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Regional differences in cardiac effects of pituitary adenylate cyclase-activating polypeptide-27 in the isolated dog heart

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Abstract

Recent observations indicate that several neuropeptides may be involved in the regulation of cardiac function, but the effects of these peptides on the atrium are not always the same as those on the ventricle. To compare the effect of pituitary adenylate cyclase-activating polypeptide (PACAP)-27 on the atrium with that on the ventricle, we investigated the effects of PACAP-27 on the sinus rate and atrial and ventricular contractility in isolated, blood-perfused dog heart preparations. PACAP-27 (0.01–0.3 nmol) caused transient positive followed by negative chronotropic and inotropic responses in a dose-dependent manner in the isolated right atrium, whereas it caused only a dose-dependent positive inotropic response in the left ventricle. After atropine treatment, PACAP-27 caused only positive cardiac responses in isolated atria. The order of the increase in response to PACAP-27 was atrial contractile force > sinus rate ≥ ventricular contractile force. Tetrodotoxin blocked the negative chronotropic and inotropic responses to PACAP-27 in isolated atria. Propranolol did not affect the positive response. PACAP-(6-27), a type I PACAP receptor antagonist, attenuated the positive responses similarly in the atropine-treated right atrium and the left ventricle. Thus, we demonstrated that (1) PACAP-27 caused negative cardiac effects in the atrium and sinoatrial node by activation of intracardiac parasympathetic nerves, but had no negative effect on the ventricle; (2) PACAP-27 had positive effects in the atrium, sinoatrial node and ventricle mediated by type I PACAP receptors, but PACAP-27 was more effective in the atrium and sinoatrial node than in the ventricle of the dog heart.

Keywords: PACAP-27 (pituitary adenylate cyclase-activating polypeptide-27); Chronotropism; Inotropism; Atrium; Ventricle; Heart; (Dog)

1. Introduction

Cardiac function is regulated by the autonomic nervous system. The magnitude of the cardiac response to stimulation of the stellate ganglia and vagus nerves parallels the density of the sympathetic and parasympathetic nerves in the heart, respectively (Levy and Martin, 1979). This relatively simple description of autonomic cardiac control has been complicated by the recent demonstration of neuropeptides in the heart. For example, neuropeptide Y caused a negative inotropic effect on the isolated dog atrium and ventricle with similar potency (Rigel et al., 1989). However, this peptide did not change the sinus rate in doses that decreased the atrial contractile force in isolated dog heart preparations (Ren et al., 1991). Vasoactive intestinal peptide (VIP) produced positive chronotropic and inotropic effects, but the positive inotropic response to VIP in the isolated ventricle was smaller than the response to VIP in the isolated atrium (Karasawa et al., 1990). These results indicate that these neuropeptides may be involved in the regulation of cardiac function, but the effects of neuropeptides on the atrium are not always the same as those on the ventricle.

Pituitary adenylate cyclase-activating polypeptide (PACAP) is a newly discovered neuropeptide that was isolated originally from the ovine hypothalamus (Miyata et al., 1989). PACAP is present in two molecular forms with 38 (PACAP-38) and 27 (PACAP-27) amino acid residues (Miyata et al., 1990). Interestingly, PACAP and VIP are members of a family of structurally related regulatory neuropeptides (Miyata et al., 1990). The concentration of radioimmunoassayable PACAP was recently measured in the rat heart (Arimura et al., 1991) and three types of PACAP receptors were detected in human and rat hearts (Inagaki et al., 1994; Sreedharan et al., 1995; Usdin et al., 1994; Wei and Mojsov, 1996). Our previous studies showed that PACAP-38 caused transient positive followed by neg-

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ative chronotropic and inotropic responses in isolated dog atria (Yonezawa et al., 1996) and PACAP-27 caused biphasic chronotropic effects in the anesthetized dog heart (Hirose et al., 1997). However, it has not yet been elucidated whether PACAP-27 affects the sinus rate, atrial and ventricular contractility similarly in the mammalian heart. Therefore, we investigated the effects of PACAP-27 on the sinus rate, atrial contractile force and ventricular contractile force in isolated, blood-perfused dog heart preparations. Additionally, to determine which receptors PACAP-27 activates in the atrium and ventricle, we studied the effects of PACAP-(6-27), a type I PACAP receptor antagonist, on the positive cardiac responses to PACAP-27 in isolated dog heart preparations.

2. Materials and methods

The animal experiments were approved by the Shinshu University School of Medicine Animal Studies Committee.

2.1. Isolated, blood-perfused dog heart preparations

Isolated right atria and left ventricles were obtained from 20 dogs (weighing 7–14 kg) anesthetized with sodium pentobarbital (30 mg/kg, i.v.). Each preparation was perfused with heparinized arterial blood from a second (support) dog. The details of these preparations have been described earlier (Chiba et al., 1975; Chiba, 1976).

The 20 support dogs, weighing 9 to 34 kg, were anesthetized with pentobarbital sodium (30 mg/kg, i.v.) and ventilated artificially through a cuffed tracheal tube with room air by using a Harvard respirator (Southnatick, MA, USA). Sodium heparin (500 USP units/kg, i.v.) was administered to each dog at the beginning of the perfusion of the isolated atrial or ventricular preparation and 200 USP units/kg was given subsequently at 1-h intervals.

After sodium heparin (200 USP units/kg, i.v.) was administered, the right atrium or the left ventricle was excised and immersed in cold Ringer's solution of the following composition (millimolar): NaCl, 154.0; KCl, 5.6; $CaCl_2$, 2.2 and $NaHCO_3$, 3.6. The wet weight of the isolated right atrial and left ventricular preparations varied from 5 to 11 g and from 8 to 17 g, respectively. The sinus node artery of the isolated right atrium or the anterior descending branch of the left coronary artery of the isolated left ventricle was cannulated and each preparation was perfused with heparinized blood from the carotid artery of the anesthetized support dog by means of a peristaltic pump (Harvard Apparatus, model 1210). A pneumatic resistance was placed in parallel with the perfusion system so that the perfusion pressure could be maintained constant at 100 mmHg. The venous effluent from the preparation was led to a collecting funnel and returned to the support dog through an external jugular vein.

The preparation was anchored to a stainless steel bar and placed in a cup-shaped glass container kept at 37°C. The upper part of the cardiac preparation was connected to

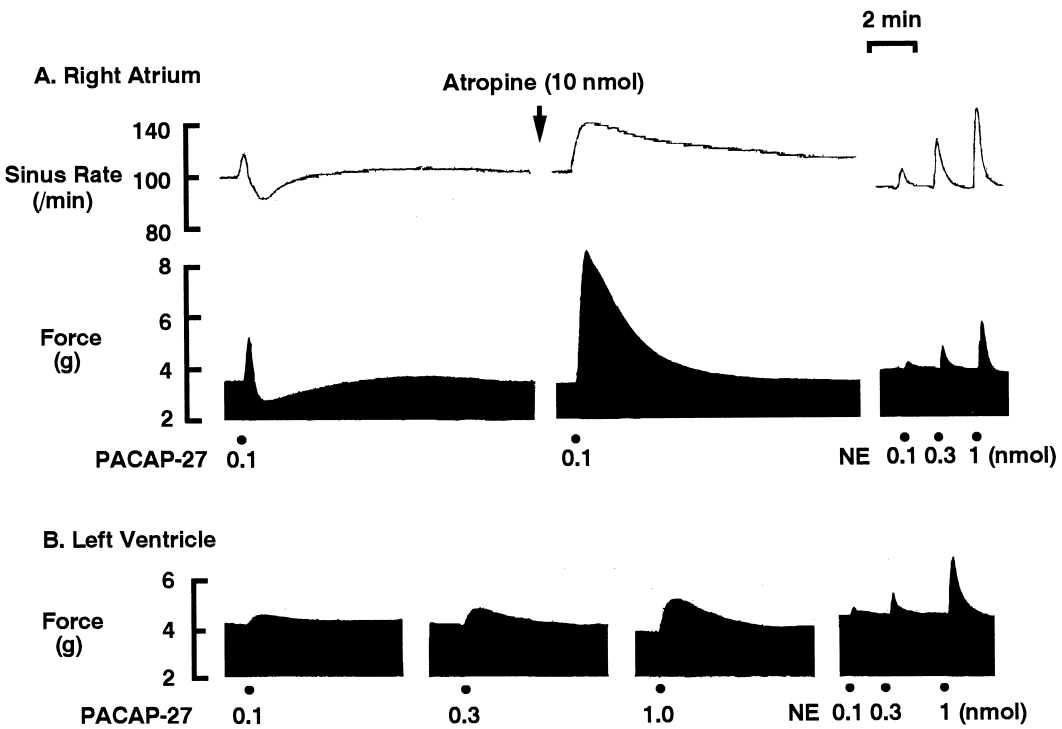

Fig. 1. (A) Effects of pituitary adenylate cyclase-activating polypeptide-27 (PACAP-27, 0.1 nmol) before and after atropine (10 nmol), and norepinephrine (NE, 0.1–1 nmol) on sinus rate and atrial contractility in an isolated, blood-perfused right atrium of the dog. (B) Effects of PACAP-27 (0.1–1 nmol) and norepinephrine (0.1–1 nmol) on ventricular contractility in an isolated, blood-perfused left ventricle of the dog.

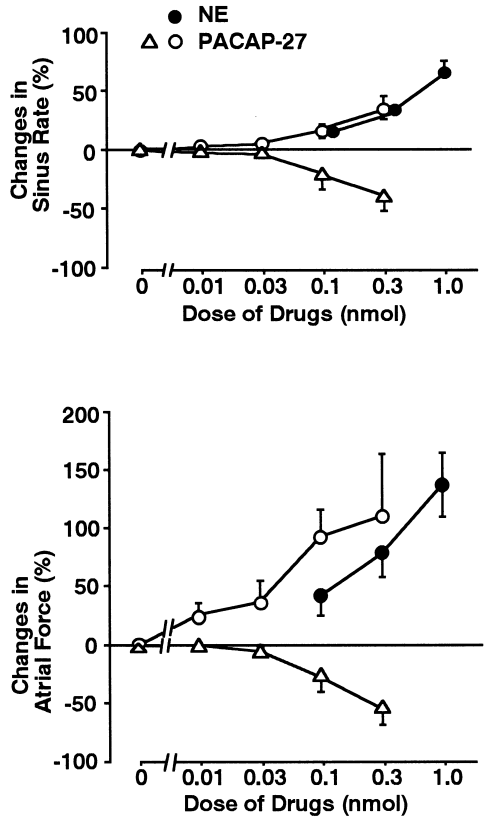

Fig. 2. The dose–response curves for mean % changes in sinus rate and atrial contractile force in response to pituitary adenylate cyclase-activating polypeptide-27 (PACAP-27, 0.01–0.3 nmol, ○ △) and norepinephrine (NE, 0.1–1 nmol, ●) in six isolated blood-perfused right atria. Vertical bars show S.E. The basal sinus rate and atrial contractile force in six atria were 104 ± 4.1 beats/min and 3.3 ± 0.5 g, respectively.

a force-displacement transducer (Nihon Kohden, Tokyo, Japan, AP-620G) by a silk thread. The cardiac tissue was usually stretched to a resting tension of 2 g. Isometric tension was recorded on a thermo-writing rectigraph (Nihon Kohden, RTA-1200). A pair of bipolar silver electrodes was brought into contact with the epicardial surface of the isolated preparation in order to record the atrial electrogram or to drive the left ventricle electrically. The left ventricular preparation was electrically paced at a frequency of 2 Hz with a 1-ms pulse duration above the threshold voltage (usually 4 V) by an electrical stimulator (Nihon Kohden, SEN 7103). The atrial rate was derived from the electrogram with a cardiotachometer (Nihon Kohden, AT-600G). Another pair of electrodes was placed posteriorly on the fatty tissue of the caval margin of the atrium and was used to stimulate the intracardiac parasympathetic nerve fibers (Furukawa et al., 1980).

2.2. Experimental protocols

We carried out four series of experiments after the preparations had stabilized for 30 min. In the first series, we studied the changes in sinus rate and atrial contractile force in response to PACAP-27 (0.01–0.3 nmol, $n = 6$) and norepinephrine (0.1–1 nmol, $n = 6$) in the isolated right atrial preparations. Additionally, we examined the effects of PACAP-27 (0.1 and 0.3 nmol, $n = 4$) on the SA nodal pacemaker activity and atrial contractile force after atropine treatment (10 nmol). The chronotropic and inotropic responses to norepinephrine were characterized by comparison with the PACAP-27-induced responses.

In the second series, to examine whether neural elements participate in chronotropic and inotropic responses to PACAP-27 in isolated right atria, we studied the effects of tetrodotoxin (10 nmol, $n = 4$) on the chronotropic and inotropic responses to PACAP-27 (0.1 nmol), acetylcholine (0.3 nmol) and intracardiac parasympathetic nerve stimulation (10 Hz).

In the third series, we studied the effects of PACAP-27 (0.01–1 nmol, $n = 6$) and norepinephrine (0.03–1 nmol, $n = 6$) on the left ventricular contractile force in the isolated left ventricular preparation. The substances were injected into the anterior descending branch of the left coronary artery. Additionally, to determine whether the positive inotropic responses to PACAP-27 are mediated by an adrenergic mechanism, we also studied the effects of propranolol on the positive inotropic response to PACAP-27 (1 nmol) in three ventricular preparations. The response to PACAP-27 was determined 2 min after propranolol was administered.

Table 1
The positive cardiac response to PACAP-27 in isolated, blood-perfused right atrial and left ventricular preparations

Drug (nmol)	Sinus rate (% increase)	Atrial force (% increase)	Ventricular force (% increase)
PACAP-27			
0.1	24 ± 6.3	242 ± 63.1	34 ± 6.6
0.3	100 ± 24.0	318 ± 5.0	43 ± 8.2
Norepinephrine			
0.3	33 ± 4.8	79 ± 21.4	38 ± 6.8

Responses to pituitary adenylate cyclase-activating polypeptide-27 (PACAP-27) were determined in four atropine-treated atria or six non-treated ventricles. Responses to norepinephrine were determined in six atria or six ventricles. The basal sinus rate and atrial force in four atria were 101 ± 4.4 beats/min and 1.7 ± 0.4 g, respectively and were not significantly different from the sinus rate and atrial force recorded in the norepinephrine-injected atria. The basal ventricular force in six ventricles was 3.3 ± 0.7 g.

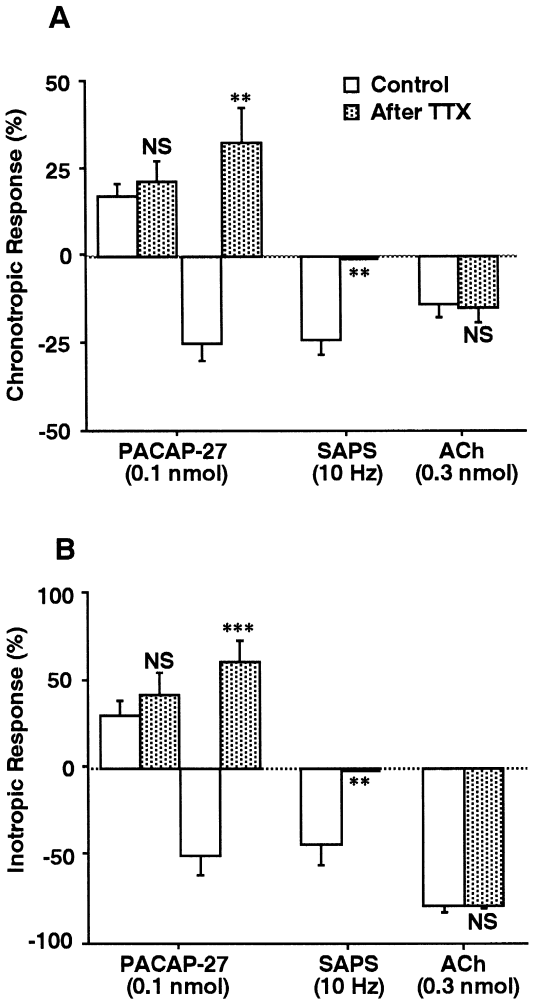

Fig. 3. The effects of tetrodotoxin (TTX, 10 nmol) on the positive followed by negative chronotropic (A) and inotropic (B) responses to pituitary adenylate cyclase-activating polypeptide-27 (PACAP-27, 0.1 nmol), and the negative chronotropic (A) and inotropic (B) responses to acetylcholine (ACh, 0.3 nmol) and intracardiac parasympathetic nerve stimulation (SAPS, 10 Hz) for 30 s in four isolated, blood-perfused right atria. Open and dotted columns show the response to intervention before and after treatment with TTX, respectively. Vertical bars show S.E. The basal sinus rate and atrial contractile force in four atria were 112 ± 7.6 beats/min and 3.9 ± 0.4 g, respectively. $** P < 0.01$; $*** P < 0.001$; NS, not significant vs. control.

In the fourth series, to determine whether PACAP receptors mediate the positive cardiac responses to PACAP-27, we studied the effects of PACAP-(6-27) (10 nmol), a type I PACAP receptor antagonist, on the positive cardiac responses to PACAP-27 (0.1 nmol) in four isolated right atria treated with atropine (30 nmol) and in three isolated left ventricles.

We studied the effects of a blocker on the chronotropic and inotropic responses to PACAP-27 1 h after the determination of the control responses to PACAP-27. The response to PACAP-27 was tested 2 and/or 60 min after the blocker was administered.

2.3. Drugs

Drugs were mixed fresh for each experiment. Pituitary adenylate cyclase-activating polypeptide-27 (Human) (PACAP-27, Peptide Institute, Osaka, Japan) and pituitary adenylate cyclase-activating polypeptide 6-27 (Human, Ovine, Rat) (PACAP-(6-27), Peninsula Laboratories, St. Helens, Merseyside, England) were dissolved in distilled water and kept frozen at -20°C as stock solutions, and diluted immediately before use. Acetylcholine chloride (Daiichi, Tokyo, Japan), atropine sulfate (Wako Pure Chemicals, Tokyo), norepinephrine hydrochloride (Sankyo, Tokyo), propranolol hydrochloride (Sigma, St Louis, MO, USA) and tetrodotoxin (Wako) were dissolved and diluted in 0.9% NaCl. Drugs were injected into the sinus node artery of the isolated atrium or the anterior descending branch of the left coronary artery of the isolated ventricle through a rubber tube by means of a microsyringe. The amount of drug solution injected was 0.01–0.03 ml over a period of 4 s.

2.4. Statistical analysis

All data are presented as % changes from the respective control and expressed as means $\pm$ S.E. An analysis of variance with Bonferroni's test was used for the statistical analysis of multiple comparisons of data. Student's *t*-test for unpaired data was used for comparison between two groups. Values of $P < 0.05$ were considered statistically significant.

3. Results

3.1. Effects of PACAP-27 on SA nodal pacemaker activity and atrial contractility

When PACAP-27 was injected into the sinus node artery of an isolated, blood-perfused right atrium, PACAP-

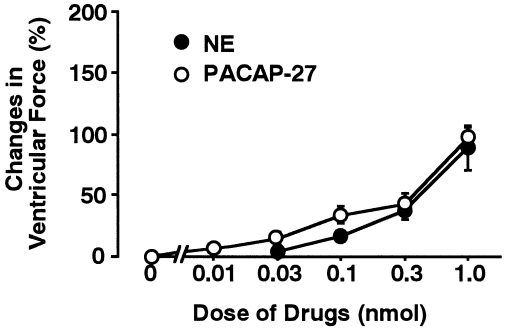

Fig. 4. The dose–response curves for mean % changes in ventricular contractile force in response to pituitary adenylate cyclase-activating polypeptide-27 (PACAP-27, 0.01–0.3 nmol, ○), norepinephrine (NE, 0.1–1 nmol, ●) in six isolated blood-perfused left ventricles. Vertical bars show S.E. The basal ventricular contractile force in six left ventricles was 3.3 ± 0.7 g.

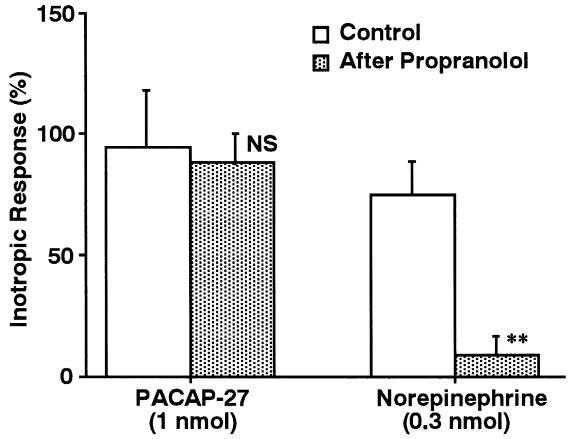

Fig. 5. The effects of propranolol (30 nmol) on the positive inotropic response to pituitary adenylate cyclase-activating polypeptide-27 (PACAP-27, 1 nmol) and norepinephrine (NE, 0.3 nmol) in three isolated, blood-perfused left ventricular preparations. Open and dotted columns show the response to intervention before and after treatment with propranolol, respectively. Vertical bars show S.E. The basal ventricular contractile force in three left ventricles was 2.6 ± 0.6 g. NS, not significant vs. control; $** P < 0.01$ vs. control.

27 produced an increase followed by a decrease in sinus rate and atrial contractility (Fig. 1A). Norepinephrine increased the sinus rate and atrial contractile force. Fig. 2 shows summarized data for the effects of PACAP-27 on six isolated right atrial preparations. PACAP-27 (0.01–0.3 nmol) increased the sinus rate ($P < 0.001$) and atrial contractile force ($P < 0.05$) in a dose-dependent manner. PACAP-27 also decreased both the sinus rate ($P < 0.01$) and atrial contractile force ($P < 0.001$) dose dependently. The threshold dose for the positive inotropic effect of PACAP-27 was 0.01–0.03 nmol. Norepinephrine (0.1–1 nmol) increased the sinus rate and atrial contractile force dose-dependently.

After atropine treatment, PACAP-27 caused only positive chronotropic and inotropic effects in the isolated-blood perfused dog atrium (Fig. 1A). Atropine (10 nmol) abolished the $11 \pm 2.9\%$ decrease in sinus rate and the $84 \pm 2.4\%$ decrease in atrial contractile force elicited by acetylcholine (1 nmol) in four right atria, and suppressed the negative chronotropic and inotropic responses to PACAP-27 at 0.1 or 0.3 nmol. After atropine treatment, PACAP-27 only increased the sinus rate and atrial contractile force in four isolated atria (Table 1).

3.2. Effects of tetrodotoxin on the responses to PACAP-27 in isolated right atria

To determine whether neural elements participate in the chronotropic and inotropic responses to PACAP-27, we examined the effect of tetrodotoxin on the cardiac responses to PACAP-27 in isolated right atria. Tetrodotoxin (10 nmol) blocked the negative chronotropic and inotropic responses to intracardiac parasympathetic nerve stimulation (10 Hz) but not to acetylcholine (0.3 nmol), and abolished the negative chronotropic ($P < 0.01$) and inotropic ($P < 0.001$) responses to PACAP-27 (0.1 nmol) in four isolated blood perfused dog atria (Fig. 3). That is, PACAP-27 caused only positive chronotropic and inotropic responses. Tetrodotoxin did not affect the initial positive cardiac responses to PACAP-27 when the re-

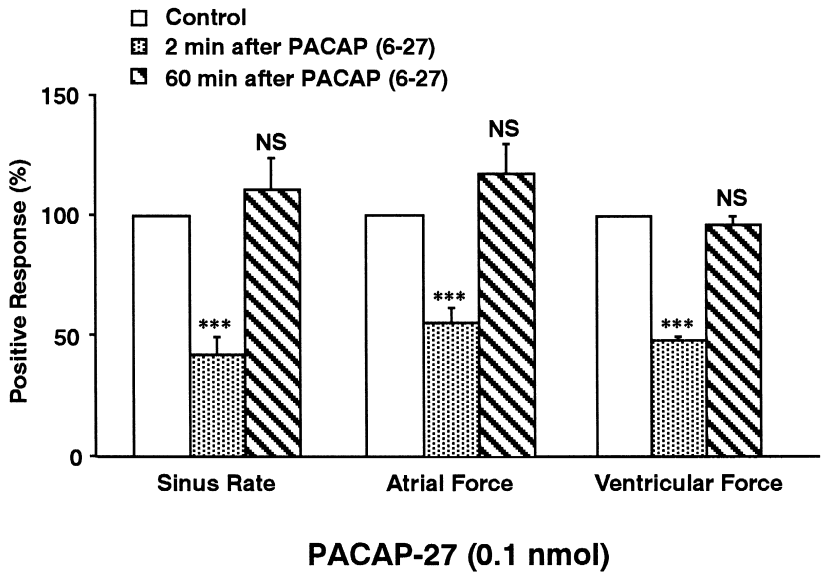

Fig. 6. The effects of pituitary adenylate cyclase-activating polypeptide-6-27 (PACAP-(6-27), 10 nmol) on the positive cardiac responses to PACAP-27 (0.1 nmol) in four isolated, blood-perfused atropine-treated right atria or three isolated, blood-perfused left ventricles. Mean increases in sinus rate, atrial force and ventricular force in response to PACAP-27 were 51 ± 12.8 beats/min, 2.6 ± 0.5 g and 2.5 ± 0.5 g, respectively in the control experiment. Vertical bars show S.E. The basal sinus rate and atrial contractile force in four atria were 117 ± 5.8 beats/min and 4.5 ± 1.5 g, respectively. The basal ventricular force in three left ventricles was 7.0 ± 1.0 g. NS, not significant vs. control; $*** P < 0.001$ vs. control.

sponse was determined at the same phase before and after tetrodotoxin treatment (Fig. 3).

3.3. Effects of PACAP-27 on ventricular contractility

When PACAP-27 (0.1, 0.3 and 1 nmol) was injected into the anterior descending branch of the left coronary artery of an isolated, blood-perfused left ventricle, it increased the ventricular contractile force dose dependently (Fig. 1B). Fig. 4 shows summarized data for the effects of PACAP-27 on six isolated left ventricular preparations. PACAP-27 (0.01–1 nmol) and norepinephrine (0.03–1 nmol) increased the ventricular contractile force in a dose-dependent manner ($P < 0.001$). The threshold dose for the inotropic effect of PACAP-27 was 0.01–0.03 nmol.

Additionally, we tested whether the positive inotropic response to PACAP-27 was mediated by β-adrenergic mechanisms in the isolated left ventricle. While propranolol (30 nmol) significantly ($P < 0.01$) inhibited the positive inotropic response to norepinephrine (0.3 nmol), it did not attenuate the positive response to PACAP-27 (1 nmol) in three isolated left ventricular preparations (Fig. 5).

3.4. Effects of PACAP-(6-27)

To determine whether PACAP receptors mediate the positive cardiac responses to PACAP-27, we studied the effects of PACAP-(6-27), a type I PACAP receptor antagonist, on the positive cardiac responses to PACAP-27 in isolated atropine-treated right atria and in non-treated left ventricles. In four isolated atria treated with atropine, PACAP-(6-27) at 10 nmol significantly ($P < 0.001$) attenuated the increases in sinus rate and atrial force in response to 0.1 nmol of PACAP-27 (Fig. 6). PACAP-(6-27) also reduced the increase in ventricular force in response to PACAP-27 in three left ventricles (Fig. 6). When PACAP-27 was tested again 60 min after PACAP-(6-27) treatment, it caused cardiac effects similar to those before PACAP-(6-27).

PACAP-(6-27) itself did not change the basal sinus rate and contractility.

4. Discussion

We demonstrated in the present study that PACAP-27 produced an increase followed by a decrease in sinus rate and atrial contractility in isolated right atrial preparations, whereas it dose dependently increased the ventricular contractile force in isolated left ventricular preparations (Figs. 1, 2 and 4). After atropine treatment, PACAP-27 caused only positive cardiac responses in the isolated right atrium and the positive inotropic response to PACAP-27 in the atrium was larger than that in the left ventricle (Table 1).

In this study, PACAP-27 produced the negative inotropic response in a dose-dependent manner (Fig. 2). Our previous study showed that PACAP-38 did not cause a negative inotropic response before physostigmine treatment. Therefore, our data indicate that PACAP-27 is more potent than PACAP-38 in decreasing the atrial contractility of the dog heart. Our present study demonstrated that PACAP-27 at a dose of 0.3 nmol increased the sinus rate and right atrial contractile force by 100 ± 24.0 and $318 \pm 5.0\%$, respectively, in the isolated, atropine-treated right atria and it was approximately 3- to 4-fold more potent than norepinephrine in augmenting pacemaker activity and contractility (Table 1). Our previous study showed that, after atropine, 0.3 nmol of PACAP-38 increased the sinus rate and atrial contractile force by approximately 10 and 70%, respectively, in the isolated right atria (Yonezawa et al., 1996). Additionally, Karasawa et al. (1990) demonstrated that VIP at the same dose increased the sinus rate and atrial contractile force by approximately 100 and 250% in the isolated right atria. In these three studies from our laboratory, the positive cardiac response to norepinephrine was similar in isolated blood-perfused right atrial preparations of dogs. Therefore, the potency order for the positive cardiac responses to PACAP-27, PACAP-38, VIP and norepinephrine is PACAP-27 $\geq$ VIP $>$ norepinephrine $>$ PACAP-38 in the dog atrium.

PACAP-27 and norepinephrine evoked similar positive inotropic effects in the isolated left ventricular preparations, whereas the positive inotropic responses to VIP and PACAP-38 were much smaller than those to norepinephrine, indicating that the potency order for the positive inotropic response to PACAP-27, PACAP-38, VIP and norepinephrine is PACAP-27 $\geq$ norepinephrine $>$ VIP $>$ PACAP-38 in the ventricle (Karasawa et al., 1990; Yonezawa et al., 1996).

The effects of PACAP-27 on atrial contractility were different from those on ventricular contractility in isolated dog hearts (Figs. 1, 2 and 4). PACAP-27 produced a negative inotropic effect in the atrium but not in the ventricle. Both atropine and tetrodotoxin blocked the negative cardiac responses to PACAP-27 in the isolated right atrium (Fig. 3 and Table 1). The regional differences in the negative effects of PACAP-27 are consistent with those in the cardiac parasympathetic nerve innervation (Levy and Martin, 1979). We have previously demonstrated that PACAP-38 produces negative chronotropic and inotropic responses by activating intracardiac postganglionic parasympathetic nerves in isolated dog atria (Yonezawa et al., 1996). Therefore, we suggest that PACAP causes the negative response by activating intracardiac parasympathetic nerves in the atrium.

The PACAP-27-induced positive inotropic effects were not different before and after propranolol in doses that suppressed norepinephrine-induced positive inotropic effects in left ventricular preparations (Fig. 5), indicating that the positive inotropic response to PACAP-27 is not mediated through β-adrenoceptors in the dog heart. Propranolol also did not attenuate the positive cardiac re-

sponse to PACAP-38 in isolated, blood-perfused dog atria (Yonezawa et al., 1996).

PACAP-(6-27), a specific antagonist of the type I PACAP receptor (Robberecht et al., 1991), attenuated the increases in sinus rate and atrial and ventricular contractility elicited by PACAP-27 to a similar extent (Fig. 6). In anesthetized dog hearts, PACAP-(6-27) inhibited the positive chronotropic response to PACAP-27, but VIP-(10-28), a VIP receptor antagonist, did not (Hirose et al., 1997). These results suggest that PACAP-27 increases atrial and ventricular contractile force as well as sinus rate mediated by type I PACAP receptors in the dog heart. PACAP-27 and PACAP-38 cause several effects that are mediated by three types of PACAP receptors (Harmar and Lutz, 1994). The type I PACAP receptor is subdivided into two subtypes on the basis of binding studies. $PACAP_{1A}$ receptors bind PACAP-27 with a slightly higher affinity than PACAP-38, while $PACAP_{1B}$ receptors bind PACAP-38 with high affinity and PACAP-27 with low affinity. The type I PACAP receptor is coupled to adenylate cyclase and phospholipase C (Spengler et al., 1993). The type II PACAP receptor is identical to the type I VIP receptor (Ishihara et al., 1992; Sreedharan et al., 1995). The type II PACAP receptor binds PACAP-27, PACAP-38 and VIP with similar affinity (Shivers et al., 1991) and is coupled to adenylate cyclase only (Ishihara et al., 1992; Spengler et al., 1993). The third type of receptor is identical to the type II VIP receptor (Lutz et al., 1993) and three neuropeptides activate the type II VIP receptor with the same rank order as for stimulation of cAMP production in the cAMP reporter LVIP cells (Usdin et al., 1994). Therefore, we suggest that the positive cardiac responses to PACAP-27 are mediated by the same subclass of type I PACAP receptors, probably $PACAP_{1A}$ or $PACAP_{1A}$-like receptors, in the dog heart.

After atropine treatment, the % increases in atrial contractile force in response to PACAP-27 were greater than those in ventricular contractile force (Table 1). The response to acetylcholine or VIP in the ventricle was much smaller than that in the atrium (Chiba et al., 1975; Karasawa et al., 1990). The density of muscarinic acetylcholine receptors in the atrium is greater than that in the ventricle in the rat (Linden et al., 1985), whereas β-adrenoceptor densities are almost equal in the atrium and ventricle (Brodde et al., 1982; Linden et al., 1985). The changes in the number of VIP receptors are also reported to correspond to changes in the contractility of the ventricular trabeculae from non-failing and failing human hearts (Hershberger et al., 1989). Additionally, PACAP-27 and -38 stimulate cAMP and inositol trisphosphate production with different potency in five splice valiants of type I PACAP receptors (Spengler et al., 1993). We suggest that the positive cardiac responses to PACAP-27 in the atrium and ventricle are mediated by the same type I PACAP receptors. Therefore, the regional differences in the efficacy of PACAP-27 to elicit a positive cardiac response might be due to differences in the density of type I PACAP-27 receptors between the atrium and ventricle and/or different post-receptor transduction between them.

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