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Effects of Tizanidine on Reflex Bradycardia in Rats

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ABSTRACT

The effects of intravenous (i. v.) administration of tizanidine on cardiovascular functions were assessed in both normal and catecholamine-depleted rats. Administration of tizanidine (1-10 μg/kg, i. v.) caused an initial increase in blood pressure, then followed by a dose-related decrease in both arterial pressure and heart rate in normal as well as catecholamine-depleted rats. The cardiovascular responses recovered about 20 min after the tizanidine injection. With regard to the reflex bradycardia produced by infusion of epinephrine (1. 25 or 2. 5 μg/kg i. v.) in rats, it was found that i. v. pretreatment of animals with tizanidine did not change the epinephrine-induced pressor effect. However, it did enhance the epinephrine-induced reflex bradycardia in normal rats, but attenuate that in catecholamine-depleted rats. Thus, the data indicate that tizanidine acts through a catecholaminergic mechanism to enhance the reflex bradycardia, which is mediated through baroreceptor reflexes in response to acute increase in blood pressure.

Key words: Tizanidine, Reflex bradycardia, Epinephrine, Baroreceptor.

Tizanidine, 5-chloro-4-(2-imidazolin-2-yl-amino)-2, 1, 3-benzothiadiazole, is a muscle relaxant with an imidazole structure and a pharmacological profile different from that of currently used, well-established myotonolytic drugs such as diazepam, baclofen, chlorphenesin, tolperisone and dantrolene. In previous paper, it reported that tizanidine could induce sedation, drowsiness and muscle relaxation in monkeys, inhibit spontaneous movement in mice, exert a powerful anti-spastic action on drug-induced convulsion in mice, and inhibit nictitating membrane contraction[1,2,3]. Furthermore, the studies on the effects of tizanidine on the spontaneous electroencephalogram and monoamine levels in rats indicated that tizanidine could increase dopamine levels in the cortex, decrease homovanillic acid (HVA), 3-methoxy-4-hydroxyphenyleneglycol (MHPG) and 5-hydroxyindole acetic acid (5-HIAA) levels in the cortex and HVA and MHPG in the brainstem, decrease slow wave sleep and increase regular θ wave in the cortex in the chronically electrode-implanted rats[4]. It has also been re-

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ported that tizanidine could inhibit α-and γ-rigidity in rats, reflex muscle tone in rabbits and the linguomandibular reflex in cats, but it had little or no effect on gross spinal reflexes or electrically induced sagmental reflexes in cats[5,6]. In the present study, the effects of tizanidine on the reflex bradycardia in response to arterial pressure elevation induced by i. v. infusion of epinephrine were studied in the normal and catechalamine-depleted rats.

MATERIALS AND METHODS

Animals and drugs

Male rats of Wistar strain, weighing about 250 g were used. Animals were housed at 25± 1°C and 50% humidity under a 12 hr light/dark cycle and had free access to food and water.

The drugs used in this research were obtained from the following sources: α-methyl-p-tyrosine (α-MT), reserpine and epinephrine, Sigma Chemical Co. (St. Louis, MO); tizanidine, Sandoz. Tizanidine, α-MT and epinephrine were dissolved in 0.9% saline, reserpine was dissolved in a few drops of glacial acetic acid and then diluted to required concentration with 50% of sucrose solution. All drug solutions were freshly prepared on the day of testing. Appropriate vehicle-injected controls were always run simultaneously.

Catecholamine-depleted rats were induced by pretreating the rats with reserpine combined with α-MT. Reserpine, 7. 5 mg/Kg was injected subcutaneously 24 hr before the experiment and α-MT, 250mg / Kg, was administrated intraperitoneally (i. p.), 6 hr before experiment.

Measurements of cardiovascular function

The animals were anesthetized with sodium pentobarbital (40mg/kg i. p.). Rectal temperature was maintained at 37. 5 ± 0. 5 °C throughout the course of the experiments by irradiation with infrared light. The trachea was cannulated and the left femoral artery was catheterized. The femoral arterial pressure was monitored with a Gould P23ID transducer, and heart rate was monitored with a Gould Biotach amplifier triggered by arterial pulses. The right femoral vein was cannulated for i. v. injection. All recordings were made on a four-channel Gould 2400S polygraph. The reflex bradycardia was induced by i. v. infusion of epinephrine in rats.

RESULTS

Effects of tizanidine on the basal levels of cardiovascular responses in normal and catecholamine-depleted rats

Administration of tizanidine (1-10 μg/kg, i. v.) caused an initial, slight increase in blood pressure, then followed by a slight but dose-related decrease in blood pressure and heart rate in normal (Fig. 1) as well as catecholamine-depleted rats (Fig. 2). The cardiovascular responses recovered about 20 min after tizanidine injection.

Effects of tizanidine on the epinephrine-induced reflex bradycardia in normal and catecholamine-depleted rats[7].

Intravenous administration of epinephrine produced a dose-related change in both arterial pressure and heart rate in rats. The reflex bradycardia was produced by an elevation in arterial pressure induced by epinephrine. Table 1 summarized the cardiovascular responses to epinephrine (1. 25 μg/kg and 2. 5 μg/kg i. v.) in both 0. 9% saline-controlled and the tizanidine-treated animals. Tizanidine (1-10 μg/kg i. v.)

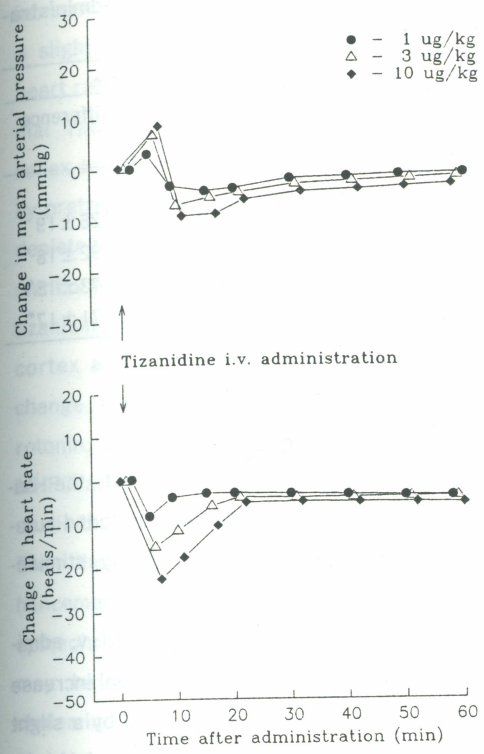

Fig. 1. Dose-related effects of tizanidine on blood pressure and heart rate in normal rats. Each plot denotes the mean value of 6 animals.

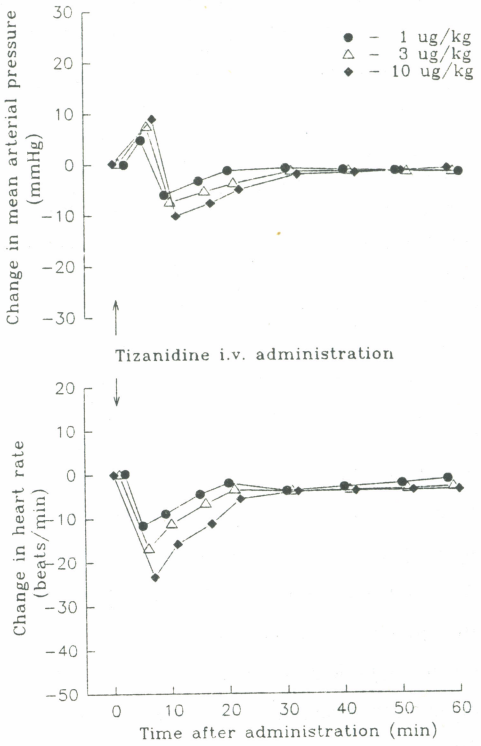

Fig. 2. Dose-related effects of tizanidine on blood pressure and heart rate in catecholamine-depleted rats. Each plot denotes the mean value of 6 animals.

Table 1. Effects of tizanidine on the cardiovascular responses induced by intravenous administration of epinephrine in normal rats

Treatment	Mean arterial pressure, mm Hg.			Heart rate, beats/min		
	Control	After epinephrine	Difference	Control	After epinephrine	Difference
Epinephrine 1.25 μg/kg i.v.						
0.9% saline	108±13	161±20	53± 6	405±51	353±61	52± 7
Tizanidine 1 μg/kg	110±17	160±23	50± 2	403±68	340±57	63± 9
Tizanidine 3 μg/kg	107±15	160±27	53± 8	410±59	329±60	81±10*
Tizanidine 10 μg/kg	105±12	161±30	56± 7	408±72	309±48	99±15*
Epinephrine 2.5 μg/kg i.v.						
0.9% saline	109±18	174±28	65± 9	406±65	336±60	70± 9
Tizanidine 1 μg/kg	104±11	167±25	63±11	411±73	319±53	92±11
Tizanidine 3 μg/kg	110±16	177±31	67±15	405±68	296±49	109±14*
Tizanidine 10 μg/kg	105±15	166±30	61±11	409±83	290±51	119±19*

*Significantly different from corresponding control value (0.9% saline group), $P<0.05$ (Student's t-test). The values are expressed as the mean ± SE of 10 animals.

Table 2. Effects of tizanidine of the cardiovascular responses induced by intravenous administration of epinephrine in catecholamine-depleted rats

Treatment	Mean arterial pressure, mm Hg.			Heart rate, beats/min		
	Control	After epinephrine	Difference	Control	After epinephrine	Difference
Epinephrine 2.5 μg/kg i.v.						
0.9% saline	76±13	149±19	73±10	323±37	197±30	126±19
Tizanidine 1 μg/kg	76±21	147±25	71±13	328±48	236±39	92±18
Tizanidine 3 μg/kg	78±25	145±21	67±11	321±59	248±26	73±15*
Tizanidine 10 μg/kg	78±15	144±16	66± 8	324±38	253±28	71±17*

* Significantly different from corresponding control value (0.9% saline group), $P<0.05$ (Student's t-test). The values are expressed as the mean ± SE of 10 animals.

were administered 20 min before the i. v. administration of epinephrine. As shown in Table 1, in animals pretreated with tizanidine, the bradycardic responses were enhanced significantly, although the responses of arterial pressure were not significantly different from that of control. The enhancement of bradycardia responses induced by tizanidine administration were significantiy attenuated in catecholamine-depleted rats as shown in Table 2.

DISCUSSION

It is well accepted that the key to the regulation of cardiovascular function lies in the reciprocal relationships between vagal tone and sympathetic efferent activity. The activity in the afferent fibers from baroreceptors activated by an increase in arterial blood pressure provides a major inhibitory force of central vasomotor tone[8,9]. The central baroreceptor arc is polysynaptic, with the primary synapse in the nucleus of the tractus solitarius(NTS), while its inhibiting neurons interposed between the NTS and the cardiovascular center. It is known that the baroreceptor afferents not only exert an influence on neuronal activity in the pons and medulla oblongata but also affect the supramedullary regions from which profound autonomic responses can be electrically stimulated[10,11].

The present results showed that i. v. administration of tizanidine caused an initial increase in blood pressure and then followed by a slight dose-related decrease in the basal level of both heart rate and arterial blood pressure in normal rats. In addition, the epinephrine-induced reflex bradycardia was enhanced by pretreatment of animals with tizanidine. In order to assess whether the catecholamine containing neurons are involved in the regulation of tizanidine on cardiovascular functions, reserpine and α-MT are employed to deplete catecholamine contents in this experiment. Reserpine depletes the stores of catecholamine of adrenergic nerve endings in the brain as well as the peripheral organs[12,13], yet α-MT inhibits the activity of tyrosine hydroxylase which is the enzyme implicated in the rate-limiting step in the biosynthesis of catecholamine[14]. The effects of reserpine and α-MT on endogenous catecholamine levels and its synthesis had been reported[15,16]. Following these pretreatment to deplete catecholamine levels, tizanidine still caused an initial

increase in blood pressure and then followed by a slight decrease in the basal levels of both heart rate and arterial pressure as found in normal rats. However, the epinephrine-induced reflex bradycardia was significantly attenuated by pretreatment of tizanidine in catecholamine-depleted rats. In our previous report, as mentioned in introduction, it indicated that tizanidine increased the level of dopamine in the cortex and brainstem, but it almost did not change the levels of noradrenaline and serotonin. On the other hand, the levels of HVA, MHPG and 5-HIAA decreased in the cortex and so did that of HVA and MHPG in the brainstem[4]. From these results, it came to light that the enhancement caused by tizanidine on the epinephrine-induced reflex bradycardia of rats may be due to the inhibition of the catecholamine turnover and then through a mechanism to enhance the vagal tone and/or to attenuate the preganglionic sympathetic efferent activity which leads to enhance the epinephrine induced reflex bradycardia.

The arterial baroreflex system is regarded as one of the most powerful and rapidly acting homeostatic mechanisms for the regulation of blood pressure. It is also well known that baroreflex sensitivity is significantly depressed in human[17,18] and experimental hypertensive animals[19,20]. In fact, more and more accumulated evidences have suggested that the monoaminergic system within the brain forms a certain essential links in the central connections of arterial baroreceptor reflexes. For example, it has been shown that the development of neurogenic hypertension in the rabbit was accompanied with an increase in norepinephrine turnover in the thoracic and lumbar spinal cord[21] and the hypertension caused by the excision of the baroreceptor nerves could be prevented by central administration of 6-hydroxydopamine which selectively destroyed the catecholaminergic fibers[22]. Moreover, the neurogenic hypertension in the rat induced by the destruction of the nuclei solitarius could also be prevented by central administration of 6-hydroxydopamine[23]. Depletion of epinephrine in the brain by means of intraperitoneal administration of phenylethanolamine N-methyltransferase inhibitors had been shown to facilitate the reflex bradycardia[24]. On the other hand, it was found that intracisternal injection of apomorphine (a dopamine receptor stimulating agent) caused an increase in arterial pressure in rats[25]. Local injection of apomorphine into the caudate-putamen complex facilitated the reflex bradycardia, while intra caudate-putamen complex injection of dopamine antagonist such as haloperidol and pimozide inhibited it[26]. As for the serotoninergic system, elevating brain content of serotonin depressed the epinephrine-induced bradycardia, whereas depleting that content enhanced the epinephrine-induced bradycardia in rats[27]. Therefore, the interrelationships between the brain catecholaminergic neurons and tizanidine-sensitive neurons in the regulation of cardiovascular function need further investigation.

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Tizanidine 對反射性徐脈之作用

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本實驗係探討靜脈注射 Tizanidine 對正常及兒茶酚胺耗竭大白鼠心血管功能之影響。無論是正常或是兒茶酚胺耗竭之大白鼠，靜脈注射 Tizanidine (1-10μg/kg)皆先導致血壓上升，接著產生與劑量有關的血壓下降與心跳減緩。這種心血管反應約在注射 Tizanidine 20 分鐘之後恢復正常。至於由靜脈注射腎上腺素(1. 25, 2. 5μg/kg)所產生的反射性徐脈現象，事先投予 Tizanidine 並不影響腎上腺素造成的血壓效應，然而它會增強腎上腺素對正常鼠引起之反射性徐脈現象，卻減弱兒茶酚胺耗竭大白鼠之反射性徐脈。由這些數據顯示，Tizanidine 可能是作用於兒茶酚胺機轉以增強在急性血壓上升後，經由壓力接受體反射途徑所產生反射性徐脈現象。

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