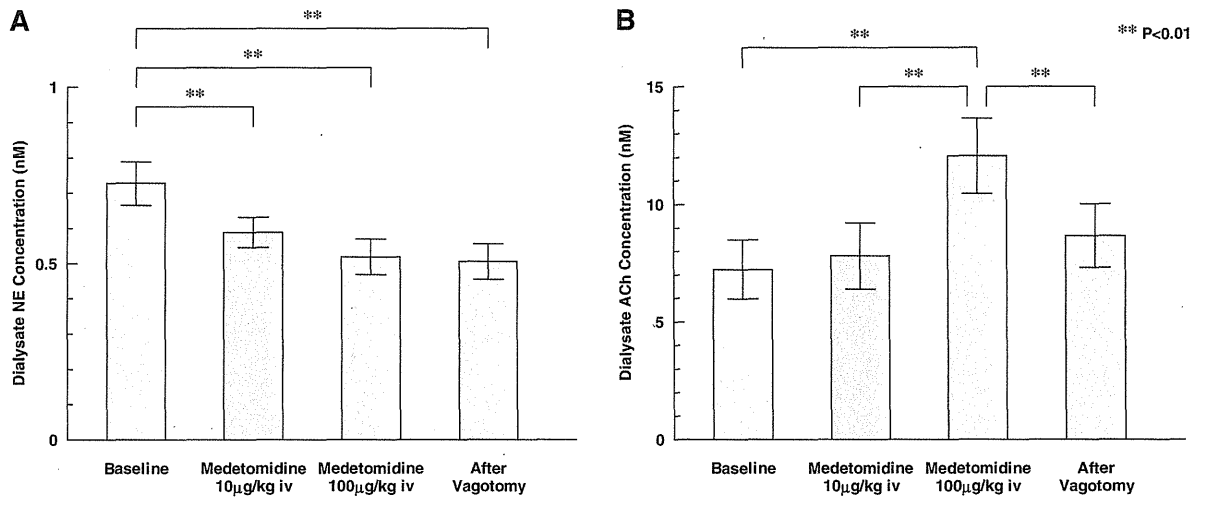

Figure 2. Effects of intravenous administration of low and high doses of medetomidine on dialysate NE (A) and ACh concentrations (B). Medetomidine at 10 and 100 μg/kg similarly decreased dialysate NE concentration (A). Medetomidine at 100 μg/kg significantly increased dialysate ACh concentration, but 10 μg/kg had no effect (B). Vagotomy suppressed the increase in dialysate ACh concentration. **P<0.01. ACh, acetylcholine; NE, norepinephrine.

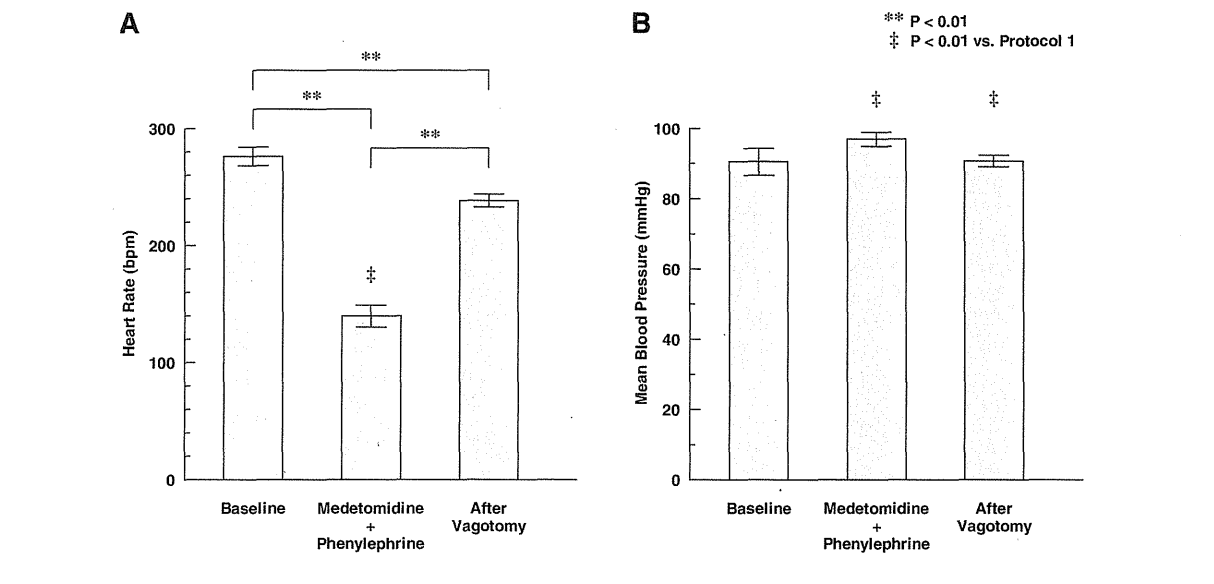

Figure 3. Effects of simultaneous intravenous administration of medetomidine and phenylephrine on heart rate (A) and mean blood pressure (B). Medetomidine combined with phenylephrine significantly decreased heart rate (A), and the decrease was significantly greater compared with medetomidine alone (protocol 1: Figure 1A). Medetomidine combined with phenylephrine maintained mean blood pressure (B) at the baseline level. Vagotomy partially restored the decrease in heart rate but had no effect on mean blood pressure. **P<0.01; ‡P<0.01 vs. medetomidine alone (protocol 1: Figure 1A).

markedly increased dialysate ACh concentration in the medetomidine-treated group (Figure 5B). The average slopes of the regression lines between mean BP and log dialysate ACh concentration were 0.0018±0.0004 in the control and 0.0062±0.0006 in the medetomidine-treated group. The slope was significantly steeper in the medetomidine-treated group than that in the control (P<0.01). However, the intercept did not differ significantly between the control (0.59±0.05) and medetomidine-treated (0.68±0.07) groups.

Protocol 4

The 20-Hz electrical stimulation of the right vagal nerve significantly decreased heart rate from 271±11 beats/min at the baseline to 112±6 beats/min and increased dialysate ACh concentration from 7.1±0.9 nmol/L at the baseline to 38.5±7.2 nmol/L (P<0.01). However, both 10 and 100 μg/kg of me-

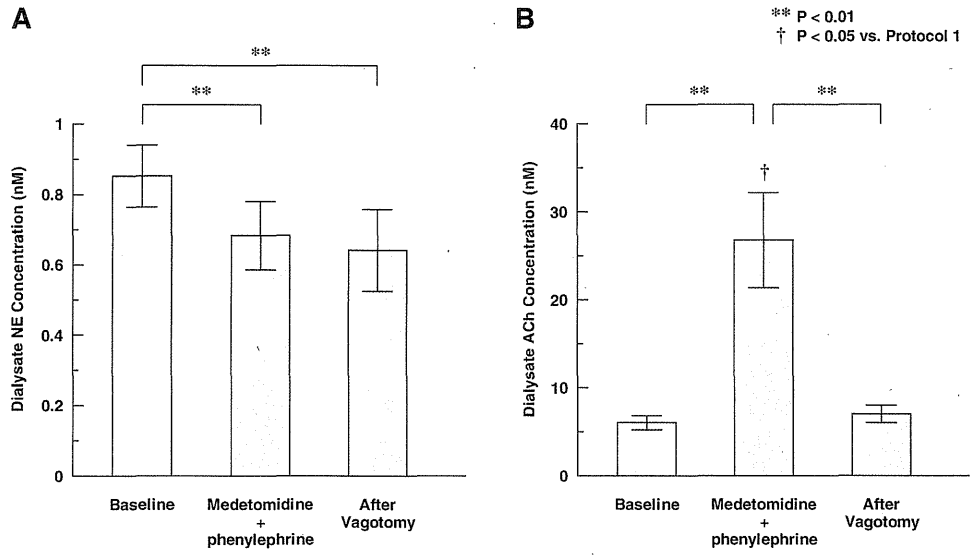

Figure 4. Effects of simultaneous intravenous administration of medetomidine and phenylephrine on dialysate NE (A) and ACh concentrations (B). Medetomidine combined with phenylephrine decreased dialysate NE concentration (A), and the decrease was similar to medetomidine alone (protocol 1: Figure 2A). Medetomidine combined with phenylephrine caused a marked increase in dialysate ACh concentration (B) and the increase was significantly greater than medetomidine alone (protocol 1: Figure 2B). Vagotomy suppressed the increase in dialysate ACh concentration. **P<0.01; †P<0.05 vs. medetomidine alone (protocol 1: Figure 2B). ACh, acetylcholine; NE, norepinephrine.

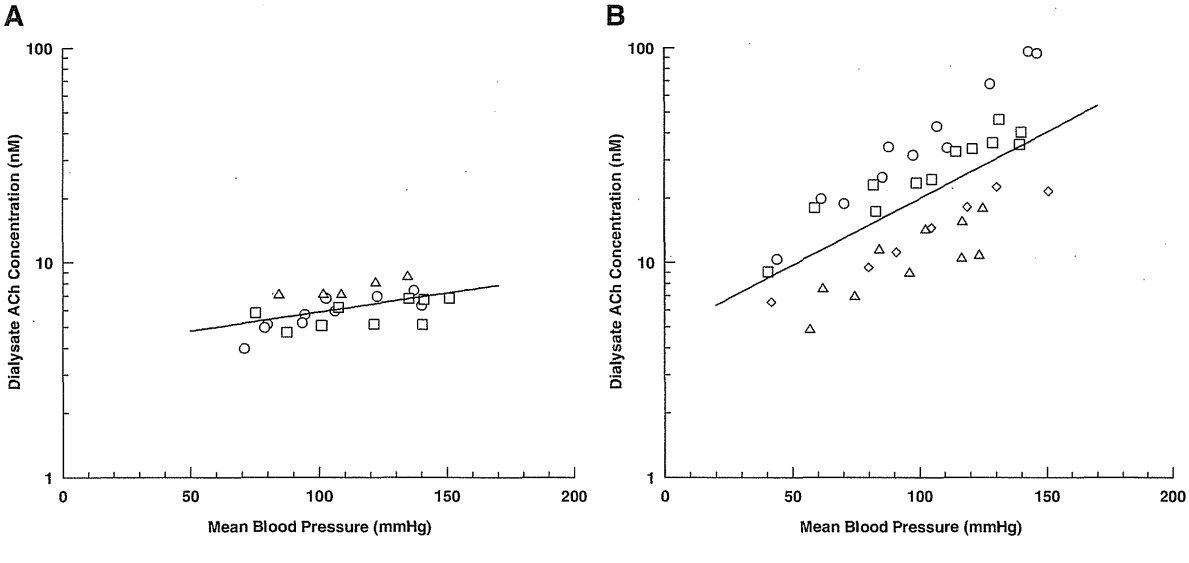

Figure 5. Regression lines of dialysate ACh concentration vs. MBP in control group (A) and medetomidine-treated group (B). In the control group (n=5), the increase in MBP had little effect on dialysate ACh concentration (A). In the medetomidine-treated group (n=7), dialysate ACh concentration was elevated with increase in MBP (B). Average regression line for control group: log[ACh]= 0.0018×MBP+0.59. Average regression line for medetomidine-treated group: log[ACh]=0.0062×MBP+0.68. Each symbol represents the data of 1 animal. ACh, acetylcholine; MBP, mean blood pressure.

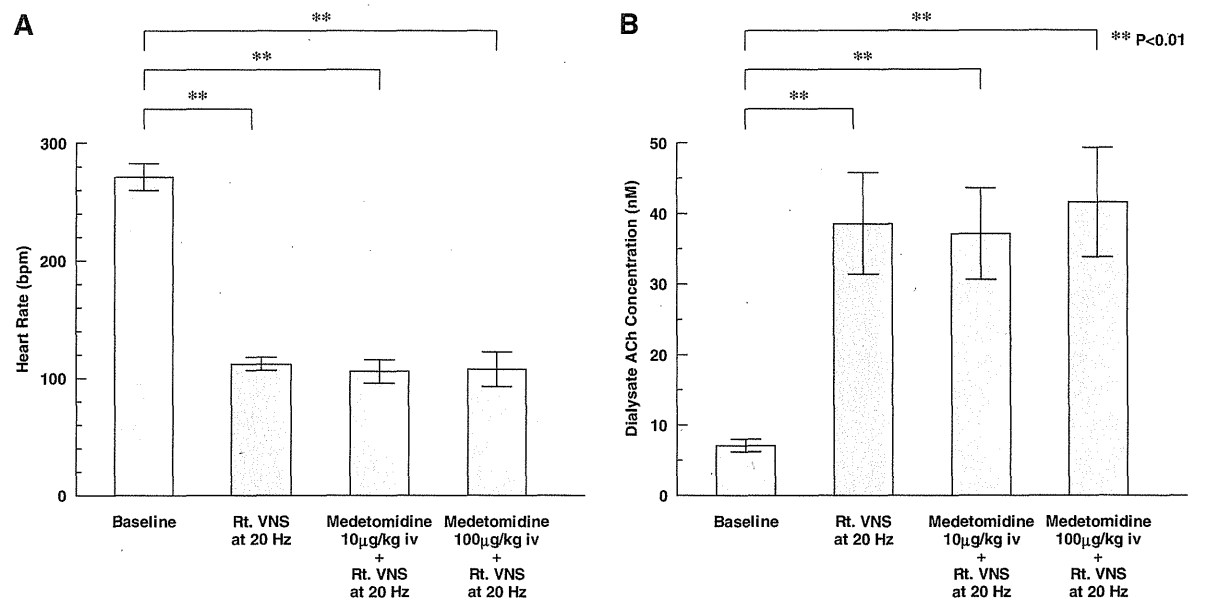

Figure 6. Peripheral effects of medetomidine on heart rate (A) and dialysate acetylcholine (ACh) concentration (B) under electrical stimulation of the right cervical vagal nerve. The 20-Hz electrical stimulation of the right cervical vagal nerve significantly decreased heart rate and increased dialysate ACh concentration. However, medetomidine did not affect the heart rate and dialysate ACh concentration under electrical vagal nerve stimulation (VNS).

detomidine did not affect heart rate and dialysate ACh concentration under the electrical stimulation (106±9.9 beats/min and 37.1±6.1 nmol/L at 10 μg/kg, 108±15 beats/min and 41.6±7.7 nmol/L at 100 μg/kg).

Discussion

We have elucidated the effects of medetomidine on cardiac sympathetic and vagal nerve activities simultaneously using cardiac microdialysis technique. Intravenous administration of 10 μg/kg of medetomidine significantly decreased sympathetic NE release to the SA node, while intravenous administration of 100 μg/kg of medetomidine significantly increased vagal ACh release to the SA node in addition to sympathetic suppression.

α_2-Adrenergic Agonist and Cardiac Sympathetic Nerve Activity

It is well-documented that α_2-adrenergic agonist suppresses sympathetic nerve activity.[15] Oku et al reported that dexmedetomidine suppressed renal sympathetic nerve discharge in baroreceptor-denervated rabbits.[16] In the present study, low-dose medetomidine decreased heart rate and mean BP through inhibiting sympathetic nerve activity, without affecting cardiac vagal nerve activity. High-dose medetomidine also suppressed NE release to the same level as low-dose medetomidine.

Several mechanisms may be involved in the sympathoinhibitory effect of α_2-adrenergic agonist. The rostral ventrolateral medulla has been reported to serve as an important site in mediating the hypotensive and sedative effects of α_2-adrenergic agonist.[17] McCallum et al reported that the central sympathoinhibitory effects of α_2-adrenoceptor stimulation are augmented by peripheral inhibition of ganglionic transmission.[18] The results obtained from protocol 1 indicate that low-dose medetomidine may induce a vagal-dominant condition through suppression of the cardiac sympathetic nerve without direct activation of the cardiac vagal nerve.

α_2-Adrenergic Agonist and Cardiac Vagal Nerve Activity

Kamibayashi et al reported that the vagus nerve played an important role in the antidysrhythmic effect of dexmedetomidine.[3] However, because it is difficult to selectively monitor cardiac vagal nerve activity, there is little direct evidence that α_2-adrenergic agonists can directly increase cardiac vagal nerve activity. In the present study, high-dose medetomidine significantly decreased heart rate and mean BP compared with low-dose medetomidine in protocol 1, and analyses of NE and ACh release by microdialysis technique proved that these decreases in heart rate and mean BP were associated with an increase in vagal ACh release to the heart. Histocytological studies demonstrated the presence of α_2-adrenergic receptors in the vagal dorsal motor nucleus and nucleus tractus solitarius.[19] Therefore, it is possible that α_2-adrenergic agonists directly activate the cardiac vagal nerve. It is also possible that intravenous medetomidine also modulates vagal ACh release through ganglionic transmission and the direct action to nerve endings. In protocol 4, however, medetomidine did not affect heart rate or the dialysate ACh concentration under electrical stimulation of the right efferent vagal nerve. Thus, in our experimental setting the peripheral effects of medetomidine on cardiac vagal nerve activity may be small compared with its central effects.

To exclude the possibility that medetomidine-induced hypotension affects local ACh concentrations, the mean BP was maintained constant by co-administration of phenylephrine in protocol 2. High-dose medetomidine combined with phenylephrine enhanced the decrease in heart rate and the increase in dialysate ACh concentration without medetomi-

dine-induced hypotension, indicating that hypotension occurring in protocol 1 had actually reduced ACh release in response to high-dose medetomidine. The results also suggest an interaction between baroreflex-induced and medetomidine-induced vagal nerve activation, which was extensively examined in protocol 3. In protocol 3, medetomidine steepened the slope of the regression line between mean BP and log dialysate ACh concentration, without affecting the intercept. In other words, medetomidine enhanced the baroreflex-induced ACh release from cardiac vagal nerve endings. Because the central pathway of baroreflex includes the vagal dorsal motor nucleus and nucleus tractus solitarius, in which α_2-adrenergic receptors have been demonstrated,[19] medetomidine may act on this pathway and modulate baroreflex-induced ACh release.

Clinical Implication

The selective α_2-adrenergic agonist, dexmedetomidine, is widely used for sedation in intensive care units. Bradycardia and hypotension are known to be unfavorable events during dexmedetomidine sedation.[20] Some cases of dexmedetomidine-induced atrioventricular block followed by cardiac arrest have been reported.[21,22] This critical complication may be associated with direct vagal activation by the α_2-adrenergic agonist. Compared with our previous results of electrical cervical vagal nerve stimulation in rabbits,[9] intravenous administration of 100 μg/kg of medetomidine had an effect equivalent to electrical vagal stimulation at 10 Hz. Furthermore, when the mean BP was maintained constant using phenylephrine, medetomidine had a stronger effect on cardiac vagal nerve activity, which is similar to 20-Hz electrical vagal stimulation, and this magnitude may sometimes cause atrioventricular block or sinus arrest.

Notwithstanding these adverse effects, vagal activation has several favorable cardioprotective effects. Our study proved that medetomidine, a selective α_2-adrenergic agonist, is a strong activator of cardiac vagal nerve. Vanoli et al[6] reported that vagal stimulation after acute ischemia can prevent ventricular fibrillation. Ando et al reported that efferent vagal nerve stimulation prevented ischemia-induced arrhythmias by preserving connexin 43 protein.[23] Our results suggest that vagal activation in addition to sympathetic suppression probably contributes to the antiarrhythmic effect of medetomidine.

Because inhibition of the sympathetic nerve system has been the cornerstone of drug therapy for heart failure,[24] a selective α_2-adrenergic agonist may be a potential therapeutic option for heart failure. Recent studies have shown that electrical vagal nerve stimulation also improves the outcomes in patients with heart failure.[25] Electrical stimulation of carotid baroreceptor has recently been reported to be a therapeutic option for heart failure. Sabbah et al reported that chronic electrical stimulation of the carotid sinus baroreflex improved left ventricular function and promoted reversal of ventricular remodeling in dogs with advanced heart failure.[26] Our study demonstrated that medetomidine modulates baroreflex control to enhance vagal nerve activity, which may also induce further cardioprotective effects.

Study Limitations

First, ACh is degraded by ACh esterase immediately after release. Therefore, detection of in vivo ACh release requires the addition of eserine, a specific ACh esterase inhibitor, into the perfusate. The presence of eserine around the semipermeable membrane might have affected ACh release in the vicinity of the semipermeable membrane. Eserine could have activated regulatory pathways such as autoinhibition of ACh release via muscarinic receptors.

Second, medetomidine is a chiral imidazole derivative. Thus, imidazoline receptors may also be involved in the cardiac vagal activation by medetomidine. Further investigation is necessary to clarify the influence of imidazoline receptors on cardiac vagal nerve activity. However, because an α_2-adrenergic antagonist, atipamezole, abolished the hemodynamic responses to medetomidine, we think that the cardiovascular effects of medetomidine are mainly related to the direct action of α_2-adrenergic receptors.

Third, the interactive effects between sympathetic and vagal nerve endings remain uncertain in the present study. Thus, we need further investigations including the open-loop approach where baroreceptor input pressure is strictly controlled.

Conclusion

A selective α_2-adrenergic agonist, medetomidine, directly activates cardiac vagal nerve and enhances the baroreflex control of vagal nerve activity. Medetomidine may be a therapeutic option for life-threatening arrhythmia or heart failure if the adverse effects are properly managed.

Acknowledgments

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Perfusion

Extracorporeal membrane oxygenation following pediatric cardiac surgery: development and outcomes from a single-center experience

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Abstract

Extracorporeal membrane oxygenation (ECMO) has emerged as an effective mechanical support following cardiac surgery with respiratory and cardiac failure. However, there are no clear indications for ECMO use after pediatric cardiac surgery. We retrospectively reviewed medical records of 76 pediatric patients [mean age, 10.8 months (0–86); mean weight, 5.16 kg (1.16–16.5)] with congenital heart disease who received ECMO following cardiac surgery between January 1997 and October 2010. Forty-five patients were treated with an aggressive ECMO approach (aggressive ECMO group, April 2005–October 2010) and 31 with a delayed ECMO approach (delayed ECMO group, January 1997–March 2005). Demographics, diagnosis, operative variables, ECMO indication, and duration of survivors and non-survivors were compared. Thirty-four patients (75.5%) were successfully weaned from ECMO in the aggressive ECMO group and 26 (57.7%) were discharged. Conversely, eight patients (25.8%) were successfully weaned from ECMO in the delayed ECMO group and two (6.5%) were discharged. Forty-five patients with shunted single ventricle physiology (aggressive: 29 patients, delayed: 16 patients) received ECMO, but only 15 (33.3%) survived and were discharged. The survival rate of the aggressive ECMO group was significantly better when compared with the delayed ECMO group ($p<0.01$). Also, ECMO duration was significantly shorter among the aggressive ECMO group survivors (96.5 ± 62.9 h, $p<0.01$). Thus, the aggressive ECMO approach is a superior strategy compared to the delayed ECMO approach in pediatric cardiac patients. The aggressive ECMO approach improved our outcomes of neonatal and pediatric ECMO.

Keywords

extracorporeal membrane oxygenation; congenital heart disease; cardiac surgery; pediatric; hypoplastic left heart syndrome

Introduction

Extracorporeal membrane oxygenation (ECMO) has emerged as an effective mechanical support following cardiac surgery with respiratory and cardiac failure. In 1976, Bartlett et al.[1] reported the successful use of ECMO for a neonatal patient with respiratory failure and, since then, ECMO has been used effectively for a variety of indications, including preoperative hemodynamic support, low cardiac output after cardiopulmonary bypass (CPB), sudden cardiac arrest, and as a bridge to heart transplantation[2]. The Extracorporeal Life Support Organization (ELSO) Registry reports that the rates of survival off ECMO and survival to discharge among neonatal cardiac patients are 59% and 39%, respectively; in pediatric patients, these values are 62% and 46%, respectively. The rates of survival off ECMO and to discharge among neonates subject to extracorporeal cardiopulmonary resuscitation are 63% and 37% and, in pediatric patients, these rates are 52% and 38%, respectively[3]. Outcomes of ECMO have been developing and still have been keeping the improvement scope for better outcomes, especially in patients with congenital heart disease.

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In our institute, we did not have any criteria for ECMO introduction until March 2005, but we changed our strategy of ECMO therapy and introduced an aggressive approach toward indications for ECMO from April 2005 wherein we did not hesitate to introduce ECMO. Before we had changed our strategy of ECMO therapy in April 2005, we had felt the negative image of ECMO because of our poor results of survival off ECMO in patients with congenital heart disease.

The purpose of this study was to evaluate the effects of this aggressive approach of ECMO initiation since we changed our strategy compared with the previously followed delayed approach for introducing ECMO after pediatric cardiac surgery, by reviewing our single center experiences.

Materials and methods

We retrospectively reviewed medical records of 76 pediatric patients with congenital heart disease who received ECMO following cardiac surgery between January 1997 and October 2010 at Okayama University Hospital, Japan.

Two groups were compared: those who were treated with the aggressive ECMO approach (45 patients from April 2005 to October 2010) and those who were treated with the delayed ECMO approach (31 patients from January 1997 to March 2005). The aggressive ECMO approach was defined as commencement of ECMO as early as possible before end-organ dysfunction or complete circulatory collapse, and without hesitation for the introduction of ECMO. The outcomes were categorized as follows: survived off ECMO (successful weaning from ECMO support), survival to hospital discharge, and survival during outpatient follow-up. Demographics, diagnosis, operative variables, and ECMO indication and duration of survivors and non-survivors were compared. We compared the lactate levels of both groups before and 2 hours after the introduction of ECMO.

We defined the indications for aggressive ECMO as follows: mean arterial blood pressure less than 35 mmHg, anuria, high lactate level (greater than 5.0 mmol/L) and acidosis with a pH less than 7.3, hypoxia with arterial oxygen saturation less than 60% with 100% FiO_2 (fraction of inspired oxygen) on mechanical ventilatory support. ECMO was established via a median sternotomy and cannulation of the ascending aorta or inominate artery for return, and drainage via a 3-mm polytetrafluoroethylene graft to the right atrium (in cases after a Norwood stage 1 procedure). For patients with a right ventricle (RV) to pulmonary artery (PA) shunt after a Norwood stage 1 procedure in hypoplastic left heart syndrome, we opened the RV-PA shunt and controlled the pulmonary blood flow by the degree of clipping the shunt.

Table 1. The demographic data of aggressive and delayed ECMO groups

Aggressive ECMO(n=45)			
Biventricle (n=16)		Single ventricle (n=29)	
Critical AS	4	HLHS	9
DORV	3	HLHS variant	7
TOF	3	SA/SV	5
PA/VSD	2	TA	3
TAPVC	2	DILV	2
CoA/VSD	1	PA/IVS	1
BEG syndrome	1	IAA	1
		Congenital MS	1
Delayed ECMO (n=31)			
Biventricle (n=15)		Single ventricle (n=16)	
DORV	3	HLHS	5
PA/VSD	3	HLHS variant	3
AVSD	2	PA/IVS	3
TOF	2	DILV	2
TAPVC	1	SA/SV	2
TGA	1	TA	1
Critical AS	1		
PTA	1		
Myocarditis	1		

AS, aortic stenosis; DORV, double-outlet right ventricle; TOF, tetralogy of Fallot; PA, pulmonary atresia; VSD, ventricular septal defect; TAPVC, total anomalous pulmonary venous connection; CoA, coarctation of the aorta; BWG, Bland-White-Garland; HLHS, hypoplastic left heart syndrome; SA, single atrium; SV, single ventricle; TA, tricuspid atresia; DILV, double-inlet left ventricle; PA, pulmonary atresia; IVS, intact ventricular septum; IAA, interrupted aortic arch; MS, mitral stenosis; AVSD, atrioventricular septal defect; TGA, transposition of the great arteries; PTA, persistent truncus arteriosus

We used a hollow-fiber membrane oxygenator (Biocube®; Nipro, Osaka, Japan) and a centrifugal pump (Gyro®; Kyocera, Kyoto, Japan) with a 6-mm heparin-coated tube (Biomate®; Toyobo, Osaka, Japan) for the ECMO device. The initial ECMO flow rate was set at 150–180 ml/kg/min. Anticoagulation was accomplished by drip infusion of sodium heparin to maintain an activated clotting time of 150–200 seconds. The hemoglobin concentration was maintained above 10 g/dL by transfusion of packed red blood cells during full support. We maintained the bladder temperature at 35°C and applied minimal ventilatory support during ECMO. The intrathoracic cavity was irrigated with sterile, warmed, normal saline every 3 days.

We tried to wean the patient off ECMO when the lactate level was less than 2.0 mmol/L, the urine output had increased to equal or greater than 1.0 ml/kg/h for as long as 24 h, and the arterial pulse pressure had increased to equal or greater than 10 mmHg. We started inotropic agents for 6 h and gradually reduced the ECMO flow rate

Table 2. Comparison of extracorporeal membrane oxygenation (ECMO) decannulation and hospital discharge ratio

	Aggressive ECMO (n=45)	Delayed ECMO (n=31)	*P*-value
Survived off ECMO	34 (75.5%)	8 (25.8%)	$P < 0.01$
Survived to Discharge	26 (57.7%)	2 (6.5%)	$P < 0.01$

Table 3. Comparison of patients with biventricular and single ventricle physiology

		Aggressive ECMO (n=45)	Delayed ECMO (n=31)	*P*-Value
Biventricle	Non-Survivors	5	13	$P<0.01$
	Survived to discharge	11	2	$P<0.01$
Singleventricle	Non-Survivors	14	16	$P<0.01$
	Survived to discharge	15	0	$P<0.01$

Table 4. Comparison of ECMO duration (mean ± s)

	Aggressive ECMO	Delayed ECMO	*P*-Value
Non-Survivors	105.8 ± 80.8 hours	292.1 ± 249.1 hours	$P=0.034$
Survived to Discharge	96.5 ± 62.9 hours	184.5 ± 166.1 hours	$P=0.032$

Table 5. Patient profiles

	Aggressive ECMO (n = 45)	Delayed ECMO (n = 31)	*P*-Value
Timing of introduction			
In OR	19 (42%)	11 (35%)	$P=0.341$
In ICU, average days after operation	3.64 days	7.88 days	$p<0.01$
Reason for indication			
LCOS	19 (42%)	8 (26%)	$P=0.032$
Cardiac Arrest	12 (26%)	10 (32%)	$P=0.159$
Hypoxia	7 (16%)	4 (13%)	$P=0.052$
Hypercapnia	2 (4%)	0	$P=0.116$
Failure to wean from CPB	5 (12%)	10 (32%)	$P=0.236$

This table shows the comparison between groups A and B in terms of time to introduction of ECMO and indication criteria for ECMO. OR: operating room; ICU: intensive care unit; LCOS: low cardiac output syndrome; n.s.: non-significant difference. Numbers inside ( ) indicate the number of patients.

Table 6. Lactate level of before and after ECMO

	Aggressive ECMO	Delayed ECMO	*P*-value
Before ECMO	5.5 ± 2.2 (mmol/L)	6.8 ± 3.1 (mmol/L)	$P=0.234$
After introduced ECMO	2.5 ± 1.2 (mmol/L)	2.9 ± 1.8 (mmol/L)	$P=0.435$

under stable hemodynamic conditions. We weaned the patient off ECMO within 12 h.

Descriptive statistics are expressed as mean ± standard deviation (*s*). The χ^2 test for a (2 × N) table, Kruskal-Wallis test and Student's t-test were used to evaluate differences between the groups for statistical significance. A p-value of <0.05 was considered to have statistical significance.

Results

The mean age of all patients was 10.8 months (range, 0 days–86 months) and the mean weight was 5.16 kg (1.16–16.5 kg). The demographic data of both aggressive ECMO and delayed ECMO are shown in Table 1. Cardiopulmonary bypass time (aggressive: 187.20 ± 117.48 min, delayed: 194.90 ± 115.33; p=0.400) and aortic cross-clamp time (aggressive: 79.93 ± 44.01 min, delayed: 64.62 ± 44.78 min; p=0.105) during surgery were not significantly different. Thirty-four patients (75.5%) from the aggressive ECMO group (n = 45) survived off ECMO and 26 patients (57.7%) survived to hospital discharge (Table 2). Eight patients (25.8%) from the delayed ECMO group (n = 31) survived off ECMO and 2 patients (6.5%) survived to hospital discharge (Table 2). Thirty-one patients (31/76, 40.8%) had biventricular physiology and 45 patients (45/76, 59.2%) single ventricle physiology. All the patients with single ventricle physiology treated with delayed ECMO died in the hospital (Table 3). The patients with biventricular physiology treated with the aggressive ECMO had significantly better results than the other groups. The patients who survived off ECMO and hospital discharge following the aggressive ECMO had significantly shorter ECMO duration than those following delayed ECMO (Table 4).

The timing of the introduction of ECMO is shown in Table 5. When ECMO was indicated for patients in the intensive care unit (ICU), the aggressive ECMO group had a significantly shorter duration of ECMO days after operation than the delayed ECMO group.

Our indication criteria for ECMO are shown in Table 5. In the aggressive ECMO group, low cardiac output syndrome was the significant reason for ECMO. In the delayed ECMO group, cardiac arrest and hypoxia were the two major reasons for ECMO.

The lactate levels before and after the introduction of ECMO are shown in Table 6. The lactate levels of both groups showed no significant difference.

Discussion

ECMO support following pediatric cardiac surgery provides effective support for postoperative cardiac and pulmonary dysfunction refractory to conventional medical management[4]. ECMO is an advanced therapy for acute cardiac and/or respiratory failure associated with congenital heart disease and pulmonary disease[5]. Indications for ECMO are affected by many factors, including ventricular function, magnitude of conventional inotropic support, and pulmonary function. No standard indication criteria or management guidelines have been established for ECMO in congenital heart disease because of its complex nature and specificity of use[6].

At our institution, in April 2005, the approach for ECMO was changed from a delayed ECMO approach, where patients were managed without ECMO for as long as possible, to an aggressive ECMO approach, where patients were indicated for ECMO as early as possible before a catastrophic event such as cardiac arrest occurred. In our results, the aggressive ECMO approach revealed a significantly better result in terms of ECMO survival and survival to hospital discharge than the delayed ECMO approach.

Ungeleider et al. have suggested that mechanical assistance should be routine after a Norwood stage 1 procedure and described an aggressive approach of mechanical assistance improving hospital survival rate[7]. Cooper et al. have suggested that the initiation of ECMO support should be based on "urgent" rather than "emergency" criteria, i.e., before the occurrence of end-organ dysfunction or circulatory collapse[8]. The risk factors for mortality due to ECMO are as follows: age below 1 month, male gender, long duration of mechanical ventilation support prior to introducing ECMO, and the development of renal or hepatic dysfunction during ECMO[6,9]. Following pediatric cardiac surgery, it is critical to maintain both systemic and pulmonary blood flow. The advantage of aggressive ECMO is the prevention of ventilator-induced lung injury caused by respiratory care injuries resulting from setting up mechanical ventilation.

Booth et al. demonstrated that the survival of cardiac patients supported by ECMO is associated with indication and cardiac diagnosis[2]. The ELSO Registry reported that rates of survival to weaning from ECMO and survival to hospital discharge were 59% and 39% in neonatal patients, and 62% and 46% in pediatric patients, respectively[3]. In our study, rates of survival to weaning from ECMO and survival to hospital discharge were 75.5% and 57.7%, respectively. Our aggressive ECMO approach was shown to be superior to that of the ELSO Registry average. Hence, the aggressive ECMO approach may result in better ECMO outcomes for patients with congenital heart disease than the delayed ECMO approach.

Patients with single ventricle physiology, especially those with hypoplastic left heart syndrome, had a significantly higher mortality rate than those with biventricular physiology[6,10]. In our review, the aggressive ECMO approach brings better outcomes, at least in patients with single ventricle physiology, than the delayed ECMO approach.

It is well known that long ECMO duration increases the risk of complications, such as bleeding, hemolysis, and systemic inflammatory syndrome. Long ECMO duration also increases the mortality rate associated with ECMO and may affect the progression of multiple organ dysfunction and have a negative influence on immunological systems[6,11-13]. A previous study of ours, as well as one by Baslaim et al., clearly indicated that patients with long ECMO duration (more than 3 days) may benefit less from ECMO support and may have an increased risk of mortality.[6,10]

In general, the purpose of pediatric ECMO as a mechanical circulatory support following cardiac surgery is recovery of cardiac function by unloading the right and left ventricle preloads and increasing the ECMO flow rate, as well as resting the lungs from the high demand for oxygen-saturated blood by the body. On the other hand, increasing the ECMO flow rate increases the left ventricular afterload and wall stress. These ECMO mismatches have a negative impact and outcome during pediatric ECMO following cardiac surgery. Hence, not only early introduction of ECMO, but also weaning as early as possible from ECMO might help to get better outcomes.

Several groups have reported improved survival in patients placed on ECMO in the operating room compared with those cannulated in the ICU[12,13]. Chaturvedi reported the avoidance of severe end-organ damage before restoration of adequate perfusion to pursue an aggressive approach for early indication of ECMO with the aim of reducing the morbidity and mortality associated with prolonged periods of hypoperfusion and cardiac arrest[13]. Our review also showed that it might be better to choose an aggressive ECMO approach of an early introduction of ECMO before progress to multiple organ dysfunctions could possibly prevent cardiac arrest, which results in a poor outcome. The ELSO Registry Report in 2009 showed a poor outcome for extracorporeal cardiopulmonary resuscitation after cardiac arrest, with survival rates of 26% in neonates, 47% in infants, and 39% in pediatric patients[3].

In conclusion, we recommend an aggressive ECMO approach following pediatric cardiac surgery, which requires early introduction and early discontinuation of ECMO support before end-organ dysfunction and circulatory collapse, rather than the conventional approach of delaying ECMO introduction. The aggressive ECMO

approach improved outcomes of ECMO therapy in our institution.

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Conflict of Interest Statement

I declare, on behalf of myself and all authors, the following: We have no material, financial, or other relationship with any healthcare-related business or other entity whose products or services may be discussed in, or directly affected in the marketplace, by this manuscript.

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Artificial
Organs

Atrionatriuretic Peptide Improves Left Ventricular Function After Myocardial Global Ischemia–Reperfusion in Hypoxic Hearts

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Abstract: Atrionatriuretic peptide (ANP) is reported to be useful for attenuating myocardial ischemia–reperfusion injury and improving left ventricular function after reperfusion. However, ANP may be either ineffectual or harmful in cases where the myocardium has been chronically hypoxic since birth. This can be a result of the concomitant high levels of cyclic guanosine monophosphate (cGMP) produced within the myocardium. This study aimed to verify the validity of using ANP to improve left ventricular function after myocardial ischemia–reperfusion injury. For this purpose, a cyanotic congenital disease model that was developed using isolated rat hearts was used. Hearts were obtained from Sprague-Dawley rats that were housed from birth until 6 weeks of age either in a hypoxic environment with 13–14% FiO_2 (hypoxic group) or in ambient air (normoxic group). These hearts were subjected to 30 min of normothermic global ischemia followed by 30 min of reperfusion using the Langendorff technique. Left ventricular functional recovery in hearts administered ANP (0.1 μM) into the reperfusion solution was compared with those hearts that were not administered ANP in both hypoxic (without ANP: $n = 6$, with ANP: $n = 6$, with ANP and HS-142-1[an antagonist of ANP]: $n = 6$) and normoxic hearts (without ANP: $n = 6$, with ANP: $n = 6$). In the hypoxic hearts, ANP administration improved the percent recovery of the left ventricular developed pressure (76.3 ± 9.2% without ANP vs. 86.9 ± 6.7% with ANP), maximum first derivative of the left ventricular pressure (82.4 ± 1.1% without ANP vs. 95.8 ± 6.5% with ANP), and heart rate (85.6 ± 4.7% without ANP vs. 96.1 ± 5.2% with ANP) after reperfusion. The improvement and recovery of these cardiac functions were closely related to significantly increased levels of postischemic cGMP release after ANP administration. The effect of ANP was blocked by HS-142-1. The improvements observed in the hypoxic group were similar to those found in the normoxic group. ANP administration during reperfusion improved left ventricular function after myocardial acute global ischemia–reperfusion equally in both the chronically hypoxic and age-matched normoxic groups. **Key Words:** Atrial natriuretic factor—Hypoxia—Myocardium—Reperfusion.

Hospital mortality rates for surgeries involving the repair of cyanotic congenital heart defects have decreased dramatically. However, acute cardiac failure during the postoperative period remains one of the leading causes of death among these patients (1). Ischemia–reperfusion injury is one of the causative factors of postoperative acute cardiac failure and involves damage to cardiomyocytes, vascular smooth muscle cells, and endothelial cells (2). Therefore, identifying drugs that attenuate acute myocardial ischemia reperfusion is important in improving the clinical outcomes of cardiac surgery.

Certain drugs are listed as clinical candidates for the prevention of myocardial ischemia–reperfusion injury during cardiac surgery. Nitric oxide (NO) donors have been found to improve left ventricular function after normothermic global ischemia in normal rabbit hearts. We previously reported that 0.1 μM atrionatriuretic peptide (ANP) administered

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during reperfusion following 30 min of normothermic global ischemia in an isolated normal rat heart model improves left ventricular function by increasing the rate of release of cyclic guanosine monophosphate (cGMP) (3).

However, there is a significant risk in concluding that the cardioprotective effects of these drugs are also useful to treat patients with chronic hypoxia because the myocardial response to these drugs in a chronically hypoxic heart may be different from those in a normoxic heart. In support of this, Baker et al. (4) reported that a NO donor does not improve left ventricular function after myocardial ischemia–reperfusion in rabbit hearts subjected to chronic hypoxia from birth; this indicates that NO donors do not exert a protective effect on chronically hypoxic hearts. Although the cause of the diminished cardioprotective effect is unknown, it may result from an overlapping mechanism of both the cardioprotective effect of a NO donor and chronic hypoxia. Chronic hypoxia has a cardioprotective effect against subsequent global ischemia (5,6); this effect is mediated by the activation of NO production and subsequent myocardial cGMP synthesis (4,6,7). Replicating the same cardioprotective effect may be of no benefit and may even be harmful. In contrast, the cardioprotective effect of ANP has a different mechanism from that of NO donors (8). However, it remains unclear whether ANP can additively improve left ventricular function after myocardial ischemia–reperfusion injury. Therefore, this study aimed to elucidate whether ANP improves left ventricular function after global myocardial ischemia–reperfusion in congenital hypoxic hearts.

MATERIALS AND METHODS

Congenital hypoxic rat heart model (hypoxia from birth)

Male Sprague-Dawley rats were used in this study. They were treated in compliance with the "Principles of Laboratory Animal Care," which was established by the National Society for Medical Research, and the "Guide for the Care and Use of Laboratory Animals," which was published by the US National Institutes of Health (NIH publication no. 85–23, revised 1996). This study was approved by the institutional ethics review board of Okayama University Hospital. To determine the effects of ANP on rat myocardium subjected to chronic hypoxia since birth, an original congenital cyanotic heart model was developed in rats. The rats of the hypoxic group were housed in a normobaric, hypoxic chamber from the time of birth until 6 weeks of age as described previously (9). The oxygen and carbon dioxide concentrations in the chamber were maintained at 13–14% and <0.4%, respectively, and continuously monitored using both a gas analyzer (ML206; ADInstruments, Sydney, Australia) and a commercially available software (PowerLab; ADInstruments). The hypoxic chamber humidity was maintained at <75%, and the temperature was maintained between 23 and 27°C with continuous hygrothermometer monitoring (TRH-7X; Shinyei, Kobe, Japan). To characterize the relationship between chronic hypoxia and ANP, a control group consisting of normoxic rats was housed in ambient air in the same environment for the same period; the effects of ANP administration were measured in these two groups. Both groups were kept in the same room under the same light–dark cycle. Rat chow and tap water were provided ad libitum.

Isolated perfused rat heart

The apparatus used for the perfusion of isolated rat hearts previously described by Hearse et al. (10) was modified and used for the experiments. This apparatus was designed to work in two interchangeable perfusion conditions: the unloaded and loaded modes. In the unloaded mode, the hearts were perfused through the aorta at a pressure of 80 cm H_2O and continued to beat independently. In the loaded mode, the hearts were perfused in the same fashion, but beat with external force; these hearts were not paced. The coronary effluent was discarded. The perfusate was a modified Krebs-Henseleit bicarbonate buffer (KHB: pH 7.4) containing 118.0 mM NaCl, 25.0 mM $NaHCO_3$, 2.5 mM $CaCl_2$, 1.2 mM $MgSO_4$, 4.7 mM KCl, 1.2 mM KH_2PO_4, and 11.0 mM glucose. The buffer was bubbled with 95% oxygen and 5% carbon dioxide gas at 38.0°C, to maintain the aortic partial pressure of oxygen >400 mm Hg: the buffer was filtered through a cellulose acetate membrane (pore size: 0.45 μm) to remove particulate contaminants (9).

The rats were anesthetized with an intraperitoneal injection of 50 mg/kg pentobarbital, and 100 IU/100 g body weight (BW) heparin was injected into the exposed right femoral vein. A blood sample was collected from the exposed left femoral vein to determine the hematocrit value (Htc) by using a blood gas analyzer (ABL 555: Radiometer, Copenhagen, Denmark). The heart was promptly harvested and then immersed in cold (4°C) heparinized KHB. The aorta was connected to the perfusion apparatus within 1 min of excision, and Langendorff perfusion was established. The pulmonary artery was incised to facilitate coronary drainage. The heart was then perfused in a retrograde manner under the perfusion

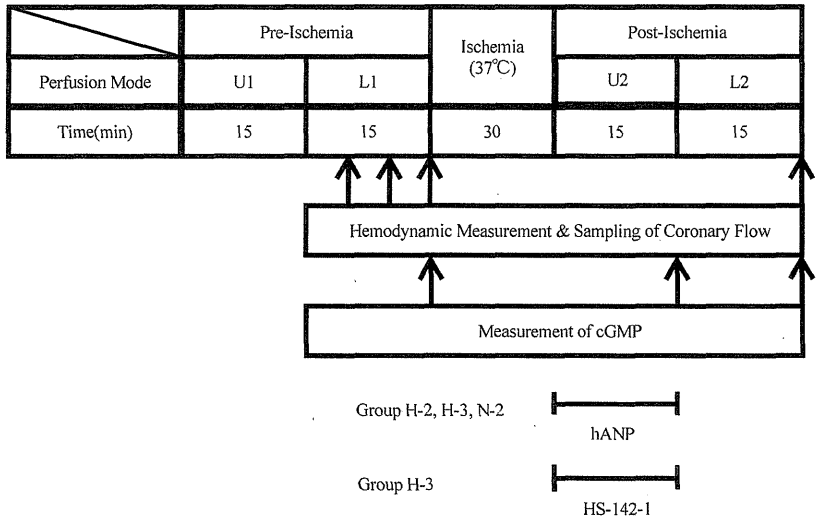

FIG. 1. Experimental protocol. cGMP, cyclic guanosine monophosphate; U, unloaded mode; L, loaded mode.

conditions of the unloaded mode at 38.0°C for 15 min. Changes in the heart rate (HR), left ventricular developed pressure (LVDP), and maximum first derivative of the left ventricular pressure (dP/dt max) were monitored in the loaded mode with PowerLab using an intraventricular balloon inserted through the mitral annulus and inflated with distilled water. During the pressure measurement, the left ventricular end-diastolic pressure was maintained at 4 mm Hg with the intraventricular balloon. The coronary flow (CF) was measured by direct collection of coronary effluent dripping from the heart for 1 min.

Study protocol

The experimental protocol is illustrated in Fig. 1. After an initial 15 min of washout (unloaded mode 1: U1), the hearts were perfused in the loaded mode for 15 min (loaded mode 1: L1). During this phase, HR, LVDP, dP/dt max, and CF were measured every 5 min, and the averages were recorded and assigned as the pre-ischemic indices. After 15 min of the loaded mode perfusion, the hearts were subjected to 30 min of global normothermic ischemia by clamping the aortic inflow line. Thirty minutes of global ischemia was selected because ischemia for periods exceeding 25 min induces sufficient myocardial damage (11). The sealed heart chamber temperature was monitored continuously during ischemia and maintained between 36.5 and 37.0°C. Hearts were reperfused for 15 min in the unloaded mode (unloaded mode 2: U2), followed by perfusion in the loaded mode for the next 15 min (loaded mode 2: L2). Postischemic cardiac function was evaluated at the end of this phase. The postischemic HR, LVDP, dP/dt max, and CF were calculated as percentages of their respective pre-ischemic values.

Thirty hearts were divided into five groups ($n = 6$ per group). In group H-1, the hearts of hypoxic rats were perfused without ANP. In group H-2, the hearts of hypoxic rats were perfused with 0.1 μM ANP (α-human atrionatriuretic peptide; Sankyo-Daiichi, Tokyo, Japan) during the U2 phase. In group H-3, the hearts of hypoxic rats were perfused with 0.1 μM ANP and 6 mg/L HS-142-1 (Kyowa Hakko Kogyo, Tokyo, Japan), a specific antagonist of the guanylyl cyclase ANP receptor during the U2 phase. In group N-1, the hearts of normoxic rats were perfused without ANP. In group N-2, the hearts of normoxic rats were perfused with 0.1 μM ANP during the U2 phase. In all groups, the release of cGMP into the CF was measured at the end of the L1, U2, and L2 phases.

The hearts were removed from the apparatus at the end of each experiment. The right ventricle (RV) and left ventricle (LV) were weighed separately. The interventricular septum was included in the LV. The ratio of wet RV/wet LV (RV/LV), wet RV/BW, and wet LV/BW were calculated. The hearts were heated to 70°C for 14 days and reweighed to determine the dry weight of the ventricular myocardium. The concentration of cGMP was determined by radioimmunoassay as described previously (12) and is expressed in picomoles per gram dry weight per min.

Exclusion criteria

Hearts presenting with HRs less than 250 beats/min after the first hemodynamic evaluation were excluded from the study because they were deemed

TABLE 1. *Characteristics of rats (final conditions and pre-ischemic values)*

Variable	Hypoxic groups			Normoxic groups	
	H-1 ($n = 6$)	H-2 ($n = 6$)	H-3 ($n = 6$)	N-1 ($n = 6$)	N-2 ($n = 6$)
BW (g)	154 ± 8*	167 ± 16**	164 ± 7***	207 ± 20	207 ± 12
Htc (%)	48 ± 3*	50 ± 7**	50 ± 2***	39 ± 3	39 ± 3
RV/LV	0.57 ± 0.11*	0.46 ± 0.04**	0.60 ± 0.08***	0.23 ± 0.01	0.24 ± 0.02
RV/BW (g/kg)	1.27 ± 0.33*	1.11 ± 0.28**	1.82 ± 0.29***	0.51 ± 0.05	0.50 ± 0.05
LV/BW (g/kg)	2.22 ± 0.23	2.38 ± 0.38	3.06 ± 0.11***	2.22 ± 0.31	2.08 ± 0.14
LVDP (mm Hg)	126 ± 7	121 ± 8	117 ± 5	122 ± 7	115 ± 9
dP/dt max (mm Hg/s)	3604 ± 187	3694 ± 570	3520 ± 286	4038 ± 341	3611 ± 223
HR (beat/min)	316 ± 13	291 ± 14	299 ± 24	312 ± 30	312 ± 22
CF (mL/min)	11.1 ± 3.3	9.6 ± 1.7	12.4 ± 1.6	7.6 ± 1.2	9.3 ± 2.3
cGMP (pmol/dry weight/min)	34.5 ± 13.0	38.7 ± 14.5	37.1 ± 12.6	15.3 ± 7.8	21.1 ± 20.4

Values are represented as mean ± standard deviation.
* $P < 0.05$, group H-1 vs. group N-1 and group H-1 vs. group N-2.
** $P < 0.05$, group H-2 vs. group N-1 and group H-2 vs. group N-2.
*** $P < 0.05$, group H-3 vs. group N-1 and group H-3 vs. group N-2.
BW, final body weight; CF, coronary flow; cGMP, cyclic guanosine monophosphate; dP/dt max, maximum first derivative of left ventricular pressure; HR, heart rate; Htc, hematocrit value; LV, left ventricular wet weight; LVDP, left ventricular developed pressure; RV, right ventricular wet weight.

to have sustained severe myocardial damage during the preparation.

Statistical analysis

All data are expressed as mean ± standard deviation. Statistical analysis was performed using commercially available software (SPSS Ver.19 for Windows; SPSS Japan, Tokyo, Japan). One-way analysis of variance (ANOVA) followed by Fisher's protected least significant difference test was used to compare data among the five groups. To determine the effect of ANP among the three hypoxic groups, one-way ANOVA followed by Scheffe's test was used. The interaction between ANP and chronic hypoxia with respect to myocardial ischemia–reperfusion injury was evaluated with a two-way ANOVA followed by Scheffe's test using the results of groups H-1, H-2, N-1, and N-2. A paired *t*-test was used to compare cGMP production in the CF of each group. A P value of <0.05 was regarded as statistically significant.

RESULTS

Pre-ischemic data

Table 1 shows the characteristics of the rats used in this study. The final BWs of the rats of the hypoxic groups were significantly lower than those of the normoxic groups. The pre-experimental Htc was significantly higher in the hypoxic groups than in the normoxic groups. The ratio of wet RV/wet LV was significantly higher in the hypoxic groups than in the normoxic groups. The wet RV/BW ratio was significantly higher in the hypoxic groups than in the normoxic groups. These results indicate that in the hypoxic groups, RV hypertrophy tends to be significantly more prevalent than LV hypertrophy. There were no statistically significant differences in HR, dP/dt max, and LVDP among the hypoxic groups. CF tended to increase in the hypoxic groups compared to the normoxic groups. The rate of cGMP drainage in CF tended to be greater in the hypoxic groups than in the normoxic groups. The average pre-ischemic cGMP drainage in CF was significantly larger in the hypoxic groups than in the normoxic groups ($P < 0.01$, 36.7 ± 12.7 vs. 20.3 ± 14.1 pmol/dry weight/min, respectively).

Postischemic cardiac functional recovery and changes in cGMP release rate

The postischemic recovery of LVDP, dP/dt max, HR, and CF are expressed as percentages of the pre-ischemic value and are listed in Table 2. The extent of the postischemic recovery of LVDP, dP/dt max, and HR was significantly better in group H-2 than in groups H-1 and H-3. There was no significant difference in the extent of cardiac function recovery between group H-1 and H-3. In the normoxic groups, the extent of postischemic recovery of LVDP, dP/dt max, and HR was greater in group N-2 than in group N-1. In the groups reperfused without ANP (group H-1 and N-1), the extent of postischemic recovery of LVDP, dP/dt max, and CF was significantly better in group H-1 than in group N-1.

Changes in the release of cGMP are shown in Fig. 2. The rate of cGMP release tended to be higher in the hypoxic rats than in the normoxic ones. In both the hypoxic and normoxic groups, the cGMP release occurring at the end of the U2 phase increased sig-

TABLE 2. *Postischemic recovery of left ventricular developed pressure, first derivative of left ventricular pressure, heart rate, and coronary flow*

	Hypoxic groups			Normoxic groups	
Variables	H-1 ($n=6$)	H-2 ($n=6$)	H-3 ($n=6$)	N-1 ($n=6$)	N-2 ($n=6$)
LVDP (%)	76.3 ± 9.2†	86.9 ± 6.7*	76.8 ± 4.2**	66.0 ± 11.9	77.1 ± 7.3***
dP/dt max (%)	82.4 ± 1.1†	95.8 ± 6.5*	82.8 ± 13.4**	64.0 ± 12.5	83.1 ± 8.3***
HR (%)	85.6 ± 4.7	96.1 ± 5.2*	92.7 ± 11.1**	86.3 ± 8.4	96.5 ± 5.0***
CF (%)	96.6 ± 25.7†	92.4 ± 20.9	81.5 ± 3.8	79.5 ± 8.3	89.1 ± 30.3

Values are represented as mean ± standard deviation.
* $P < 0.05$, group H-1 vs. group H-2.
** $P < 0.05$, group H-2 vs. group H-3.
*** $P < 0.05$, group N-1 vs. group N-2.
† $P < 0.05$, group H-1 vs. group N-1.
CF, coronary flow; cGMP, cyclic guanosine monophosphate; dP/dt max, maximum first derivative of left ventricular pressure; HR, heart rate; LVDP, left ventricular developed pressure.

nificantly with ANP administration in groups H-2 and N-2. The release of cGMP in both groups decreased to pre-ischemic levels after the end of the L2 phase. There was no difference in the rate of cGMP release between groups H-1 and H-3 at any time point.

Postischemic cardiac functional recovery was closely correlated with a significantly increased rate of cGMP release as a result of ANP administration; this cardioprotective effect was blocked by a specific antagonist of the guanylyl cyclase ANP receptor.

Interaction between the cardioprotective effects of ANP and chronic hypoxia

The interactions between the effects of ANP and chronic hypoxia on cardiac function recovery are shown in Fig. 3. ANP administration improved the recovery of the LVDP (10.6% improvement in hypoxic hearts vs. 11.1% in normoxic hearts), dP/dt max (13.4% vs. 19.1%), and HR (10.5% vs. 10.2%) after reperfusion of both hypoxic and normoxic hearts to the same degree. No interaction was identified between ANP and chronic hypoxia with respect to the recovery of LVDP ($P = 0.9270$), dP/dt max ($P = 0.4045$), HR ($P = 0.9583$), and CF ($P = 0.5030$) using the two-way ANOVA followed by Scheffe's test.

DISCUSSION

Effect of ANP on congenitally hypoxic heart

The present study provides the first direct evidence that ANP improves left ventricular function after myocardial acute global ischemia–reperfusion equally, in both chronically hypoxic and normal

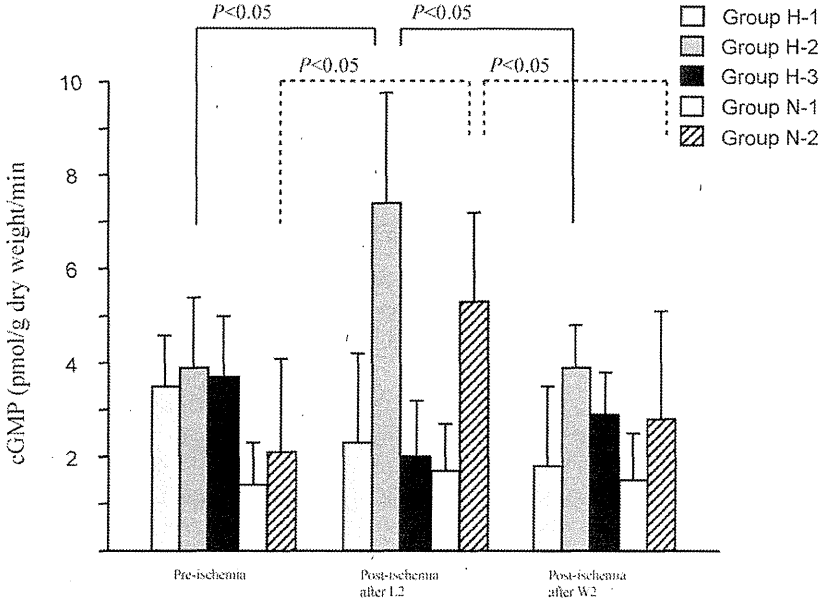

FIG. 2. Changes in cyclic guanosine monophosphate (cGMP) leakage. The rate of cGMP release tended to be higher in the hypoxic rats than in the normoxic rats. In groups H-2 and N-2 (both hypoxic and normoxic rats), cGMP was released at the end of the U2 phase and increased significantly with ANP administration. The release of cGMP in both groups decreased to the pre-ischemic levels after the end of the L2 phase. Bars represent standard deviation.

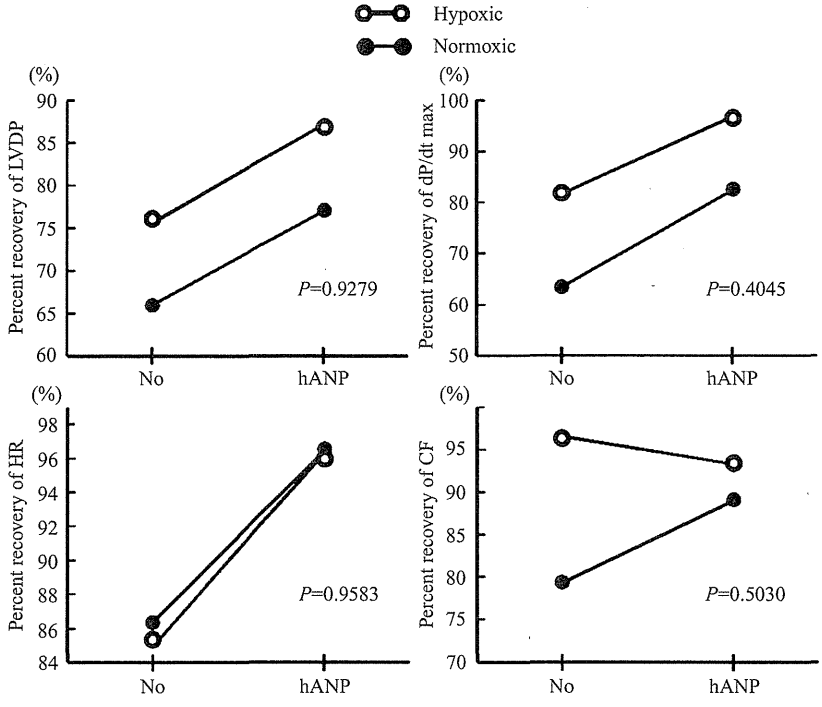

FIG. 3. Interactions between ANP and chronic hypoxia on left ventricular developed pressure, first derivative of left ventricular pressure, heart rate, and coronary flow. There was no interaction between ANP and chronic hypoxia from birth. LVDP, left ventricular developed pressure; dP/dt max, maximum first derivative of left ventricular pressure; HR, heart rate; CF, coronary flow; cGMP, cyclic guanosine monophosphate.

hearts. The increase in the release rate of cGMP following the initial 15 min of reperfusion and the lack of a cardioprotective effect in the presence of an ANP receptor blocker indicate that the protective effects of ANP against ischemia–reperfusion injury are partly mediated by the direct effect of cGMP on cardiomyocytes. ANP is known to activate a particulate guanylyl cyclase associated with the ANP receptor and increase the rate of intracellular cGMP synthesis (3). However, the relationship between the cGMP concentration in the reperfused myocardium and the extent of the protective effects against reperfusion injury is not known. It is suggested that overstimulation of cGMP synthesis could be detrimental to the reperfused myocardium and may result in increased apoptotic cell death (13). Padilla et al. (14) clearly demonstrated the negative effects of excessive cGMP in isolated rat hearts that received large doses of urodilatin (a member of the natriuretic peptide family) after 40 min of ischemia. Therefore, it is important to determine the doses of agents necessary to obtain the targeted increase in cGMP in the reperfused myocardium. In this study, 0.1 μM ANP was used because the previous study demonstrated that 0.1 μM ANP induces an increase in cGMP release without a significantly negative hemodynamic effect in normoxic rat hearts (3). The results of this study indicate that 0.1 μM ANP is an acceptable level even in chronically hypoxic hearts.

Interaction between ANP and chronic hypoxia

This study shows that ANP equally improves left ventricular function following an acute myocardial ischemia–reperfusion injury in chronically hypoxic and normal rat hearts. Our results are very different from those of Baker et al. who attempted to demonstrate that a NO donor exerts a cardioprotective effect against ischemia–reperfusion injury in chronically hypoxic hearts by using an isolated rabbit heart model. In that study, no improvement in left ventricular function after myocardial acute global ischemia–reperfusion was observed in rabbit hearts reperfused with the NO donor; this finding is in contrast with those in the rabbit hearts reperfused without NO donor (4).

The results of this study lead us to question why the cardioprotective effect of ANP was not diminished in hearts that were subjected to chronic hypoxia. Two potential reasons can be considered. First, the mechanisms activating cGMP synthesis may be different for ANP and chronic hypoxia. Hypoxia preconditioning is predominantly dependent upon increased NO synthesis; furthermore, increased NO levels directly activate soluble guanylyl cyclase. This mechanism for activating cGMP synthesis is the same as that used by the NO donor. On the other hand, ANP binds to the ANP receptor and indirectly activates the particulate guanylyl cyclase associated with the ANP receptor (8). Although both types of guanylyl cyclase activate

cGMP synthesis, the intramyocardial cGMP is known to be highly compartmentalized. Because these compartmentalized cGMPs often have different effects, chronic hypoxia (which involves soluble guanylyl cyclase-dependent cGMP synthesis) and ANP administration (which involves particulate guanylyl cyclase-dependent cGMP synthesis) may have different effects on the myocardium (8,15,16). Su et al. report that particulate guanylyl cyclase-dependent cGMP predominantly contributes to a decrease in intracellular Ca^{2+} levels, while soluble guanylyl cyclase-dependent cGMP predominantly contributes to a reduction in the sensitivity of myofilaments to Ca^{2+} (8).

ANP also appears to have an additional cardioprotective effect against myocardial ischemia–reperfusion injury. Another study provides evidence for the existence of an independently functioning and local renin–angiotensin system in the heart (17). In an isolated perfused rat heart, angiotensin II was found to exacerbate ischemia-induced ventricular fibrillation and to impair cardiodynamics; these effects were blocked by ANP (18). Morales et al. (19) report that directly blocking the local renin-angiotensin system with angiotensin II receptor antagonists relieves myocardial stunning after global ischemia. Therefore, the functional antagonism of angiotensin II may underlie the protective effect of ANP against ischemia–reperfusion injury. This cardioprotective effect of ANP, which is not related to cGMP, may therefore contribute to the further cardioprotective effects observed in the hypoxic hearts investigated in this study. Further study is needed to elucidate the full consequences of the interaction.

Study limitations

There are several limitations to the model system employed in this study. First, an isolated perfused preparation was used. Although the preparations were denervated, direct cardiac responses can be studied independent of the systemic effects of ANP. Second, a crystalloid solution was used in the perfusion circuit, which might have produced results different from those in a blood perfusion (20). The fact that various blood components play different roles during ischemia and reperfusion could have affected the results. Therefore, we used a simple crystalloid solution to simplify the study's model. Third, the animals were subjected to the protocol on the 1st day after birth. In contrast, children with cyanotic congenital heart disease are generally cyanotic since birth, having never been exposed to normoxia. Fourth, neonate animals were intermittently reoxygenated when they were exposed to air for feeding and maintenance. The effect of such brief normoxia has not yet been clarified. Finally, it should be noted that this study does not confirm that ANP is safe for use in clinical situations involving acute ischemia–reperfusion of chronic hypoxic hearts. The experimental conditions of this study have many differences from clinical situations. Further study is required to evaluate the validity of the findings for applications in patients with congenital hypoxia.

CONCLUSION

Atrionatriuretic peptide administration during the reperfusion of congenital hypoxic rat hearts following global ischemia improved the recovery of left ventricular function to the same degree as age-matched normal rat hearts. Although ANP and chronic hypoxia increased the rate of myocardial cGMP synthesis, there appears to be no interaction between ANP and chronic hypoxia with respect to their beneficial effects on left ventricular function following ischemia–reperfusion. These results indicate that ANP may have a cardioprotective pathway independent of its promotion of cGMP synthesis.

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Echocardiography

Transcatheter Closure of a Large Atrial Septal Defect under Microprobe Transesophageal Echocardiographic Guidance

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We present a case of an atrial septal defect (ASD) in a 59-year-old man with an indication for ASD closure who also had a history of chronic obstructive pulmonary disease. Because of his decreased respiratory function with multiple bullae in his lungs, the procedure was performed without general anesthesia under the guidance of fluoroscopy and two-dimensional (2D) transesophageal echocardiography (TEE) using a transesophageal echocardiographic microprobe (micro-TEE) (S8-3t; Philips Medical Systems, Andover, MA, USA). The micro-TEE probe was inserted into the esophagus smoothly and easily in the supine position without sedation. It revealed a deficient superior-anterior rim and adequate rims elsewhere, and the maximal diameter of ASD was measured to be 25 mm. Balloon sizing resulted in a stretched defect diameter of 29 mm using the stop-flow technique. A 30-mm AMPLATZER Septal Occluder (AGA Medical, Plymouth, MN, USA) was deployed. The micro-TEE demonstrated that both disks were on the appropriate sides of the interatrial septum and the device was not interfering with surround cardiac structures. Residual shunt flow was not detected with color Doppler. The device was released successfully without any complications. Recently introduced multiplane micro-TEE can provide adequate information about a large ASD with a less invasive procedure in adult patients. Micro-TEE has a potential to become a novel imaging option for interventions of the interatrial septum. (Echocardiography 2012;29:E94-E96)

Key words: transesophageal echocardiography, atrial septal defect, transcatheter closure device

A 59-year-old man with a history of chronic obstructive pulmonary disease who presented with progressive exertional dyspnea was found to have a secundum-type atrial septal defect (ASD) and a dilated right ventricle on transthoracic echocardiography (TTE). He was referred to our hospital for evaluation and transcatheter ASD closure.

Right heart catheterization demonstrated a pulmonary-to-systemic flow ratio of 1.8:1. His respiratory function was decreased due to multiple bullae in his lungs. And an intracardiac echocardiography was unavailable in our hospital. Therefore, transcatheter ASD closure was performed without general anesthesia under the guidance of fluoroscopy and two-dimensional (2D) transesophageal echocardiography (TEE) using a transesophageal echocardiographic microprobe (micro-TEE) (S8-3t; Philips Medical Systems, Andover, MA, USA) (Fig. 1). No sedatives were used, and local pharyngeal anesthesia was induced with oral liquid containing lignocaine. The micro-TEE probe was inserted into the

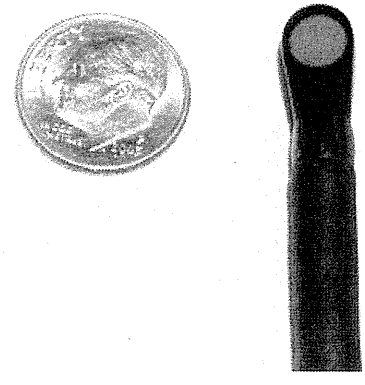

Figure 1. Miniaturized micro-TEE probe.

There are no disclosures about this report.

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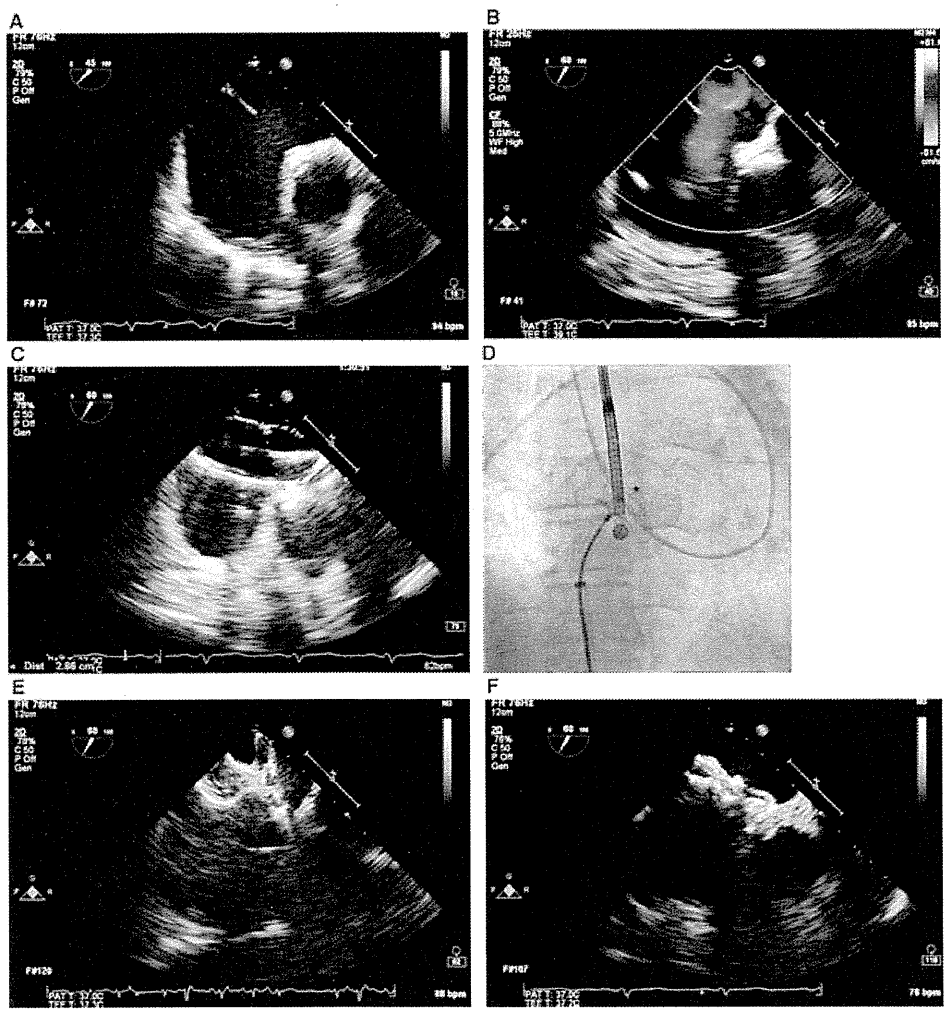

Figure 2. Transcatheter atrial septal defect closure under micro-TEE and fluoroscopic guidance. **A.** Micro-TEE demonstrates a large atrial septal defect (ASD) and a deficient superior-anterior rim. **B.** With color Doppler image, left-to-right shunt flow is recognized. **C.** Balloon sizing using the stop-flow technique **D.** Fluoroscopic view and **E.** micro-TEE view deploying a 30-mm AMPLATZER Septal Occluder **F.** Device is deployed successfully.

esophagus smoothly and easily in the supine position. It revealed a deficient superior-anterior rim and adequate rims elsewhere (Fig. 2A and B, movie clip S1), and the maximal diameter of ASD was measured to be 25 mm. Balloon sizing with a 34-mm AGA balloon (AGA Medical, Plymouth, MN, USA) resulted in a stretched defect diameter of 29 mm using the stop-flow technique (Fig. 2C). A 12-French AGA sheath was used to deliver the device. A 30-mm AMPLATZER Septal Occluder (AGA Medical, Plymouth, MN, USa) was deployed (Fig. 2D and E, movie clip S2). The micro-TEE clearly demonstrated that both disks were on the appropriate sides of the interatrial septum and the device was not interfering with surround cardiac structures. Residual shunt flow was not detected with color Doppler. The device was released successfully without any complications (Fig. 2F).

The extremely miniaturized multiplane micro-TEE has 18.5 mm of tip length, 7.5 mm of tip width, and 5.5 mm of tip height. The shaft size is 5.2 mm which is about a half size of the standard TEE probe for adults. The transducer consisted of 32 elements and has frequency from 3.2 MHz to 7.4 MHz. 2D, as well as M-mode, color Doppler, pulse-wave wave Doppler, and continuous-wave Doppler are available.

Echocardiography plays a pivotal role in guiding interventions of structural heart diseases. There are several imaging tools of echocardiographic guidance for structural heart interventions including 2D TTE, 2D TEE,[1] intracardiac echocardiography,[2,3] and recently introduced