THE EFFECTS OF CYANOSIS ON MYOCARDIAL BLOOD FLOW, OXYGEN UTILIZATION, AND LACTATE PRODUCTION IN DOGS

To elucidate differences in myocardial blood flow and metabolism between cyanotic and normal hearts, a model of chronic cyanosis was created in five adult mongrel dogs by anastomosing the inferior vena cava to the left atrium. After 6 to 9 months, myocardial blood flow, the ratio of subendocardial to subepicardial flow, oxygen consumption, oxygen extraction ratio, and lactate consumption in these cyanotic dogs and five control dogs were determined under baseline conditions and during pharmacologic stress with isoproterenol (0.2 $\mu\text{g/kg/min}$). Radioactive microspheres were used to determine left and right ventricular blood flow rates, and arterial and coronary sinus differences in oxygen and lactate levels were measured. At baseline and during stress, oxygen consumption and oxygen extraction ratios were identical in control and cyanotic hearts. Total myocardial blood flow was increased with stress and did not differ between cyanotic and control hearts. Left ventricular muscle from cyanotic hearts did exhibit lower endocardial/epicardial blood flow ratios than those of control hearts at rest, and the relative subendocardial flow decreased further with stress. During isoproterenol infusion, myocardial lactate production, indicative of anaerobic metabolism, was evident in two of five cyanotic animals and none of the control dogs. The relative subendocardial ischemia and its further aggravation by stress in cyanotic hearts may contribute to the pathophysiologic basis of myocardial dysfunction in cyanotic heart disease. (*J THORAC CARDIOVASC SURG* 1995;109:849-53)

Ara K. Pridjian, MD, Edward L. Bove, MD, and Flavian M. Lupinetti, MD,
Ann Arbor, Mich.

Cyanotic hearts often exhibit decreased baseline and postoperative contractile function in addition to poor metabolic reserve.¹⁻³ Although the reasons for this have not been completely explained, previous studies have suggested that myocardial perfusion may be abnormal in cyanotic hearts.⁴⁻⁶ It has been postulated that during periods of stress, oxygen demands in cyanotic hearts exceed supply. This results in episodes of myocardial hypoxia that may lead to myocardial necrosis and fibrosis. The subendocardium is likely to be particularly susceptible, and pathologic changes have been observed in the subendocardium of children with cyanotic heart disease.⁷

Early studies of children with cyanotic heart dis-

ease showed that total coronary artery blood flow was decreased, perhaps as a result of polycythemia.^{5,6} The oxygen extraction was slightly elevated in these children, so that oxygen consumption was equal to that of normal hearts. However, the methods used to measure coronary artery flow in these human subjects were necessarily indirect, and conclusions regarding oxygen consumption may not be valid. Furthermore, cyanotic heart disease is often accompanied by some degree of ventricular hypertrophy, and this can produce abnormalities in blood flow distribution and high energy phosphate metabolism.^{8,9} This study was designed to assess the effects of cyanosis without hypertrophy on myocardial blood flow and oxygen metabolism.

Material and methods

Procedures were done under sterile conditions in a veterinary operating room. Five adult mongrel dogs were anesthetized with sodium pentobarbital, intubated, and the lungs mechanically ventilated. Cefazolin 25 mg/kg was given by intramuscular injection. The chest was entered through the sixth intercostal space on the right. Animals were heparinized (100 mg/kg). The inferior vena cava was divided and anastomosed to the inferior pulmonary vein/left atrial junction in an end-to-side fashion with running

From the Thoracic Surgery Research Laboratory, Section of Thoracic Surgery, Department of Surgery, The University of Michigan School of Medicine, Ann Arbor, Mich.

Received for publication April 21, 1994.

Accepted for publication August 15, 1994.

Address for reprints: Ara K. Pridjian, MD, Ochsner Clinic, 1514 Jefferson Highway, New Orleans, LA 70121.

Copyright © 1995 by Mosby-Year Book, Inc.

0022-5223/95 \$3.00 + 0 12/1/59970

Table I. Heart rates and blood pressures in control and cyanotic dogs at baseline and during stress

	Control	Cyanotic
Baseline heart rate (beats/min)	123 ± 10	121 ± 5
Stress heart rate (beats/min)	225 ± 22	210 ± 18
Baseline systolic blood pressure (mm Hg)	90 ± 9	84 ± 8
Stress systolic blood pressure (mm Hg)	110 ± 15	102 ± 12
Baseline diastolic blood pressure (mm Hg)	50 ± 7	47 ± 9
Stress diastolic blood pressure (mm Hg)	55 ± 12	57 ± 10

Table II. Oxygen extraction ratios in control and cyanotic dogs at baseline and during stress

	Control	Cyanotic
Baseline (%)	61 ± 4	61 ± 4
Stress (%)	61 ± 4	62 ± 3

5-0 polypropylene suture. The thoracotomy was closed and animals were allowed to recover for 6 to 9 months. The control group comprised five animals that did not have an operation.

At the time of the experiment animals were again anesthetized and the lungs ventilated with 100% oxygen. A median sternotomy was done and catheters were placed in the left atrium, the descending aorta, and the coronary sinus. Radioactive microspheres (15 μ m, ^{141}Ce and ^{113}Sn) were injected into the left atrium as blood was withdrawn from the aorta. Coronary sinus and aortic samples were drawn simultaneously for oxygen content and lactate analysis. The oxygen extraction ratio was calculated by dividing the arteriovenous difference in oxygen by the arterial oxygen content. Lactate level was determined with the use of a colorimetric assay (Kodak Ektachem WR-700, Kodak Co., Rochester, N.Y.). Intravenous fluid administration was minimized so that the cyanotic animals remained polycythemic. All measurements were obtained under baseline conditions and were repeated 10 minutes after a continuous intravenous infusion of isoproterenol, 0.2 μ g/kg per minute, was begun.

At the end of the experiment hearts were excised and left and right ventricles were separated and subdivided. The ventricles were weighed and indexed to the body surface area.¹⁰ Each piece was then sectioned into endocardial, middle, and epicardial thirds, which were weighed and counted in a gamma counter with reference blood samples. Standard formulas were used to determine total myocardial blood flow and the ratio of subendocardial to subepicardial flow.¹¹ The interventricular septum was not used for calculation of these ratios. With this flow data and the aorta to coronary sinus differences in oxygen and

Table III. Myocardial oxygen consumption in control and cyanotic dogs at baseline and during stress

	Control	Cyanotic
Baseline (ml oxygen/min/gm ventricle)	1.17 ± 0.05	1.29 ± 0.10
Stress (ml oxygen/min/gm ventricle)	2.22 ± 0.17	2.09 ± 0.18

lactate levels, myocardial oxygen consumption and lactate utilization or production were determined.

Results are given as the mean plus or minus the standard error of the mean. Student's *t* test was used to compare groups. Differences were considered significant at *p* < 0.05.

Animals received humane care in compliance with the "Guide for the Care and Use of Laboratory Animals" published by the National Institutes of Health (NIH Publication No. 85-23, revised 1985).

Results

Six to 9 months after creation of the model, mean arterial oxygen tension and oxygen saturation in room air were 48 ± 4 mm Hg and $76\% \pm 4\%$, respectively, compared with 85 ± 5 mm Hg and $97\% \pm 1\%$ in control dogs. Hematocrit values averaged $56\% \pm 2\%$ in cyanotic dogs compared with $34\% \pm 2\%$ in control dogs. Differences in oxygen tension, oxygen saturation, and hematocrit value were significant (*p* < 0.05). Indexed left and right ventricular weights in cyanotic dogs were 115 ± 7 and 29 ± 2 gm/m², respectively, compared with 119 ± 5 and 36 ± 2 gm/m² in control animals. Mean right ventricular mass in cyanotic dogs was significantly less than that of control dogs (*p* < 0.05).

In Table I, baseline and stress heart rates and blood pressures are shown. There were no differences in these parameters between control and cyanotic groups. The oxygen extraction ratios and myocardial oxygen consumption are shown in Tables II and III. The oxygen extraction ratio was the same in control and cyanotic groups and did not change with stress. Myocardial oxygen consumption increased with stress in both groups and was not statistically different between groups.

Myocardial blood flow and the subendocardial/subepicardial ratio of blood flow are reported in Table IV. Under baseline conditions, left and right ventricular myocardial blood flow values were nearly identical in control and cyanotic hearts. With isoproterenol, myocardial blood flow nearly doubled in both groups. Differences between groups were not significant. The left ventricles of cyanotic hearts

Fig. 1. Myocardial lactate metabolism in control and cyanotic dogs. Two of five cyanotic dogs had lactate production during stress.

Table IV. Myocardial blood flows and subendocardial/subepicardial blood flow ratios in control and cyanotic hearts at baseline and during stress

	Control flow (ml/min/gm ventricle)	Cyanotic flow (ml/min/gm ventricle)	Control ratio	Cyanotic ratio
Baseline				
Left ventricle	1.34 ± 0.03	1.38 ± 0.07	1.27 ± 0.02	$1.02 \pm 0.07^*$
Right ventricle	1.12 ± 0.04	1.14 ± 0.05	0.95 ± 0.1	1.01 ± 0.09
Stress				
Left ventricle	2.16 ± 0.15	1.91 ± 0.15	0.94 ± 0.02	$0.85 \pm 0.08^*$
Right ventricle	2.25 ± 0.07	2.19 ± 0.22	0.93 ± 0.04	1.01 ± 0.04

* $p < 0.05$ versus control.

exhibited significantly lower endocardial/epicardial ratios than those of control hearts at rest ($p < 0.05$). Right ventricular blood flow ratios were comparable. With stress, the relative blood flow decreased further in cyanotic left ventricles, again with a significantly lower endocardial/epicardial ratio than that in control hearts ($p < 0.05$).

Lactate metabolism for control and cyanotic dogs is shown in Fig. 1. Net lactate consumption is depicted by a *positive bar*, and a *negative bar* signifies lactate production. All hearts consumed lactate at rest. Lactate was produced in two of five cyanotic hearts during stress, but in none of the control hearts.

Discussion

The cause of subendocardial underperfusion in the systemic ventricle of cyanotic hearts is probably multifactorial. Cyanotic ventricles are often exposed to conditions of abnormal pressure and volume loading. This can lead to increased wall tension and decreased subendocardial perfusion. In a clinical study, Graham and associates¹ found that the left ventricular end-diastolic volume nearly doubled after a systemic-pulmonary artery shunt procedure and that the degree of dilatation of the ventricle correlated with postoperative pump function.

Heart rates and blood pressures in the two groups were not significantly different at baseline or with

stress. This is important to the validity of our findings relating to the distribution of blood flow. With lower diastolic blood pressure, the driving pressure for coronary artery perfusion decreases, particularly affecting the subendocardium. Lower perfusion pressure was therefore not responsible for our findings. Diastolic blood pressure can be quite low in patients with large systemic-pulmonary artery shunts, resulting in a decrease in myocardial perfusion pressure with relative and actual myocardial ischemia.

Our model produced cyanosis without hypertrophy, as shown by the equal left ventricular weights in both groups. Right ventricular mass in cyanotic animals was significantly smaller than normal. In this model the right ventricle receives only superior vena caval and coronary sinus blood. The smaller right ventricular mass is presumably a result of directing lower body systemic venous return away from the right ventricle and into the left.

Polycythemia can increase blood viscosity and coronary vascular resistance, and this may affect subendocardial more than subepicardial vessels.¹² In an earlier study that used the same model of chronic cyanosis, Lupinetti and colleagues⁴ showed that the ratio of subendocardial to subepicardial blood flow in cyanotic hearts was normal at baseline. However, in that study the cyanotic animals were treated with hemodilution to make hematocrit values normal before blood flow rates were assessed. It appears that at baseline, the only significant difference between the original study and the present experiment is the polycythemia, which suggests that this may contribute to subendocardial underperfusion.

The myocardial oxygen extraction was nearly identical in control and cyanotic hearts. This is in contrast to the findings of earlier clinical studies in which cyanotic patients were found to have decreased myocardial blood flow and increased oxygen extraction compared with respective findings in normal control subjects.^{5,6} However, methods of determining myocardial blood flow and weight in these human subjects were necessarily imprecise. In addition, the patients had a variety of congenital anomalies and did not comprise a homogenous population. Whereas it is possible that the cyanotic and hypertrophied hearts from these patients may have demonstrated supernormal oxygen extraction values, there is little basis for hypothesizing that a heart can become a more efficient extractor of oxygen.

Only two of five cyanotic dogs exhibited lactate production during stress. This finding is in agree-

ment with findings of the clinical study from Friedli and associates.¹³ Hearts from 5 of 14 cyanotic patients stressed by atrial pacing produced lactate, which indicated a shift to anaerobic metabolism. All other patients demonstrated normal aerobic metabolism at baseline and during stress. Myocardial metabolism in cyanotic congenital heart disease is normal at rest, but oxygen supply does not meet the demand in some subjects during stress. In the study of Friedli and associates,¹³ these were the patients who were thought to be at particularly high risk for myocardial fibrosis from repeated episodes of inadequate oxygen supply.

Hemoglobin itself provides a compensatory mechanism in cyanosis. Cyanotic patients function on the steepest portion of the oxyhemoglobin dissociation curve, where oxygen is rapidly unloaded from hemoglobin.¹⁴ It is tempting to speculate that erythrocyte 2,3-diphosphoglycerate may be elevated in cyanosis. By lowering the oxygen affinity of hemoglobin, this molecule shifts the dissociation curve to the right, favoring release of oxygen to the tissues.¹⁵

In this model of cyanosis the subendocardium of the systemic ventricle received less blood flow than normal. Clinically, the subendocardium of patients with cyanotic congenital heart disease may be particularly susceptible to ischemic injury, and subendocardial ischemia may play an important role in the pathophysiologic basis of myocardial dysfunction in cyanotic heart disease. This study lends support to the importance of pursuing strategies that avoid or minimize the duration of cyanosis and polycythemia in patients with congenital heart disease.

REFERENCES

1. Graham TP, Erath HG, Boucek RF, Boerth RC. Left ventricular function in cyanotic congenital heart disease. *Am J Cardiol* 1980;45:1231-6.
2. Borrow KM, Keane JF, Castaneda AR, Freed MD. Systemic ventricular function in patients with tetralogy of Fallot, ventricular septal defect, and transposition of the great arteries repaired during infancy. *Circulation* 1981;64:878-85.
3. Graham TP, Erath HG, Buckspan GS, Fisher RD. Myocardial anaerobic metabolism during isoprenaline infusion in a cyanotic animal model: possible cause of myocardial dysfunction in cyanotic congenital heart disease. *Cardiovasc Res* 1979;13:401-6.
4. Lupinetti FM, Wareing TH, Huddleston CB, et al. Pathophysiology of chronic cyanosis in a canine model. *J THORAC CARDIOVASC SURG* 1985;90:291-6.

5. Rudolf W. Myocardial metabolism in cyanotic congenital heart disease. *Cardiology* 1971;56:209-15.
6. Scheuer J, Shaver JA, Kroetz FW, Leonard JJ. Myocardial metabolism in cyanotic congenital heart disease. *Cardiology* 1970;55:193-210.
7. Franciosi RA, Blanc WA. Myocardial infarcts in infants and children: I—a necropsy study in congenital heart disease. *J Pediatr* 1968;73:309-19.
8. Silverman NA, Kohler J, Levitsky S, Pavel DG, Fang RB, Feinberg H. Chronic hypoxemia depresses global ventricular function and predisposes to the depletion of high-energy phosphates during cardioplegic arrest: implication for surgical repair of cyanotic congenital heart defects. *Ann Thorac Surg* 1984;37:304-8.
9. Bache RJ, Vrobel TR, Arentzen CE, Ring WS. Effect of maximal coronary vasodilation on transmural myocardial perfusion during tachycardia in dogs with left ventricular hypertrophy. *Circ Res* 1981;49:742-50.
10. Cowgill GR, Drabkin DL. Determination of formula for surface area of dog together with consideration of formulae available for other species. *Am J Physiol* 1927;81:36-61.
11. Domenech RF, Hoffman JIE, Noble MIM, et al. Total and regional coronary blood flow measured by radioactive microspheres in conscious and anesthetized dogs. *Circ Res* 1969;25:581-96.
12. Replogle RL, Merrill EW. Experimental polycythemia and hemodilution. *J THORAC CARDIOVASC SURG* 1970;60:582-8.
13. Friedli B, Haenni B, Moret P, Opie LH. Myocardial metabolism in cyanotic congenital heart disease studied by arteriovenous differences in lactate, phosphate, and potassium at rest and during atrial pacing. *Circulation* 1977;55:647-52.
14. Kilmartin JV. Interaction of haemoglobin with protons, CO₂, and 2,3-diphosphoglycerate. *Br Med Bull* 1976;32:209-22.
15. Marschner JP, Seidlitz T, Rietbrock N. Influence of 2,3-diphosphoglycerate on oxygen dissociation kinetics of human hemoglobin. *Int J Clin Pharmacol Ther Toxicol* 1992;30:546-7.

Bound volumes available to subscribers

Bound volumes of THE JOURNAL OF THORACIC AND CARDIOVASCULAR SURGERY are available to subscribers (only) for the 1995 issues from the Publisher, at a cost of \$91.00 for domestic, \$118.77 for Canadian, and \$111.00 for international subscribers for Vol. 109 (January-June) and Vol. 110 (July-December). Shipping charges are included. Each bound volume contains a subject and author index and all advertising is removed. Copies are shipped within 60 days after publication of the last issue of the volume. The binding is durable buckram with the JOURNAL name, volume number, and year stamped in gold on the spine. *Payment must accompany all orders.* Contact Mosby-Year Book, Inc., Subscription Services, 11830 Westline Industrial Drive, St. Louis, Missouri 63146-3318, USA; phone 1 (800) 453-4351 or (314) 453-4351.

Subscriptions must be in force to qualify. Bound volumes are not available in place of a regular JOURNAL subscription.