EFFECTS OF SYNTHETIC HUMAN ATRIAL NATRIURETIC PEPTIDE ON THE HUMAN CORONARY CIRCULATION IN SUBJECTS WITH NORMAL CORONARY ARTERIES

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Atrial natriuretic peptide (ANP) is recognized as an "endogenous vasodilator". The purpose of this study was to determine the effects of a clinical therapeutic dose of synthetic alpha-human ANP on the coronary circulation in 15 subjects with normal coronary arteries and normal ventricular function. The epicardial coronary arterial diameter was measured by selective coronary arteriography. Coronary blood flow was estimated from the arterial cross-sectional area and the flow velocity determined using an subselective intracoronary Doppler catheter. ANP, 0.03 micrograms/min/kg given intravenously over 15 minutes, caused a dilation of the large epicardial coronary artery ($n=8$): the diameter of the proximal left anterior descending artery dilated from 2.6 ± 0.4 to 3.1 ± 0.5 mm ($p < 0.01$). Mean arterial pressure decreased from 89 ± 5 to 83 ± 5 mmHg ($p < 0.01$); heart rate did not change during ANP infusion. Estimated coronary blood flow significantly increased ($n=6$, $p < 0.01$), and thus the coronary vascular resistance decreased after ANP infusion, suggesting an ANP-induced dilation of resistance vessels.

The present study demonstrates that in human subjects a clinical dose of ANP by intravenous infusion dilates both the large epicardial and small resistance coronary vessels. These results suggest a potentially beneficial role for ANP in reducing the severity of myocardial ischemia in patients with ischemic heart disease.

SINCE the pioneering work by de Bold et al,¹ it has been recognized that atrial natriuretic peptide (ANP) is released from mammalian atria and circulates as a hormone.^{2,3} In addition to its crucial role in maintaining body fluids and electrolyte homeostasis,²⁻⁶ the peptide also has a potent vasorelaxant activity.⁷⁻⁹ Although there are some experimental studies in which ANP did not produce coronary vasorelaxation,¹⁰⁻¹²

isolated strips of a variety of vascular beds from various species and coronary artery rings from explanted hearts of patients responded to ANP by relaxation.⁷⁻⁹ Experimental studies performed in vivo have shown the dilation of coronary vessels after ANP administration.¹³⁻¹⁷ In conscious dogs, coronary blood flow and large epicardial coronary diameter increased after ANP administration into the coronary artery¹⁶ and the left atrium.¹⁷ Relaxation of vascular smooth muscles caused by ANP and by nitrate shares a common cellular pathway:

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TABLE I EFFECTS OF ANP ON HEMODYNAMIC VARIABLES (N=8)

	Before	After ANP infusion (0.03 micrograms/kg/min)
Mean AoP (mmHg)	89±5	83±5**
Heart Rate (bpm)	66±6	62±5
PAWP (mmHg)	7±3	5±4*
RAP (mmHg)	4±2	2±2*
Cardiac Output (l/min)	5.6±1.5	5.9±1.1
Rate-Pressure Products (mmHg×bpm)	6410±250	6210±200*

Data are mean±S.D.

AoP=aortic root pressure, bpm=beats/minute, PAWP=pulmonary artery wedge pressure, RAP=right atrial pressure.

p*: <0.05, *p*: <0.01 versus before ANP.

these drugs induce an increase in intracellular cGMP formation which, in turn, leads to a decrease in cytosolic free calcium concentrations^{9,29,31-32}. The vasodilating activities of those agonists are endothelium-independent.³³ However, ANP activates the particular guanylate cyclase while nitrate activates soluble guanylate cyclase.²⁹ Thus, ANP is recognized as "an endogenous vasodilator" released from the heart.

The effects of clinical doses of ANP on the coronary circulation in humans have not yet been rigorously studied. A recent report by Chu et al¹⁸ has demonstrated that an intravenous bolus administration of ANP (2.5 micrograms/kg) to patients caused a sustained dilation of the epicardial coronary artery with no increase in estimated coronary blood flow. It is necessary, however, to elucidate the effects of a clinical dose of ANP rather than a large pharmacological dose on coronary circulation in humans when one considers clinical application of the peptide.

In the present study, we examined the effects of a clinical dose of ANP (0.03 microgram/min/kg) on coronary circulation in human subjects with angiographically normal coronary arteries and normal ventricular function. We selected this dose since many studies have described the effects of a similar dose ANP on plasma concentrations, systemic hemodynamics, and the peripheral vasculature of humans.²²⁻²⁶

METHODS

Fifteen patients (10 men and 5 women, ages ranging from 38 to 56 years) undergoing

diagnostic coronary arteriography for chest pain were studied. All patients met the following criteria: 1) angiographically normal coronary arteries; 2) left ventricular ejection fraction of more than 60% by the contrast ventriculogram; and 3) no evidence of coronary vasospasm. Before each study, written informed consent was obtained from each patient after the explanation of the procedure.

Cardiac Catheterization and Quantitative Coronary Arteriography

Patients were brought to the cardiac catheterization laboratory in a fasting state. Diazepam (5 mg p.o.) was given for sedation. Cardiac medications such as beta-blockers, calcium blockers, long-acting nitrates, and converting enzyme inhibitors were withheld for at least 24 h before study. Right heart catheterization was performed with a 7F Swan-Ganz catheter. Arterial pressure was measured through a side tube of a 8F sheath catheter inserted into the femoral artery. Coronary arteriography was performed using a 8F Judkins coronary catheter with the non-ionic contrast agent, Iopamidol 370 (Nihon Shering, Osaka, Japan). Angiograms were recorded on 35 mm cinefilm at a speed of 30 frames/sec.

After selecting an appropriate projection that allowed visualization of the proximal segments of the left coronary arteries, the diameters of the coronary arterial segments were measured with a computer-assisted digital analysis system (MIPRON I, Kontron Instruments, FRG, ref. 19). An end-diastolic image was digitized and stored in a

Fig.1. Serial angiograms (end-diastolic frames) of the left coronary arteries in a right anterior oblique projection before (A) and after ANP infusion (B). The diameters of the large epicardial coronary arteries increased after ANP infusion.

512×512 matrix with an 8-bit gray scale. After arterial segments and a tip of a coronary catheter to be analyzed were identified, the edges of these structures were automatically detected by the computer, and absolute values of the arterial segments were determined by referencing the size of the coronary catheter. A 3–4 mm long arterial segment was identified and its mean diameter determined.

Estimation of Coronary Blood Flow

Coronary blood flow velocity was mea-

sured continuously using a 3F 20-MHz Doppler flow velocity catheter (Mikro-tip Doppler catheter DC-201, Millar Instruments, Houston, TX) connected to a Miller MDV-20 Doppler velocimeter^{20,21}. An 8F coronary guiding catheter was inserted into the femoral artery and positioned at the left coronary ostium. After intravenous administration of heparin 5000 unit, a 0.14 high torque floppy guide wire was introduced into the left anterior descending coronary artery (LAD) and a Doppler flow catheter was advanced into the mid-portion of the LAD over the guide wire. To obtain high-quality phasic signals of blood flow velocity, a Doppler catheter tip was usually placed at the center of the lumen and a range of Doppler sampling was adjusted.

To estimate relative changes in the volume-flow along the epicardial coronary artery, the velocity-area index (VAI) was calculated as:

$$\text{VAI (ml/min)} = \text{Mean Doppler Velocity} \times 3.14 \times (D/2)^2.$$

where D is the epicardial coronary diameter distal to the Doppler catheter tip.

Protocol

In the first 8 patients, baseline coronary arteriography of the left coronary artery was conducted for quantitative analysis. This was followed by saline or synthetic human alpha-ANP (Suntory INC., Division of Pharmaceuticals, Tokyo, Japan) infused intravenously at a rate of 0.03 micrograms/min/kg over 15 min. ANP was dissolved in sterile physiological saline at a concentration of 20 µg/ml and administered by an infusion pump. Arterial pressure, heart rate and right heart pressures were continuously measured. Thermodilution cardiac output was measured in duplicate before and after ANP infusion. Quantitative coronary angiography was repeated before and at the end of ANP infusion. Finally, intravenous nitroglycerin (TNG) was administered in a dose of 0.2 mg by bolus followed by 0.02 mg/minute and coronary angiograms were obtained 3 min later.

In the next 7 patients coronary blood flow velocity was continuously measured during ANP infusion using an intracoronary Doppler catheter, and changes in velocity-area

Fig.2. Summary of changes in the diameters of the LAD and LCX. Closed and open circles represent diameters of the proximal and distal arterial segments, respectively.

**: $p < 0.01$ versus baseline values.

ns: no significant difference between after ANP vs after ANP + TNG.

TABLE II EFFECTS OF ANP ON THE CORONARY VASCULAR BED (N=7)

	Before	After ANP infusion
Mean Aop (mmHg)	86 ± 6	$81 \pm 7^{**}$
Heart Rate (bpm)	70 ± 5	66 ± 4
LAD Diameter (mm)	2.8 ± 0.5	$3.2 \pm 0.4^{**}$
Mean CBFV (ml/min)	5.8 ± 0.1	5.9 ± 0.1
VAI (ml/min)	36 ± 2	$47 \pm 3^{**}$
Mean AoP/VAI (mmHg/ml/min)	2.3 ± 0.4	1.8 ± 0.3

Data are mean $\pm$ S.D.

VAI=velocity-area index, LAD=the left anterior descending coronary artery, CBFV=coronary blood flow velocity.

**: $p < 0.01$ versus baseline.

index were estimated. The quantitative coronary arteriography was performed before and 15 min after the initiation of ANP.

Statistical Analysis

Data were expressed as mean $\pm$ SE. Statistical analysis was done by Student's t-tests to compare 2 data points and by an analysis of variance followed by multiple comparison tests to compare more than 3 data points. A probability value of less than 0.05 was defined as a statistically significant difference.

RESULTS

Effects of ANP on Epicardial Coronary Artery Diameter

Intravenous administration of saline did not alter hemodynamic variables and coronary artery diameter. Intravenous infusion of ANP slightly but significantly decreased systemic arterial pressure, right atrial pressures and pulmonary wedge pressure (Table

I). A rate-pressure product decreased after ANP infusion. Heart rate and cardiac output unchanged after ANP infusion.

Representative angiograms before and after ANP infusion in a subject are shown in Fig. 1. Changes in coronary diameters in the first 8 patients are summarized in Fig. 2. ANP significantly increased the diameter of the proximal LAD from 2.6 ± 0.4 to 3.1 ± 0.5 mm ($p < 0.01$) and that of the proximal left circumflex coronary artery (LCX) from 2.3 ± 0.4 to 2.7 ± 0.4 mm ($p < 0.01$). The diameters of the distal segments of the LAD and LCX also increased after ANP infusion. However, there was no significant difference between the diameter after ANP infusion and that after infusion of ANP plus TNG.

Effects of ANP on Estimated Coronary Blood Flow and Resistance

Coronary flow velocity was not changed by ANP infusion (Table II). As the diameter of the LAD that contained a tip of the Doppler catheter significantly increased, the velocity-area index (VAI) consequently increased ($p < 0.01$), suggesting an increase in coronary blood flow in response to the ANP infusion. The ratio of mean arterial pressure to VAI decreased from 2.3 ± 0.4 to 1.8 ± 0.3 mmHg/ml/min ($p < 0.01$) by ANP infusion, suggesting a reduction in the coronary vascular resistance after ANP infusion (Table II).

DISCUSSION

The present study demonstrates that a clinical dose of intravenous ANP significantly dilates the large epicardial coronary artery in humans. The small resistance coronary vessels also appear to dilate following ANP infusion, because the estimated coronary vascular resistance decreased after ANP administration. The diameter of the large epicardial coronary artery after an ANP infusion was not statistically increased by an addition of TNG, suggesting that the dose of ANP used may be submaximal. The ANP-induced dilation of the large epicardial vessels may have been caused by its direct effect, but not have resulted from a flow-dependent dilation;^{27,28} because the coronary blood flow velocity did not significantly change during ANP infusion. We believe

that attempts to determine the effects of ANP on coronary vasculature in normal subjects are important, because the cardiovascular effects of ANP may be attenuated in pathologic states such as heart failure.²⁴⁻²⁶

The epicardial coronary arterial dilation observed in this study accords well with the studies by Herrmann et al^{34,35} and by Chu et al¹⁸. Herrmann et al³⁴ reported that intravenous administration of rat ANP (0.06-0.3 micrograms/kg/min) to patients with congestive heart failure resulted in a decrease in the coronary vascular resistance with no change in coronary blood flow (thermodilution methods). They also showed that intracoronary administration of rat ANP (67 ± 5 micrograms/min) to normal subjects increased the diameter of the large epicardial coronary artery by 26-27% with no direct myocardial effects.³⁵ Chu et al¹⁸ examined the effects of human ANP on the coronary vascular bed in patients and demonstrated that a bolus intravenous injection of ANP in a pharmacological dose (2.5 micrograms/kg) caused sustained dilation of the epicardial coronary artery lasting for 30 min; estimated coronary blood flow did not change within 5 min of the initiation of ANP. In the present study, we measured coronary arterial diameter and blood flow velocity during the period of ANP infusion over 15 min and showed that a clinical dose of ANP causes the dilation of the large epicardial and small resistance vessels. The present findings are in agreement with previous animal studies performed in conscious dogs^{16,17} which showed that administration of ANP into coronary arteries or the left atrium (0.5-5.0 μ g/kg) caused sustained dilation of the large epicardial vessels. Coronary blood flow significantly increased after ANP administration in those experimental studies.

The present finding of an ANP-induced reduction in the estimated coronary vascular resistance is at variance with studies in which vasomotion of the human forearm vascular bed in response to ANP was examined. A study from our institution²² and studies by Roy et al²³ and Ebert et al²⁵ failed to find a significant reduction in forearm vascular resistance during intravenous ANP infusion. Those studies used comparable doses of ANP to that used in the present study. An explanation for different responsiveness in

the coronary and forearm vascular beds to ANP is not readily available at the present time.

A limitation of the present study is that plasma ANP concentrations were not measured. However, a number of studies have indicated that doses similar to that used are safe and effective in increasing plasma ANP concentrations by several times the baseline level in healthy humans²³⁻²⁵. The ANP-induced decrease in arterial and right heart pressures with no change in heart rate observed in the present study are in agreement with previous studies^{6,22-26}.

The present findings indicate that ANP may have beneficial effects in the treatment of myocardial ischemia. The ANP-induced coronary vasodilation may reduce the severity of myocardial ischemia by reducing an imbalance between myocardial oxygen demand and supply, as does nitroglycerin. This possible beneficial action of ANP on myocardial ischemia could be potentiated by lack of reflex increase in sympathetic nerve activity during hypotension caused by ANP as compared to that during hypotension caused by nitroglycerin or nitroprusside^{22,30} because reflex sympathetic activation results in the increase in myocardial oxygen demand. In the present study, rate-pressure product (i.e. an index of myocardial oxygen demand) decreased by ANP infusion. Therefore, further studies need to be done to determine a clinical usefulness of the peptide in the treatment of myocardial ischemia.

In conclusion, the present study demonstrates that a clinical dose of ANP causes coronary vasodilation in normal subjects. The results suggest a potentially important role for ANP in the treatment of ischemic heart disease, although the assessment of its clinical application awaits further studies.

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