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The effect of atenolol on the spontaneous and reflex activity of the sympathetic nerves in the cat: influence of cardiopulmonary receptors

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1 Atenolol reduces sympathetic efferent discharge and attenuates the responses of the sympathetic nerves to changes in blood pressure. The present experiments were carried out to determine whether these changes were mediated by cardiopulmonary receptors whose afferents lie in the vagal nerves.

2 Cats were anaesthetized with α-chloralose and artificially ventilated. In one group of cats recordings were made of sympathetic efferent discharge from few-fibre preparations from the lumbar trunk, splanchnic or renal nerves over a range of blood pressures. In a second group of cats changes in heart rate and blood pressure in response to bilateral occlusion of the common carotid arteries were investigated. In all cats the influence of vagal afferent fibres was removed by cooling both vagal nerves in the neck, both before and after administration of atenolol (3 mg kg^{-1} i.v.).

3 Cooling both vagal nerves produced significant increases in blood pressure, heart rate and spontaneous sympathetic efferent discharge but did not affect the relationship between sympathetic efferent discharge and mean blood pressure or the responses to carotid occlusion. Atenolol significantly reduced blood pressure, heart rate and sympathetic efferent discharge but the change in sympathetic efferent discharge on vagal cooling was less than before giving the drug. Atenolol also attenuated the reflex responses of the sympathetic nerves to changes in blood pressure and reduced responses to carotid occlusion. This attenuation was not removed by vagal cooling.

4 Thus, neither the reduction in spontaneous sympathetic efferent discharge nor the attenuation of the baroreceptor reflex seen after atenolol, are due to an increased input to the brain from vagal afferent fibres. Other possible mechanisms whereby atenolol might exert its effects on the sympathetic nerves are discussed.

Introduction

There is now evidence that the administration of a number of β-adrenoceptor blocking agents can affect both the spontaneous and the reflexly induced activity of the sympathetic nerves (e.g. Lewis & Haeusler, 1975; Friggi, Chevalier-Cholat & Bodard, 1977a; Scott, 1981). The reduction of the sympathetic efferent discharge produced by propranolol has been used as evidence to support a central site for the hypotensive actions of this drug (e.g. Lewis & Haeusler, 1975). However, a reduction in sympathetic efferent discharge has also been observed (Scott, 1981), after the administration of atenolol. Since this drug does not readily enter the brain (Cruickshank, Neil-Dwyer, Cameron & McAinsh, 1980) these observations provide evidence for a peripheral action, at least for atenolol. Atenolol has also been shown to attenuate the responses of the sympathetic nerves to pharmacologically induced changes in the blood pressure (Scott, 1981).

Thus, there is evidence that at least one β-adrenoceptor blocking drug, atenolol, may exert its action on the sympathetic nervous system by modifying the input to the brain. These changes both in the resting level of the sympathetic efferent discharge and in the reflex responses of the sympathetic nerves to changes in the blood pressure may reflect a change in the sensitivity of the afferent limb of the baroreceptor reflex arc. However, they might also result from changes in the level of activity of other receptors. There is evidence that cardiopulmonary receptors whose afferents lie in the vagal nerves may tonically inhibit the sympathetic nerves (see Thorén, 1979 for references). After giving a β-adrenoceptor blocking drug such as atenolol there are widespread cardiovascular changes that are likely to affect the discharge from a number of cardiopulmonary receptors. Sectioning the vagi removes the inhibition on the sympathetic nerves and results in an increase in

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the level of sympathetic efferent discharge (Thorén, 1979). It has also been suggested (Koike, Mark, Heistad & Schmid, 1975) that changes in the discharge of these cardiopulmonary receptors might affect the sensitivity of the baroreceptor reflex arc.

The present experiments were carried out to assess the contribution made by cardiopulmonary receptors whose afferent fibres travel in the vagal nerves, to the changes in the sympathetic nervous discharge produced by atenolol. If the reduction in the level of sympathetic efferent discharge produced by atenolol was secondary to changes in the discharge from cardiopulmonary receptors then the effects of removal of these receptors would be greater after atenolol than before giving the drug; that is, cooling the vagal nerves would result in a greater increase in the sympathetic efferent discharge after atenolol than before giving the drug. Cooling the vagal nerves might also be expected to remove the attenuation of the reflex changes in the sympathetic nervous discharge in response to changes in the blood pressure observed after atenolol.

Methods

Experiments were carried out on 11 cats of either sex weighing between 1.8 and 4.0 kg. The animals were anaesthetized by an intraperitoneal injection of α-chloralose in polyethylene glycol (100 mg ml^{-1}) in a dose of 80 mg kg^{-1}. Further injections of α-chloralose were given intravenously in doses of 2–15 mg $kg^{-1}h^{-1}$ as necessary to maintain the animals in a steady state of light anaesthesia.

The trachea was cannulated and positive pressure ventilation was established with a mixture of approximately 40% O_2 in air. The femoral artery and vein were cannulated in the groin. The pH, P_{CO_2} and P_{O_2} of the arterial blood were monitored throughout the experiment. The ventilation volume was adjusted to maintain the arterial P_{CO_2} at approximately 35 mmHg and infusions of sodium bicarbonate (1 mEq ml^{-1}) were given as necessary to correct for the presence of any non-respiratory acidosis using the *in vivo* titration curves of Kappagoda, Linden & Snow (1970). Rectal temperature was monitored and maintained between 37° and 38°C. Dextraven 150 was given during the experiment to compensate for any loss of fluid. The total volume of fluid given over the period of an experiment into each animal was approximately 10–15% of its blood volume (assumed to be 80 ml kg^{-1}). Blood pressure was monitored throughout the experiment through a cannula inserted into the femoral artery and attached to a strain gauge manometer (Model P23 Gb, Statham Inst. Co. Inc. Puerto Rico). The mean pressure in the femoral artery was calculated by adding one third of the pulse pressure to the diastolic pressure. Pressure in the right atrium was measured through a cannula inserted into the jugular vein.

In 7 cats recordings of the sympathetic efferent discharge were made. A ventral incision was made through the skin and abdominal muscle on the left side and the aorta and renal artery were exposed by a retroperitoneal approach. The skin and muscle flaps were then retracted to form an abdominal pool. Further dissection was carried out under a dissecting microscope. Small filaments were dissected from the cut central end of the renal nerves, lumbar trunk or splanchnic nerve and placed on platinum recording electrodes. Recordings of the discharge of these few-fibre preparations were made under liquid paraffin using conventional techniques and displayed on an ultra-violet recorder. The mean frequency of sympathetic efferent discharge was recorded every second by the use of a digital counter whose output was displayed on the ultra-violet paper.

In a further 4 cats no recordings of the sympathetic efferent discharge were made. In these cats the common carotid arteries were dissected free in the neck and loose snares placed around them on both sides.

In all 11 cats both vagi were dissected free in the neck. The skin was retracted to create a pool. Small glass probes were positioned such that they could easily be placed under the vagal nerves. A mixture of iced water and salt was pumped through the glass probes by means of a peristalic pump (Model MHRE 200, Watson-Marlow Ltd, England) so that the temperature of the fluid within the probes was between −2°C and 0°C. When the vagi were placed over these probes the cardiovascular changes produced were comparable to those produced by bilateral vagal section. It was therefore concluded that this system of cooling produced a complete blocking of activity in all fibres in the vagal nerves.

Experimental protocol

In the first group of 7 cats, dissection was continued until recordings from baroreceptor-dependent fibres were obtained and control recordings were taken over a period of at least 30 min. The blood pressure was then raised or lowered by a bolus injection of phenylephrine (3–10 μg kg^{-1}) or glyceryltrinitrate (40–60 μg kg^{-1}) and recordings of the sympathetic efferent discharge were made over a wide range of blood pressures when the blood pressure had remained steady for at least 20 s. The blood pressure was allowed to return to control levels and further control records were obtained. Both vagal nerves were then placed on the cooling probes and recordings of the sympathetic efferent discharge made over a range of blood pressures as before after which the cooling probes were removed and further control

records obtained. Atenolol (i.v. 3 mg kg^{-1} Tenormin, Stuart Pharmaceuticals Ltd) was then administered. Further records were obtained at least 30 min after the administration of atenolol. The effect of cooling both vagal nerves on the relationship between the sympathetic efferent discharge and the blood pressure was investigated as before.

In the second group of 4 cats the effects of bilateral occlusion of the common carotid arteries was investigated before and at least 30 min after giving atenolol (3 mg kg^{-1}) both in the control period and when both vagal nerves were cooled in the neck.

Analysis of data

In both groups of cats a comparison of the variables measured was made during the different interventions using Student's paired *t* tests (Snedecor & Cochran, 1967).

A linear regression analysis (Snedecor & Cochran, 1967) was carried out to determine the relationship between the level of sympathetic efferent discharge and the mean blood pressure before and after the different interventions. Comparisons of the slopes of the regression lines for all the cats were made using Student's paired *t* tests.

Results

When recording started approximately 4 h after the initial dose of anaesthetic, the mean blood pressure was 103.9 ± 7.1 mmHg (mean ± s.e.mean; range 64 to 141 mmHg) and the mean heart rate was 181 ± 5.9 beats min^{-1} (range 156–212 beats min^{-1}). The mean pH, P_{CO_2} and P_{O_2} of arterial blood were 7.38 ± 0.01 (range 7.35 to 7.46), 35.2 ± 1.4 mmHg (range 27 to 40 mmHg) and 125.0 ± 7.9 mmHg (range 97 to 156 mmHg) respectively.

Effects of vagal cooling

An example of the effect of cooling both vagal nerves in one cat is shown in Figure 1. In this cat, the cooling procedure resulted in an increase in the blood pressure from a control value of 87 mmHg to 103 mmHg and in the level of sympathetic efferent discharge from 25 to 55 impulses s^{-1}. Results from 7 cats are shown in Table 1. Cooling both vagal nerves resulted in significant increases in the mean blood pressure ($P<0.001$), heart rate ($P<0.025$) and in the sympathetic efferent discharge ($P<0.005$), but did not produce any significant effects on the right atrial pressure.

In 7 cats there was an inverse relationship between the sympathetic efferent discharge and the mean blood pressure as has been reported previously (Scott, 1981), but cooling both vagal nerves had no significant effect on this relationship (see Table 1).

In a further 4 cats cooling both vagal nerves had no consistent effects on the magnitude of the blood pressure or heart rate responses to carotid occlusion (see Table 1).

Effects of atenolol

In 7 cats the administration of atenolol (3 mg kg^{-1} i.v.) resulted in significant reductions (P always <0.01) in the blood pressure, heart rate and sympathetic efferent discharge as reported previously (Table 1). However, the increase in the mean right atrial pressure was not statistically significant. Atenolol also produced a significant ($P<0.025$) attenuation of the reflex responses of the sympathetic nerves to changes in the blood pressure (see Table 1) as has been reported previously (Scott, 1981). In 4 cats atenolol produced a significant attenuation of the reflex response in blood pressure ($P<0.01$) and heart rate ($P<0.05$) to carotid occlusion (see Table 1).

Effects of vagal cooling after atenolol

After atenolol, cooling both vagal nerves still resulted in a significant increase in the sympathetic efferent discharge ($P<0.01$). However, the increase in the mean blood pressure was not statistically significant. The increases in sympathetic efferent discharge and blood pressure produced by cooling after atenolol were not significantly greater than the increases observed before giving the drug. In fact, the mean increase in sympathetic efferent discharge on cooling after atenolol was only 5.9 ± 1.5 impulses s^{-1}, significantly less ($P<0.05$) than the increase of 15.7 ± 3.6 impulses s^{-1} observed on cooling before giving the drug. Cooling both vagal nerves again resulted in a significant ($P<0.05$) increase in the heart rate. However, this change of 6 beats min^{-1} is significantly less than the mean value of 25 beats min^{-1} observed during vagal cooling before giving atenolol ($P<0.025$). Cooling had no significant effect on the right atrial pressure.

After giving atenolol, the cooling procedure still did not alter the change in sympathetic efferent discharge per mmHg change in the blood pressure and the values remained significantly smaller ($P<0.025$) than those recorded in the control situation before giving atenolol (see Table 1). Thus, cooling both vagal nerves did not remove the attenuation of the reflex responses of the sympathetic nerves produced by the administration of atenolol.

In 4 cats, cooling both vagal nerves had no significant effects on the magnitude of the responses of blood pressure or heart rate to carotid occlusion. The

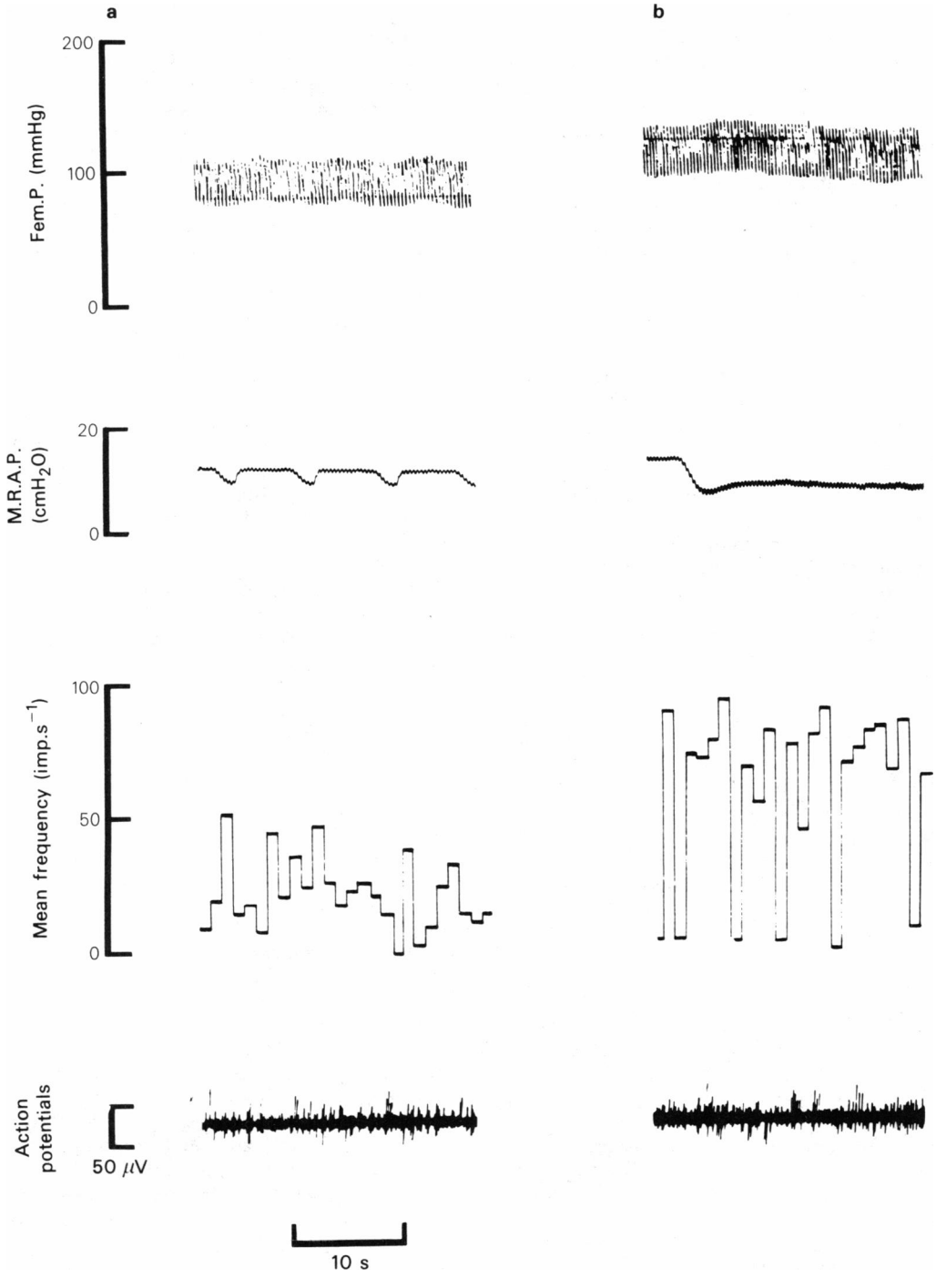

Figure 1 Original records showing the effects of cooling both vagal nerves in the neck. Left hand panel (a): before cooling; right hand panel (b): during vagal cooling. From above downwards: Fem.P., pressure in the femoral artery; M.R.A.P., mean pressure in the right atrium; mean frequency of sympathetic efferent discharge recorded every second; (action potentials) records of the discharge recorded from the splanchnic nerve.

Table 1 The effect of cooling both vagal nerves before and after the acute administration of atenolol (3 mg kg^{-1})

	n	*Before atenolol* Control	*Before atenolol* During vagal cooling	*After atenolol* (3 mg kg^{-1}) Control	*After atenolol* (3 mg kg^{-1}) During vagal cooling
Fem. P. (mmHg)	7	102.6± 5.8	121.6± 6.2	91.0±5.8	100.0±8.4
HR (b min^{-1})	7	177.0±13.5	201.6±13.1	153.9±4.0	159.7±3.9
RA_p (cm H_2O)	7	9.7± 1.5	9.1± 1.6	11.0±1.7	10.9±1.8
SED (imp s^{-1})	7	21.1± 4.1	36.8± 6.0	17.4±3.4	23.3±4.5
SED (imp s^{-1} mmHg change in Fem. P.$^{-1}$)	7	−0.44± 0.12	−0.47± 0.20	−0.12±0.05	−0.10±0.02
Control Fem. P. (mmHg)	4	92.8±11.8	116.6±18.7	89.1±5.7	101.5±5.4
ΔFem. P. (mmHg) during carotid occlusion	4	42.9± 9.0	38.0± 8.3	32.1±7.0	23.0±6.7
Control HR (b min^{-1})	4	170.6± 5.4	184.5± 9.7	151.0±4.9	151.4±5.2
ΔHR (b min^{-1}) during carotid occlusion	4	9.6± 2.2	7.0± 2.1	2.0±0.9	1.6±0.5

From above downwards: (all values mean±s.e.mean) Fem.P., pressure in the femoral artery (mmHg); HR = Heart rate (beats min^{-1}); RA_p = mean pressure in the right atrium (cmH_2O); SED (imp s^{-1}) = mean rate of sympathetic efferent discharge; SED (imp s^{-1} $mmHg^{-1}$) = mean change in SED per mmHg change in femoral arterial pressure; Control Fem.P. (mmHg) = pressure in the femoral artery before carotid occlusion; ΔFem.P. (mmHg) = change in femoral arterial pressure during carotid occlusion; Control HR (b min^{-1}) = heart rate before carotid occlusion and ΔHR (b min^{-1}) = change in heart rate during carotid occlusion.

changes in blood pressure and heart rate remained significantly less than before giving atenolol ($P<0.025$).

Discussion

The present studies confirm the work of Friggi *et al.* (1977a) and of Scott (1981) who demonstrated that, in the anaesthetized animal, the administration of atenolol reduced the spontaneous sympathetic efferent discharge. Atenolol was also found to attenuate the reflex responses to pharmacologically induced falls in blood pressure as described by Scott (1981), and to attenuate both the pressor and the heart rate response to bilateral occlusion of the common carotid arteries. This latter finding is consistent with the changes observed in the responses of the sympathetic nerves (Scott, 1981) and has also been shown for propranolol (Dunlop & Shanks, 1969). However, the present series of experiments does not provide any support for the hypotheses either that the reduction in sympathetic efferent discharge is a result of an increased inhibition of the sympathetic nerves mediated by vagal afferent fibres or that the attenuation of the reflex responses to changes in blood pressure is due to an increase in the discharge of vagal afferent fibres.

In the present study, before the administration of atenolol, removal of the influence of all vagal afferent fibres by cooling both vagal nerves to below 0°C resulted in an increase in the blood pressure and in the sympathetic efferent discharge. This tonic inhibition of the sympathetic nerves by vagal afferent fibres has been described before and is thought to be mainly due to the small unmyelinated fibres in the vagal nerves (see Thorén, 1979). Discharge in the larger myelinated afferent fibres from the complex unencapsulated endings in the atrial walls as described by Coleridge, Hemingway, Holmes & Linden, (1957) has also been shown to inhibit sympathetic efferent discharge in fibres in the renal nerves (Kidd, Linden & Scott, 1981) but does not produce the more widespread inhibitory effects such as are caused by the C fibre input. However, after atenolol, even though there was a small increase in atrial pressure, the influence of these vagal afferent fibres on the sympathetic nerves was shown to be reduced rather than increased: after atenolol, vagal cooling caused smaller increases in the sympathetic efferent discharge than were observed before giving the drug. Thus, the reduction in spontaneous sympathetic efferent discharge observed after atenolol, cannot be explained by an increase in the inhibitory effects exerted by the vagal afferent fibres on the sympathetic nerves.

There are, of course, many other explanations for the reduction in sympathetic efferent discharge observed after atenolol. Since this drug does not readily enter the brain (Cruickshank, *et al.*, 1980) its effects are most likely to be mediated at a peripheral rather than a central site. In the present series of experiments both the sympathetic afferent fibres and the carotid sinus afferent fibres remained intact. It is however, difficult to explain the reduction in sym-

pathetic efferent discharge by a change in the discharge of sympathetic afferent fibres. These fibres have been shown to increase their discharge when the atrial walls are distended (Lombardi, Malliani & Pagani, 1976), as would occur when atrial pressure increased after atenolol, and then to have an excitatory effect on the discharge of the sympathetic nerves. The reduction in sympathetic efferent discharge could however, be due to an increase in the discharge of the carotid sinus baroreceptors. Friggi, Chevalier-Cholat & Torresani, (1977b) observed a relative increase in aortic nerve discharge after propranolol, pindolol or timolol compared to the control period at the same blood pressure level. Such a resetting of the baroreceptors, producing an increase in the baroreceptor discharge in spite of the fall in blood pressure seems at present the most likely explanation for the reduced spontaneous sympathetic nervous activity observed after atenolol.

In the present series of experiments atenolol was also shown to reduce the reflex responses of the efferent sympathetic nerves to falls in the blood pressure and the pressure within the carotid sinus. This attenuation was still present when the influence of the vagal afferent fibres had been removed. Thus, in this study there was no evidence that a reduction in the discharge in vagal afferent fibres produced by cooling both vagi had any consistent effect on the sensitivity of the baroreceptor reflex arc. Although interactions between carotid sinus baroreceptors and receptors within the thorax are well documented (see Abboud, 1979 for references), it may be that in the present experiments the resting discharge of the vagal afferent fibres was too low to influence the sensitivity of the baroreceptor reflex. It is likely that, in the volume expanded animal when the discharge from these vagal afferent fibres would be greater, an increased sensitivity of the baroreceptor reflex arc could have been demonstrated on vagal cooling. However, in the present study there was no evidence to suggest that the attenuation of the reflex responses to changes in blood pressure, produced by atenolol was due to the changing influence of vagal afferent fibres.

There are, of course, other groups of receptors whose discharge might change after atenolol and which might alter the sensitivity of the baroreceptor reflex. It is conceivable that an increase in the sympathetic afferent input might modulate the baroreceptor reflex. The examples of interactions between differing groups of receptors are now so diverse (see Abboud, 1979 for references) that it would be surprising if the sympathetic afferent fibres and barorecptor fibres were found not to interact, although at present there is no firm evidence to support such a view. Interactions between carotid sinus and aortic arch baroreceptors have been shown (Kendrick, Matson & Lalley, 1979). There is also evidence of synergistic interactions between the large myelinated A fibre baroreceptors and the smaller unmyelinated C fibre baroreceptors (Aars, 1980). Since the C fibres tend to be activated at higher blood pressures it may be that after giving atenolol when the blood pressure is lowered there is a reduction in the numbers of C fibres tonically discharging and hence, due to the loss of synergistic interaction between the A and C fibres an attenuation of the reflex responses to changes in blood pressure.

Alternatively, atenolol might produce its effects on the sympathetic nervous system either by directly modifying the afferent pathway of the baroreceptor reflex, or by an indirect mechanism, for example, by altering plasma renin activity. Further work is required in order to differentiate between these many possible mechanisms of action.

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