Endothelin receptor subtype A blockade selectively reduces pulmonary pressure after cardiopulmonary bypass

Cassandra Joffs, MD
C. Allyson Walker, MS
Jennifer W. Hendrick, BS
David J. Fary, BS
Daniel K. Almany, BS
Jennifer N. Davis, DVM
Aron T. Goldberg, MD
Fred A. Crawford, Jr, MD
Francis G. Spinale, MD, PhD

Background: The bioactive peptide endothelin-1 is elevated during and after cardiopulmonary bypass and exerts cardiovascular effects through its 2 receptor subtypes, endothelin-1A and endothelin-1B. Increased endothelin-1A receptor stimulation after cardiopulmonary bypass can cause increased pulmonary vascular resistance and modulate myocardial contractility. However, whether and to what degree selective endothelin-1A blockade influences these parameters in the postbypass setting is not completely understood.

Objectives: Our objective was to measure left ventricular function and hemodynamics in a porcine model of cardiopulmonary bypass after selective blockade of endothelin-1A.

Methods: Adult pigs ($n = 23$) underwent 90 minutes of cardiopulmonary bypass and were randomized 30 minutes after bypass to receive a selective endothelin-1A antagonist (TBC 11251, 10 mg/kg; $n = 13$) or saline vehicle ($n = 10$).

Results: After bypass and before randomization, pulmonary vascular resistance rose nearly 4-fold, and left ventricular preload recruitable stroke work fell to one third of baseline values (both $P < .05$). In the vehicle group pulmonary vascular resistance continued to rise, and preload recruitable stroke work remained reduced. However, after endothelin-1A blockade, the rise in pulmonary vascular resistance was significantly blunted compared with that in the vehicle group. Moreover, the reduction in pulmonary vascular resistance with endothelin-1A blockade was achieved without a significant change in systemic perfusion pressures.

Conclusions: The present study demonstrated that increased activity of the endothelin-1A receptor likely contributes to alterations in pulmonary vascular resistance in the postbypass setting. Selective endothelin-1A blockade may provide a means to selectively decrease pulmonary vascular resistance without significant effects on systemic hemodynamics.

From the Division of Cardiothoracic Surgery, Medical University of South Carolina, Charleston, SC.

This work is supported by National Institutes of Health grants R01HL56603 and R01HL57952 and by an unrestricted grant from the Texas Biotechnology Corporation (San Diego, Calif).

Received for publication June 6, 2000; revisions requested Sept 8, 2000; revisions received Feb 2, 2001; accepted for publication Feb 5, 2001.

Address for reprints: Francis G. Spinale, MD, PhD, Division of Cardiothoracic Surgery, 114 Doughty St, Suite 625, Charleston, SC 29425.

J Thorac Cardiovasc Surg 2001;122:365-70

Copyright © 2001 by The American Association for Thoracic Surgery

0022-5223/2001 \$35.00 + 0 12/1/114938

doi:10.1067/mtc.2001.114938

Neurohormonal system activation is common after cardiopulmonary bypass (CPB).¹ For example, systemic levels of the potent bioactive peptide endothelin-1 (ET-1) increase 2-fold to 3-fold during and after CPB.^{2,3} The effects of ET-1 are mediated through 2 main receptor subtypes, endothelin-1A (ET_A) and endothelin-1B (ET_B).⁴ In vascular beds stimulation of ET_A

Figure 1. The percentage change in mean arterial pressure after infusion of 10 μ g of ET-1 in conscious pigs ($n = 5$). Infusion of the selective ET_A antagonist (10 mg/kg TBC 11251) caused a significant reduction in the ET pressor response. * $P < .05$ versus control values.

receptors results in contraction. In contrast, stimulation of endothelial ET_B receptors causes vasorelaxation by release of the vasodilators nitric oxide, epoprostenol (prostacyclin), or both.⁵ ET-1 has also been shown to affect myocardial contractility.^{1,6,7} ET-1 is normally present in the plasma at very low concentrations,⁸ but increased systemic levels of ET-1 occur in a number of disease states.⁸⁻¹² The increased ET-1 level in the post-CPB setting is associated with increased pulmonary arterial pressure and vascular resistance.¹³⁻¹⁵ The nonselective ET-1 receptor antagonist, bosentan, has been deployed in several cardiovascular disease states.^{16,17} For example, in both clinical and experimental models of pulmonary hypertension, nonselective ET-1 blockade with bosentan decreased systemic and pulmonary vascular resistance (PVR).¹⁷ The pulmonary vascular endothelium has a significant role in local regulation and maintenance of normal pulmonary vasomotor tone and in mediating the hypertensive response to CPB.^{3,18,19} Endothelial ET_B receptor stimulation induces vasorelaxation, particularly in the pulmonary bed.²⁰ Furthermore, the ET_B receptor is an important clearance mechanism for circulating ETs.²¹ Thus blockade of the ET_B receptor in the post-CPB setting may not be desirable, and selective ET_A receptor inhibition may be of greater utility. Accordingly, the goal of this study is to evaluate the effects of selective ET_A receptor antagonism after CPB on pulmonary and systemic hemodynamics.

Methods

Overview

For these studies, an intact porcine model of CPB with cardioplegic arrest was used.²² After baseline measurements under normothermic conditions, CPB with myocardial arrest using conventional hypothermic crystalloid cardioplegic solution was instituted. After reperfusion and separation from CPB, hemodynamics were assessed, and the pigs were randomized to either ET_A receptor inhibition or vehicle. Measurements were then performed for 90 minutes after separation from CPB. All animals were treated and cared for in accordance with the National Institutes of Health "Guide for the Care and Use of Laboratory Animals" (National Research Council, Washington, 1996).

Drug and Dose Selection

For these studies, the selective ET_A receptor antagonist TBC 11251 (Texas Biotechnology Corporation, Houston, Tex) was used. This compound has demonstrated high specificity for the ET_A receptor and is one of the most selective ET_A antagonists reported.²³ In an initial series of studies, chronically instrumented pigs were used to perform arterial pressure response studies, as described in previous studies.²⁴ In brief, 5 pigs (20 kg, Hambone Farms) chronically instrumented with an aortic access catheter received an intravenous injection of ET-1 (0.4 μ g/kg, Sigma Chemical Co, St Louis, Mo). Blood pressure was recorded at baseline and continuously for 20 minutes after ET-1 administration. After hemodynamics had returned to baseline values, TBC 11251 at 10 mg/kg was reconstituted to a standard volume of 5 mL by means of sterile saline solution and infused intravenously over a 10-minute period. Hemodynamic measurements were repeated at 1 to 20 minutes after injection. This dose of the ET_A antagonist did not significantly reduce resting arterial pressure. A second ET-1 challenge was administered, and blood pressure and heart rate were recorded continuously for at least 30 minutes after the challenge. Satisfactory ET_A receptor blockade was defined as an approximately 75% reduction in the ET-1-mediated increase in arterial pressure. The dose used in these studies resulted in significant inhibition of this ET-1-mediated blood pressure response, as shown in Figure 1. Thus an intravenous dose of 10 mg/kg was used in the CPB studies. This initial study used an exogenous delivery of purified ET-1 to define a dosing paradigm for the CPB study and does not necessarily reflect the actions or plasma levels of this peptide in vivo.

CPB: Instrumentation and Measurements

For the CPB studies, 23 pigs (45 kg) were instrumented to measure hemodynamics and left ventricular (LV) function.²² On the day of the study, the pigs were fully anesthetized, and arterial pressure monitoring was instituted. A multiluminal thermodilution catheter (7.5F, Baxter Healthcare Corp, Santa Ana, Calif) was then positioned in the pulmonary artery. The arterial line and pulmonary artery catheter were connected to externally calibrated transducers (Statham P23ID, Carolina Medical Inc, King, NC). A median sternotomy was performed, and the great vessels were isolated. A precalibrated microtipped transducer (7.5F; Millar Instruments Inc, Houston, Tex) was placed in the LV apex through a small stab wound and secured with a purse-string suture. A flow probe was placed over the anterior portion of the ascending aorta and connected to a digital flowmeter (HT107; Transonic Systems,

Figure 2. Schematic of the experimental design. After baseline measurements of LV function and hemodynamics, CPB with complete myocardial arrest was performed for 90 minutes. Measurements of LV function and hemodynamics were performed at 30 minutes after release of the aortic crossclamp and separation from CPB. At this time, randomization was performed in a blinded fashion in which pigs received the ET_A antagonist or vehicle. Measurements were then repeated for 60 and 90 minutes after CPB.

Inc, Ithaca, NY) for continuous measurement of LV stroke volume and cardiac output. The electrocardiographs, pressure waveforms, and flow-probe signals were recorded with a multichannel recorder (Hewlett-Packard, Palo Alto, Calif), as well as being digitized on a computer for subsequent analysis at a sampling frequency of 250 Hz (80386 processor; Zenith Data Systems, St Joseph, Mo). After collection of baseline hemodynamics and LV function, the pigs were anticoagulated with sodium heparin (300 U/kg) to achieve an extended activated clotting time of longer than 400 seconds (ACTII; Medtronic HemoTec, Inc, Englewood, Colo). Arterial cannulation was obtained, and then a 12-gauge catheter (DLP Inc, Grand Rapids, Mich) was placed at the root of the aorta for intermittent infusion of cardioplegic solution and use as an LV vent. Venous cannulation (34F; CR Bard Inc, Santa Ana, Calif) was then performed, and CPB was initiated. The CPB circuit contained a membrane oxygenator (Bentley Univox Spiral Gold, Bentley Laboratories, Irvine, Calif) and was driven by a modular roller pump system (Sarns 5000, 3M Healthcare, Ann Arbor, Mich). The aorta was crossclamped, and an oxygenated, hypothermic crystalloid cardioplegic solution (500 mL of Na⁺, 130 mmol/L Cl⁻, 109 mmol/L K⁺, 24 mmol/L Ca²⁺, 1.8 mmol/L, and 30 mEq/L HCO₃⁻ at 4°C) was delivered through the aortic root catheter. CPB was continued for 90 minutes with 500 mL of cardioplegic solution administered at 30-minute intervals after the initial cardioplegic dose. During CPB, neither systemic hypothermia nor topical cooling were used, and total flow was maintained within 2.5 to 3.0 L/min to maintain the mean systemic arterial pressure at a goal of 75 mm Hg. At completion of the cardioplegic arrest period, the crossclamp was removed, and the myocardium was reperfused. If ventricular fibrillation occurred during the initial reperfusion period, defibrillation was performed at 20 J with internal paddles. No inotropic or vasoactive agents were used at any time during the protocol, and protamine was not administered. At 30 minutes after crossclamp removal, LV function and systemic hemodynamics were recorded. After these measurements, the randomization scheme and experimental protocol were initiated.

Figure 3. PVR significantly increased after crossclamp removal and separation from CPB. In the vehicle group PVR increased in a time-dependent manner, with longer periods after separation from CPB. In marked contrast, PVR fell from 30-minute post-CPB values after infusion of the ET_A receptor antagonist. Sample sizes for each time point are shown in Table 1. **P* < .05 versus baseline values; +*P* < .05 versus 30 minutes after CPB; #*P* < .05 versus vehicle.

Randomization

At the 30-minute postbypass period, the 23 pigs were randomly assigned to receive either vehicle alone or the ET_A receptor antagonist, as shown in Figure 2. The randomization scheme was based on ear tag numbers, and the treatment assignment remained blinded to the investigators throughout the study. The cardiothoracic surgery veterinarian maintained the code list, prepared the compounds, and performed the infusion. The randomization codes were not broken until the completion of the study protocol. LV function and systemic hemodynamics were then measured at 60 and 90 minutes after separation from CPB.

Plasma Collection and Analysis

Plasma samples were obtained during the study for measurement of plasma ET-1 levels in the presence and absence of ET_A blockade. Baseline collection occurred immediately after placement of the arterial monitoring line. Blood samples were collected during CPB at 30, 60-, and 90-minute time points. The next sample collection occurred after aortic crossclamp release and a 30-minute stabilization period, just before randomization. Final samples were collected after randomization at 30 and 60 minutes after ET_A blockade or saline infusion. All blood samples were collected from arterial catheters placed after induction of anesthesia. Samples were collected in heparinized tubes and immediately centrifuged. Plasma was decanted and stored at -70°C until the time of assay. ET-1 levels were measured with a commercially available high-sensitivity radioimmunoassay kit (RIK-6901; Peninsula Laboratories, Inc, San Carlos, Calif) by using a previously described method.²⁵

Data Analysis

Changes in LV function and hemodynamics after CPB with cardioplegic arrest were initially compared with baseline values by means

Figure 4. Plasma ET-1 levels, expressed as percentage change from baseline, increased significantly after CPB. ET-1 levels continued to rise after infusion of the ET_A receptor antagonist, with significant difference from vehicle by 90 minutes after CPB. * $P < .05$ versus baseline values; + $P < .05$ versus 30 minute after CPB; # $P < .05$ versus vehicle.

of analysis of variance (ANOVA). Likewise, changes in plasma ET-1 concentrations before, during, and after CPB were compared with baseline values by using ANOVA. Specific comparisons between pre- and post-CPB values were performed with a Bonferroni adjusted t test. Comparisons between untreated and treated groups at the time points after CPB were examined with an ANOVA for repeated measures. All statistical analyses were performed with statistical software programs (BMDP Statistical Software Inc, Los Angeles, Calif). Results are presented as means $\pm$ SEM.

Results

All of the pigs entered into the protocol were successfully weaned from CPB and randomized at 30 minutes after bypass. Summary LV function and hemodynamic values are presented in Table 1. LV function and mean arterial pressure were reduced after cardioplegic arrest and CPB. Heart rate and pulmonary artery pressure were increased at 30 minutes after separation from CPB. In both the vehicle ($n = 10$) and ET_A blockade groups ($n = 13$), arterial pressure remained reduced throughout the study period. Mean pulmonary artery pressure was significantly lower in the ET_A receptor blockade group at 60 and 90 minutes after CPB. Systemic vascular resistance was reduced in the ET_A receptor blockade group when compared with that of the vehicle group but did not reach statistical significance ($P = .25$).

The change in PVR after separation from CPB is summarized in Figure 3. A significant increase in PVR occurred at 30 minutes after CPB. In the vehicle group PVR increased in a time-dependent manner after separation from CPB. In marked contrast infusion of the ET_A antagonist completely inhibited this time-dependent increase in PVR and allowed for a return toward baseline values.

The plasma ET-1 level at baseline was 6.0 ± 0.82 ng/mL, approximating reported normal values.⁸ Changes in plasma ET-1 levels are shown in Figure 4. Expressed as the percentage change from baseline, plasma ET-1 levels increased significantly from baseline at 30 minutes after CPB and continued to increase in the ET_A blockade group, with significant difference from levels in the vehicle group at 90 minutes after CPB.

Discussion

Neurohormonal system activation is a common sequelae of CPB.¹ Elevated circulating levels of the bioactive peptide ET-1 have been identified after CPB and persist into the early postoperative period.^{2,3} ET-1 has been shown to exert potent vasoconstrictive effects on the pulmonary vascular bed and to modulate myocardial contractility.^{6,7,10,20} The major findings of this study were 2-fold. First, circulating levels of ET-1 are elevated after competitive ET_A receptor blockade administered after CPB. Second, the introduction of selective ET_A receptor blockade significantly blunted the rise in PVR after CPB without significantly affecting systemic perfusion pressures.

The effects of ET-1 are mediated through 2 predominant receptor subtypes, ET_A and ET_B.^{4,10} The ET_A receptor is expressed mainly on vascular smooth muscle cells, where it activates a second messenger system, resulting in increased intracellular Ca^{2+} concentrations and contraction.²⁶ The ET_A receptor also appears to modulate the inotropic effect of ET-1.^{26,27} The ET_B receptor is found mainly on endothelial cells, where activated second messenger systems induce the release of nitric oxide and epoprostenol, with resultant vascular relaxation.^{4,5} The ET_B receptor is also a vital clearing mechanism for ET-1.²¹ Given the elevated levels of ET-1 after CPB, blockade of the potentially beneficial effects of the ET_B receptor may not be desirable. Consistent with past reports, this study demonstrated elevated circulating levels of ET-1 after CPB.^{2,3} This study also demonstrated significantly higher plasma ET-1 concentrations in the group receiving ET_A blockade. This is not unexpected because the drug used in the study is a competitive inhibitor, and elevated levels likely reflect displacement of ET-1 from ET_A receptors. This provides further evidence that the dose of ET_A antagonist used in the present study provides a pharmacologic effect at the level of the ET receptor system.

The underlying mechanisms for increased vasoconstriction within the pulmonary circuit in the early post-CPB setting are likely multifactorial. Acute ET_A receptor inhibition significantly blunted the rise in PVR after CPB without significantly affecting systemic perfusion pressures, thus providing direct evidence that ET_A receptor activation within the pulmonary circuit is a contributing factor. Right ventricular dysfunction is not uncommon after CPB,^{28,29} and

TABLE 1. Hemodynamics and cardiac performance after CPB and cardioplegic arrest: Effects of selective ET_A receptor blockade

	Time after CPB (min)					
	Baseline	30	60		90	
			Vehicle	ET _A B	Vehicle	ET _A B
Heart rate (beats/min)	86 ± 5	119 ± 6*	118 ± 6*	126 ± 6*	121 ± 5*	124 ± 6*
Cardiac output (L/min)	4.2 ± 0.2	3.5 ± 0.2*	3.5 ± 0.5	3.6 ± 0.3	3.2 ± 0.4*	3.9 ± 0.4
Stroke volume (mL/beat)	37.5 ± 2.1	18.8 ± 1.9*	23.4 ± 2.6*	16.2 ± 1.7*†	20.4 ± 2.8*	18.2 ± 2.5*
Stroke work (× 10 ⁵ dyne · cm)	48 ± 3	15 ± 2*	19 ± 2*	12 ± 2*†	17 ± 2*	14 ± 3*
Mean arterial pressure (mm Hg)	97 ± 3	58 ± 3*	62 ± 4*	55 ± 6*	62 ± 3*	56 ± 6*
LV end-diastolic pressure (mm Hg)	8.3 ± 0.7	10.1 ± 0.9	9.2 ± 1.4	8.8 ± 1.2	9.1 ± 1.6	9.1 ± 1.3
Mean pulmonary arterial pressure (mm Hg)	11.4 ± 0.8	19.2 ± 1.0*	23.1 ± 1.0*‡	16.7 ± 1.6*†	24.6 ± 1.2*†	15.8 ± 2.0*†‡
Systemic vascular resistance (dyne · s ⁻¹ · cm ⁻⁵)	1830 ± 120	1029 ± 84*	1240 ± 195*	977 ± 173*	1328 ± 237.2*	1075 ± 216*
Sample size (n)	23	23	10	13	10	13

ET_AB, Endothelin selective receptor blocker TBC 11251 (10 mg/kg).

**P* < .05 versus before CPB.

†*P* < .05 versus vehicle.

‡*P* < .05 versus 30 minutes after crossclamp release.

increased PVR can exacerbate this condition. Inhaled nitric oxide has been demonstrated to reduce PVR in patients after cardiac operations.^{30,31} ET_B receptor stimulation provides an intrinsic pathway of nitric oxide and epoprostenol production, contributing to physiologic vasodilation in the pulmonary circuit. In the present study the infusion of a selective, competitive ET_A antagonist resulted in increased circulating levels of ET-1. Increased plasma ET-1 in this setting could lead to enhanced ET_B receptor activation, with subsequent pulmonary vasodilation. Thus, the reduction seen in PVR with selective ET_A receptor blockade may occur through 2 complementary mechanisms: through the direct antagonism of ET_A receptor-mediated vasoconstriction and through the potentiation of ET_B receptor-mediated vasorelaxation.

In addition to effects in the pulmonary vascular bed, ET-1 has been shown to influence contractile function in the myocardium both in intact animal preparations and in isolated myocyte systems.^{6,7,9} Exposure of normal myocardial preparations to increasing concentrations of ET-1 has been shown to induce a mild positive inotropic effect.¹⁴ However, with underlying myocardial contractile dysfunction, increased ET-1 levels exert a negative inotropic effect.⁶ The effects of ET_A receptor blockade on indices of global LV ejection performance observed in the present study need to be considered in the context of the experimental design used versus the clinical CPB setting. Inotropic stimulation with calcium, β-adrenergic receptor antagonists, or both is commonly performed in the early post-CPB setting; however, in the present study no vasoactive or inotropic interventions were used because these would have confounding influences on the experimental design. Thus, whether and to what degree ET_A receptor blockade would influence myocardial contractile performance remains to be established.

Although a number of studies have documented increased ET-1 levels after CPB,^{2,3} the investigations with ET-1 blockade are limited, with the majority using nonselective ET-1 blockade or a converting enzyme inhibitor.³²⁻³⁴ Of the studies with selective ET_A antagonism, the primary focus has been animal models of congenital heart disease.^{35,36} For example, a recent study evaluated the effect of selective ET_A antagonism in a lamb model of experimentally induced high flow pulmonary hypertension.³⁵ The investigators were able to show that PVR was significantly lower after CPB in lambs pretreated with the selective antagonist. The present study builds on these reports by using a conventional model of CPB and specifically evaluating systemic perfusion indices and myocardial performance in addition to pulmonary hemodynamics.

Many cardiac operations require CPB, with the potential for some degree of postoperative hemodynamic instability. Although off-pump coronary revascularization is an option for selected patients, CPB is still necessary for a significant number of patients undergoing coronary bypass.^{37,38} In effect, the most compromised coronary patients are frequently selected out to undergo operation with CPB, and CPB is still necessary for most valve and extensive aortic procedures. Thus, improved strategies for preventing and treating the hemodynamic sequelae of CPB are a significant clinical goal. The present study demonstrated that selective ET_A antagonism blunted the inevitable rise in PVR in a porcine model of CPB without affecting systemic hemodynamic stability. These results suggest that selective ET_A blockade may prove a useful new therapeutic modality in the limited armamentarium for treatment of pulmonary hypertension after CPB.

We thank Simona C. Baicu, Jeffrey A. Sample, Nancy Harper, Mary C. King, Karla Zeigler, Joseph Sistino, and Dr Tom Brock for their time and expertise.

References

- Downing SW, Edmunds JH Jr. Release of vasoactive substances during cardiopulmonary bypass. *Ann Thorac Surg.* 1992;54:1236-43.
- Dorman BH, Clair MJ, Walker CA, Kratz JM, Crumbley AJ, Zellner JL, et al. Temporal synthesis and release of endothelin within the systemic and myocardial circulation during and following cardiopulmonary bypass. *Circulation.* 1998;(Suppl):1-686.
- Kirshbom PM, Page SO, Jacobs MT, Tsui SS, Bello E, Ungerleider RM, et al. Cardiopulmonary bypass and circulatory arrest increase endothelin-1 production and receptor expression in the lung. *J Thorac Cardiovasc Surg.* 1997;113:777-83.
- Martin ER, Brenner BM, Ballermann BJ. Heterogeneity of cell surface endothelin receptors. *J Biol Chem.* 1990;265:14044-9.
- de Nucci G, Thomas R, D'Orleans-Juste D, Antunes E, Walder C, Warner TD, et al. Pressor effects of circulating endothelin are limited by its removal in the pulmonary circulation and by the release of prostacyclin and endothelium-derived relaxing factor. *Proc Natl Acad Sci U S A.* 1988;85:9797-800.
- Thomas PB, Liu ECK, Webb ML, Mukherjee R, Hebbar L, Spinale FG. Evidence of an endothelin-1 autocrine loop in cardiac myocytes: relation to contractile function with congestive heart failure. *Am J Physiol.* 1996;271:H2629-37.
- Spinale FG, Walker JD, Mukherjee R, Iannini JP, Keever AT, Gallagher KP. Concomitant endothelin receptor subtype A blockade during the progression of congestive heart failure in rabbits has direct and beneficial effects on left ventricular and myocyte function. *Circulation.* 1997;95:1918-29.
- Lerman A, Edwards BS, Hallett JW, Heublein DM, Sandberg SM, Burnett JC Jr. Circulating and tissue endothelin immunoreactivity in advanced atherosclerosis. *N Engl J Med.* 1991;325:997-1001.
- Mulder P, Richard V, Derumeaux G, Hogue M, Henry JP, Lallemand F, et al. Role of endogenous endothelin in chronic heart failure: effect of long-term treatment with an endothelin antagonist on survival, hemodynamics, and cardiac remodeling. *Circulation.* 1997;96:1976-82.
- Gray GA, Webb DJ. The endothelin system and its potential as a therapeutic target in cardiovascular disease. *Pharmacol Ther.* 1996;72:109-48.
- Shiffrin EL, Intengan HD, Thibault G, Touyz RM. Clinical significance of endothelin in cardiovascular disease. *Curr Opin Cardiol.* 1997;12:354-67.
- Rosenberg AA, Kennaugh J, Koppenhager SL, Loomis M, Chatfield BA, Abman SH. Elevated immunoreactive endothelin-1 levels in newborn infants with persistent pulmonary hypertension. *J Pediatr.* 1993;123:109-14.
- Bando K, Vifayaraghavan P, Turrentine MW, Shary TG, Ensing GJ, Sekine Y, et al. Dynamic changes of endothelin-1, nitric oxide, and cyclic GMP in patients with congenital heart disease. *Circulation.* 1995;96(Suppl 9):346-51.
- Komai H, Adatia IT, Elliott MF, de Leval MR, Haworth SG. Increased plasma levels of endothelin-1 after cardiopulmonary bypass in patients with pulmonary hypertension and congenital heart disease. *J Thorac Cardiovasc Surg.* 1993;106:473-8.
- Antonelli M, Letizia C, Tritapepe L, Raponi GM, Ghezzi MC, Menichetti A, et al. Extracorporeal circulation does not induce intrapulmonary release of endothelin-1, but only a modest overproduction in pulmonary circulation. *J Thorac Cardiovasc Surg.* 1999;404:487-94.
- Teerlink JR, Loffler BM, Hess P, Maire JP, Clozel M, Clozel JP. Role of endothelin in the maintenance of blood pressure in conscious rats with chronic heart failure: acute effects of the endothelin receptor antagonist RO 47-0203 (bosentan). *Circulation.* 1994;90:2510-8.
- Roux S, Breu V, Ertel SI, Clozel M. Endothelin antagonism with bosentan: a review of potential applications. *J Mol Med.* 1999;77:364-76.
- Kirshbom PM, Jacobs MT, Tsui SS, DiBernardo LR, Schwinn DA, Ungerleider RM, et al. Effects of cardiopulmonary bypass and circulatory arrest on endothelium dependent vasodilation in the lung. *J Thorac Cardiovasc Surg.* 1996;111:1248-56.
- Shafique T, Johnson RG, Dai HB, Weintraub RM, Sellke FW. Altered pulmonary microvascular reactivity after total cardiopulmonary bypass. *J Thorac Cardiovasc Surg.* 1993;106:479-86.
- Janakidevi K, Fisher MA, Del Vecchio PJ, Tirupathi C, Figge J, Malik AB. Endothelin-1 stimulates DNA synthesis and proliferation of pulmonary artery smooth muscle cells. *Am J Physiol.* 1992;6:C1295-301.
- Dupuis J, Goresky CA, Fournier A. Pulmonary clearance of circulating endothelin-1 in dogs in vivo: exclusive role of ETB receptors. *J Appl Physiol.* 1996;81:1510-5.
- Dorman HB, Hebbar L, Zellner JL, New RB, Houck WV, Acsell J, et al. ATP-sensitive potassium channel activation prior to cardioplegia: effects on ventricular and myocyte function. *Circulation.* 1998;98:II-176-83.
- Wu C, Chan MF, Stavros F, Raju B, Okun I, Mong S, et al. Discovery of TBC 11251, a potent, long acting orally active endothelin receptor-A selective antagonist. *J Med Chem.* 1997;40:1690-7.
- Spinale FG, de Gasparo M, Whitebread S, Hebbar L, Clair MJ, Melton M, et al. Modulation of the renin-angiotensin pathway through enzyme inhibition and specific receptor blockade in pacing induced heart failure. I. Effects on left ventricular performance and neurohormonal systems. *Circulation.* 1997;96:2385-96.
- Krombach RS, Clair MJ, Hendrick JW, Mukherjee R, Houck WV, Hebbar L, et al. Amlodipine therapy in congestive heart failure: hemodynamic and neurohormonal effects at rest and after treadmill exercise. *Am J Cardiol.* 1999;84:3L-15L.
- Masaki T, Miwa S, Sawamura T, Ninomiya H, Okamoto Y. Subcellular mechanisms of endothelin action in vascular system. *Eur J Pharmacol.* 1999;375:133-8.
- Miyauchi T. Pathophysiology of endothelin in the cardiovascular system. *Ann Rev Phys.* 1999;61:391-415.
- Davila-Roman VG, Waggoner AD, Hopkins WE, Barzilai B. Right ventricular dysfunction in low output syndrome after cardiac operations: assessment by transesophageal echocardiography. *Ann Thorac Surg.* 1995;60:1081-6.
- Stein KL, Breisblatt W, Wolfe C, Gasior T, Hardesty R. Depression and recovery of right ventricular function after cardiopulmonary bypass. *Crit Care Med.* 1990;18:1197-200.
- Schmid ER, Burki C, Engel MH, Schmidlin D, Tornic M, Seifert B. Inhaled nitric oxide versus intravenous vasodilators in severe pulmonary hypertension after cardiac surgery. *Anesth Analg.* 1999;89:1108-15.
- Troncy E, Francoeur M, Blaise G. Inhaled nitric oxide: clinical applications, indications, and toxicology. *Can J Anaesth.* 1997;44:973-88.
- Goodwin AT, Amrani M, Gray CC, Chester AH, Yacoub MH. Inhibition of endogenous endothelin during cardioplegia improves low coronary reflow following prolonged hypothermic arrest. *Eur J Cardiothorac Surg.* 1997;11:981-7.
- Carteaux JP, Roux S, Siaghy M, Schjoth B, Dolofon P, Bechamps Y, et al. Acute pulmonary hypertension after cardiopulmonary bypass in pig: the role of endogenous endothelin. *Eur J Cardiothorac Surg.* 1999;15:346-52.
- Kirshbom PM, Tsui SSL, DiBernardo LR, Meliones JN, Schwinn DA, Ungerleider RM, et al. Blockade of endothelin-converting enzyme reduces pulmonary hypertension after cardiopulmonary bypass and circulatory arrest. *Surgery.* 1995;118:440-5.
- Petrossian E, Parry AJ, Reddy VH, Akkersdijk GP, McMullan DM, Thompson L, et al. Endothelin receptor blockade prevents the rise in pulmonary vascular resistance after cardiopulmonary bypass in lambs with increased pulmonary blood flow. *J Thorac Cardiovasc Surg.* 1999;117:314-22.
- Prendergast B, Newby DE, Wilson LE, Webb DJ, Mankad PS. Early therapeutic experience with the endothelin antagonist BQ-123 in pulmonary hypertension after congenital heart surgery. *Heart.* 1999;82:505-8.
- Cartier R. Systematic off-pump coronary artery revascularization: experience of 275 cases. *Ann Thorac Surg.* 1999;68:1494-7.
- Deigeler A, Matin M, Falk V, Binner C, Walther T, Autschbach R, et al. Indication and patient selection in minimally invasive and off-pump coronary artery bypass grafting. *Eur J Cardiothorac Surg.* 1999;16(Suppl 1):S79-82.