attenuated by the same concentration of propranolol.

The relaxant effect of dopamine was also unaffected by aminophylline (20 μM), which significantly reduced the relaxation induced by 1 μM adenosine from 34.7 ± 5.2% to 15.8 ± 3.6% (values relative to papaverine-induced relaxation) ($n = 10$, $P < 0.01$). Atropine (1 μM) did not reduce the relaxation in response to dopamine.

Discussion

In isolated cerebral arteries of the dog, the addition of dopamine caused a contraction, the maximum contraction and the ED_{50} being greater than those seen with noradrenaline. In anaesthetized dogs, the major effect of dopamine intravenously applied is a vasoconstriction (von Essen, 1974). Such vascular

contraction observed *in vitro* and *in vivo* was reversed to a relaxation following treatment with α-adrenoceptor blocking agents, thus the contractile response is produced by the stimulation of α-adrenoceptors as is the case with noradrenaline. However, in isolated cerebral arteries, dopamine elicited a greater contraction than did noradrenaline, while the latter elicits a greater contraction in canine isolated renal, mesenteric and femoral arteries (unpublished data). Two possibilities might account for such a discrepancy: a greater affinity of dopamine for α-adrenoceptors in cerebral arteries and a greater relaxant effect of noradrenaline at high concentrations. When concentrations of noradrenaline were raised to 50 μM or higher, marked relaxation occurred. Further, in coronary arteries in which β-adrenoceptors are predominant, noradrenaline causes a relaxation, while dopamine causes a contraction (Toda & Goldberg, 1975). These findings suggest that the latter alternative is more likely.

Relaxation induced by dopamine in phenoxybenzamine-treated and prostaglandin-contracted cerebral arteries was not significantly influenced by propranolol, atropine or aminophylline at concentrations sufficient to attenuate the relaxant effect of noradrenaline, acetylcholine or adenosine, respectively. These findings support the hypothesis that β-adrenergic, cholinergic and adenosine-related mechanisms are not involved in the genesis of dopamine-induced relaxation in cerebral arteries as is the case in coronary arteries (Toda, Hojo, Sakae & Usui, 1975). The vasodilatory response of the cerebral circulation of anaesthetized dogs to dopamine is abolished by pimozide (von Essen, 1974), while the relaxant effect of dopamine in isolated arteries, including renal, mesenteric, coronary and cerebral, is not antagonized by dopamine antagonists, haloperidol, chlorpromazine, apomorphine and bulbocapnine, since these drugs applied at high concentrations produce marked relaxation in isolated arteries (Toda & Goldberg, 1975; Goldberg & Toda, 1975).

Regional differences in the response of isolated canine arteries of the dog to vasoconstrictor and dilator agents have been demonstrated (Bohr, Goulet & Taquini, 1961; Toda & Fujita, 1973; Toda, 1974). The relaxant effect of dopamine in different arteries under the same experimental conditions are summarized in Figure 5. The potency of dopamine in causing a relaxation is in the order, mesenteric = coronary > renal > small femoral > cerebral. Large femoral arteries contract in response to dopamine even after treatment with phenoxybenzamine. Relaxation caused by stimulation of specific receptors such as β-adrenoceptors and cholinoceptors, is appreciably less in cerebral arteries as compared with the relaxation observed in peripheral arteries (Toda, 1974), and the same is true in the case of dopamine, while non-specific relaxation induced by papaverine is not markedly different in cerebral and peripheral arteries.

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