A method for collecting right coronary venous blood samples from conscious dogs

XIAOMING BIAN, BRADLEY J. HART, ARTHUR G. WILLIAMS, JR., AND H. FRED DOWNEY
Department of Integrative Physiology, University of North Texas
Health Science Center at Fort Worth, Fort Worth, Texas 76107-2699

Bian, Xiaoming, Bradley J. Hart, Arthur G. Williams, Jr., and H. Fred Downey. A method for collecting right coronary venous blood samples from conscious dogs. Am. J. Physiol. 276 (Heart Circ. Physiol. 45): H1086–H1090, 1999.—This report describes for the first time a technique to collect right coronary venous blood samples from conscious dogs. Catheters, prepared from Micro-Renathane tubing, were surgically implanted in right ventricular superficial veins of three anesthetized dogs. Also implanted were an arterial catheter, a right coronary flow transducer, and a right coronary artery constrictor. The coronary catheter was introduced at a venous bifurcation so that its side holes were positioned above the bifurcation; both ends of the catheter were exteriorized. Heparinized saline was continuously infused through the venous catheter by a battery-powered pump. The dogs were maintained for 10–13 days after surgery, and all catheters remained patent. Multiple right coronary venous samples were collected from each dog. These samples were analyzed for venous oxygen tension (PvO2) under baseline conditions, with right coronary pressure reduced to 50 mmHg, and during the reactive hyperemia after release of the right coronary artery constriction. PvO2 was 27.7 ± 1.0 mmHg at baseline, 23.4 ± 1.0 mmHg during coronary artery constriction, and 34.3 ± 1.5 mmHg during reactive hyperemia. These data and the position of the catheter at autopsy demonstrated that coronary venous blood had been sampled.

coronary venous catheter; right coronary oxygen tension

INVESTIGATIONS OF CORONARY PHYSIOLOGY and myocardial metabolism frequently require samples of coronary venous blood. The left coronary circulation drains primarily to the coronary sinus, which can be catheterized in humans and in relatively large, anesthetized, or conscious experimental animals (4, 7, 10–12). In contrast, samples of right coronary venous blood must be collected from small, superficial veins on the right ventricular surface. These veins can be catheterized in animals in experiments (6, 14), but to date this has been done only in anticoagulated, anesthetized animals. Because the right ventricle performs less work, uses less oxygen, and requires less blood flow than the left ventricle, extrapolations of left ventricular findings to the right ventricle are suspect. For example, oxygen tension measured in right coronary venous blood of anesthetized dogs is greater than that of left coronary venous blood (2, 5, 6, 9, 11, 14). However, the possibility of a selective effect of anesthesia on the right coronary circulation or on right ventricular metabolism cannot be ruled out. Thus it was essential to develop a technique to sample right coronary venous blood in the conscious dog. With this new capability, many investigations of right coronary and right ventricular physiology that require the conscious state can, for the first time, be conducted.

MATERIALS AND METHODS

Coronary venous catheter. The coronary venous catheter was prepared from Micro-Renathane tubing [type MRE-025, 0.025-in. outer diameter (OD), 0.012-in. inner diameter (ID), Braintree Scientific]. Three or four side holes (~0.012 × 0.024 in.) were cut in the central region of a 5- to 6-ft length of tubing. The holes were cut by bending the tubing and excising small ovals along its edge with micro iris curved scissors. A 5-mm² Silastic anchoring patch was glued to the tubing 0.5–1 cm from the side holes. Figure 1 shows a microphotograph of the catheter. This catheter was designed so that the side holes could be positioned in a coronary vein and both ends of the catheter could be exteriorized as described below. Having access to both ends of the venous catheter facilitated flushing and treatment with antithrombotic agents, if necessary, and provided redundant paths for blood to be withdrawn.

Animal instrumentation. Three mongrel dogs of either sex, weighing 24–27 kg were instrumented. Thirty minutes after preanesthesia treatment with acepromazine maleate (0.03 mg/kg im), anesthesia was induced by thiopental sodium (5 mg/kg im). After endotracheal intubation, a surgical plane of anesthesia was maintained by mechanical ventilation with isoflurane gas (1–3%) with equal offset of oxygen (1 liter). Under sterile conditions, a thoracotomy was performed in the fourth right intercostal space. A Tygon catheter (0.04-in. ID, 0.07-in. OD) was inserted into the aorta through the right internal mammary artery to measure aortic pressure and collect arterial samples. A 1- to 2-cm nonbranching section of the proximal right coronary artery was dissected free for attachment of a single crystal Doppler flow probe and a hydraulic occluder. The occluder was positioned distal to the flow probe so that changes in vessel cross-sectional area beneath the velocity-sensing Doppler crystal would not occur when the occluder was inflated.

A Micro-Renathane catheter (type MRE-033, 0.033-in. OD, 0.014-in. ID) was inserted into the right coronary artery distal to the occluder through a small side branch to the right...
atrium. The tip of this catheter was positioned at the origin of the branch. In the territory perfused by the right coronary artery distal to the occluder, one or two superficial veins, each with a bifurcation, were identified (Fig. 2). A 20-gauge, 1.25-in. intravenous Insyte catheter-needle unit (Insyte-W, Becton-Dickinson Vascular Access) was inserted into one branch of the bifurcation, the needle was withdrawn, and the catheter hub was cut off. A Micro-Renathane venous catheter, prepared as described above, was inserted through the Insyte catheter until most of the tubing proximal to the side holes had been advanced centrally through the vein and into the right atrium. Through a purse-string suture sewn in the right atrium above the catheterized vein, a right-angle clamp was inserted to withdraw the venous catheter. The Insyte catheter was removed from the coronary vein. Because the Insyte catheter was positioned between the anchoring patch and the vein, it had to be cut lengthwise so that it could be removed from the venous catheter. The Insyte catheter was then anchored to the right ventricle with silk sutures. This positioned the side holes of the venous catheter in the right coronary superficial vein just central to its bifurcation. Thus, even though a thrombus eventually formed in the catheterized branch, blood continued to flow through the other branch of the bifurcation, so venous blood samples could be collected from either end of the catheter. Immediately after the venous catheter was secured in place, 27-gauge Luer stub adapters with stopcocks were inserted in both ends of the catheter and heparinized saline (10 U/ml) was infused through both ends of the catheter. In two dogs, one right coronary venous catheter was implanted. In one dog, two right coronary venous catheters were implanted.

At the conclusion of instrumentation, catheters and wires were brought out of the thorax through the third and fifth right intercostal spaces, tunneled under the skin, and exteriorized between the shoulders through individual puncture wounds. The chest was closed, and the pneumothorax was evacuated through a chest tube.

The dog was fitted with a jacket (Alice King Chatham). An Ambulatory Infusion Pump (model ML-6-4, Cormed) was positioned in a pocket of the jacket. This pump was connected to the venous end of the right coronary venous catheter so that heparinized saline (5 U/ml) could be continuously infused at 0.02–0.03 ml/min. Because the right atrial end of the catheter was closed, the heparinized saline flowed out of the catheter through the side holes and maintained a patent pathway for intermittent sampling of coronary venous blood. In the dog with two right coronary venous catheters, only one catheter was attached to a pump. The other catheter was filled with heparinized saline (5,000 U/ml) and flushed daily with heparinized saline (10 U/ml).

Antibiotics (Clavamox, 6.25 mg/lb, twice daily) and aspirin (162–300 mg) were given for 10 days after surgery. The right}

**Fig. 1.** Microphotograph of a portion of right coronary venous catheter showing Silastic anchoring patch and side holes.

**Fig. 2.** Schematic diagram of experimental preparation. A Doppler flow probe and a hydraulic occluder were positioned proximally on the right coronary artery. RC pressure (RCP) was monitored through a catheter implanted in a right atrial (RA) branch. Venous samples were withdrawn through sides holes of the venous catheter. Side holes were positioned in an RC superficial vein, central to its bifurcation. A Cormed Ambulatory Infusion Pump was connected to the venous end of the RC venous catheter for infusion of heparinized saline. RV, right ventricle; SVC, superior vena cava; Ao, aorta.
coronary arterial catheter was filled with heparinized saline (5,000 U/ml) and flushed daily with heparinized saline (10 U/ml). The aortic catheter was treated similarly at 3-day intervals.

After the dogs were euthanized, autopsies were performed to confirm that all the side holes of the catheter were still in the superficial right ventricular vein.

Protocol and data collection. Starting from the fifth day after surgery, right coronary venous and systemic arterial blood samples were collected with the animal standing quietly in a sling. These samples were collected daily for 4 days from one dog and every other day for 6 days from two dogs. Blood could be easily withdrawn from the patent catheters at ~0.2 ml/min. One-minute collection provided a sufficient sample for analyses of blood gases and hemoglobin (Instrumentation Laboratories model Synthesis 30). Coincident with blood sampling, right coronary and systemic arterial blood pressure and right coronary flow velocity (CFV) were measured.

Pressure transducers (Narco Telecare model LDI-5) were positioned at midheart level. CFV was measured with a Triton Technology model 100 pulsed Doppler flowmeter. Pressure and velocity signals were averaged electronically and recorded. Coronary venous blood samples and hemodynamic data were collected under baseline conditions. The hydraulic occluder was then inflated to reduce right coronary arterial pressure to 50 mmHg, and blood samples and hemodynamic data were collected. The occluder was then released, and blood samples and hemodynamic data were collected during the ensuing reactive hyperemia.

Statistical analyses. Data are presented individually and as means ± SE. Variables measured under baseline, partial coronary occlusion, and reactive hyperemia were compared with repeated-measures ANOVA. Differences with P < 0.05 are described as statistically significant.

RESULTS

The dogs were maintained for 10–13 days after surgery. In all three dogs, the pump-perfused right coronary venous catheters remained patent at all times. In the dog with an extra, nonperfused right coronary venous catheter, blood samples could be also collected from this catheter, but it was sometimes necessary to flush the catheter with heparinized saline or, on a few occasions, infuse streptokinase (12,500 IU/ml) before blood could be withdrawn.

To verify that the collected blood was from the right coronary venous drainage, the right coronary artery was partially constricted to reduce right coronary arterial pressure to 50 mmHg. This constriction reduced right coronary arterial flow from 0.56 ± 0.02 to 0.35 ± 0.01 ml·min⁻¹·g⁻¹. After arterial and right coronary venous blood samples were collected, the constriction was released. Reactive hyperemia of 1.01 ± 0.05 ml·min⁻¹·g⁻¹ was observed, and during this hyperemia, arterial and right coronary venous blood samples were collected. These maneuvers were expected to reduce and then increase the oxygen tension of the right coronary venous blood.

Systemic hemodynamic variables, arterial and coronary venous hemoglobin, and arterial PO₂ are summarized in Table 1. Heart rate, mean arterial pressure, and arterial PO₂ were normal and not affected by restricting and releasing the right coronary artery. PO₂ of the blood sampled from the right coronary venous catheter (PvO₂) is shown in Fig. 3. The baseline PvO₂ was 27.7 ± 1.0 mmHg. During constriction of the right coronary artery and the subsequent reduction in right coronary flow, PvO₂ fell to 23.4 ± 1.0 mmHg (P < 0.05). During right coronary reactive hyperemia, PvO₂ increased to 34.3 ± 1.5 mmHg (P < 0.05). As shown in Fig. 3, all venous samples had decreases in PvO₂ during right coronary constriction and increases in PvO₂ during reactive hyperemia.

Autopsy confirmed that all of the side holes of the catheter had remained positioned in the right coronary superficial vein.

DISCUSSION

This report describes for the first time a technique to collect right coronary venous blood samples from conscious dogs.

Table 1. Summary of systemic hemodynamic variables, CBF, PaO₂, and Hb

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>Coronary Artery Constriction</th>
<th>Reactive Hyperemia</th>
<th>ANOVA</th>
</tr>
</thead>
<tbody>
<tr>
<td>HR, beats/min</td>
<td>106 ± 5</td>
<td>105 ± 6</td>
<td>107 ± 6</td>
<td>NS</td>
</tr>
<tr>
<td>P, mmHg</td>
<td>103 ± 4</td>
<td>104 ± 4</td>
<td>103 ± 4</td>
<td>NS</td>
</tr>
<tr>
<td>CBF, ml·min⁻¹·g⁻¹</td>
<td>0.36 ± 0.02</td>
<td>0.35 ± 0.01</td>
<td>1.01 ± 0.05</td>
<td>P &lt; 0.05</td>
</tr>
<tr>
<td>PaO₂, mmHg</td>
<td>94.7 ± 2.2</td>
<td>94.0 ± 2.4</td>
<td>94.9 ± 2.1</td>
<td>NS</td>
</tr>
<tr>
<td>Hb, g</td>
<td>12.1 ± 0.7</td>
<td>11.8 ± 0.8</td>
<td>12.3 ± 1.1</td>
<td>NS</td>
</tr>
<tr>
<td>PvO₂, g</td>
<td>12.0 ± 0.6</td>
<td>12.1 ± 0.9</td>
<td>11.9 ± 0.8</td>
<td>NS</td>
</tr>
</tbody>
</table>

Values are means ± SE. Right coronary pressure was 103 ± 4 mmHg at baseline and 50 mmHg during coronary artery constriction. HR, heart rate; P, systemic arterial pressure; CBF, coronary blood flow; PaO₂, arterial oxygen tension; Hb, arterial hemoglobin; PvO₂, venous hemoglobin.
scious dogs. Preliminary data on oxygen tension of these samples is also presented.

To date, investigations of right coronary physiology and right ventricular metabolism have been largely restricted to anesthetized, open-chest models, because right coronary venous blood samples could not be collected from conscious animals. This problem resulted from the small size of the right ventricular superficial veins and the absence of a large common drainage pathway, such as the coronary sinus of the left coronary circulation, which could be chronically catheterized. Furthermore, important differences in right ventricular power output, wall thickness, luminal pressure, and coronary flow limit extrapolations that might be made from left ventricular data. Whereas studies on open-chest dogs have provided information on right ventricular oxygen extraction and consumption under various conditions (6, 14), investigators have been concerned that anesthesia and the acute effects of extensive surgery may have produced artifacts. These concerns have increased with reports of significant differences in right ventricular oxygen extraction and right coronary autoregulatory ability compared with findings in the left ventricle (1–3, 6). Clearly, a need exists to extend these studies to the intact circulation of the conscious animal.

To collect right coronary venous blood from the conscious dog, we developed a novel catheter and catheterization procedure. By entering the venous drainage at a bifurcation, we could position the side holes of our catheter in a stream of flowing blood, even if the entry vessel became obstructed by a thrombus. Because our catheter did not terminate in the vein from which blood was sampled, irritation of the vessel wall by the catheter tip was avoided and the likelihood of thrombus formation was lessened. Finally, with continuous perfusion of heparinized saline through the catheter and out from its side holes, patency of the catheter was maintained, and continued blood flow though the venous drainage was probably enhanced. However, this continuous perfusion may not be essential, because we were able to withdraw blood from one catheter that was not perfused. However, it was necessary to intermittently treat this catheter with streptokinase, a thrombolytic agent. Having external access to both ends of the coronary venous catheter facilitated sampling by providing redundant access to the side hole sampling ports. We believe our catheter and procedure can also be successfully applied to the superficial veins of the left ventricle and, most likely, to the venous drainage of other organs. In such applications, the catheter would have to be exteriorized at an appropriate point downstream from the point of cannulation.

The traditional approach of implanting a single-ended catheter has been successful for collecting samples of left coronary venous blood from the coronary sinus of conscious dogs (7–9, 11) and the anterior interventricular vein of conscious ponies (13). We did not evaluate this approach for collecting right coronary venous blood from conscious dogs because we were unable to maintain such catheters patent in the anterior interventricular vein of dogs.

Right coronary venous blood was analyzed for oxygen tension under baseline and other conditions to verify the position of the sampling ports with the right coronary vein. Under baseline conditions, the $P_{O_2}$ of the sampled blood averaged 27.7 mmHg. Obviously, this is much less than the $P_{O_2}$ of mixed venous blood in the right atrium, indicating that coronary rather than central venous blood was sampled. Furthermore, the $P_{O_2}$ of the sampled blood decreased during constriction of the right coronary artery, and the $P_{O_2}$ rose during the reactive hyperemia after release of the constriction. These findings, along with our observation of the catheter position at autopsy, verified that the catheter remained in place and provided samples of right coronary venous blood. Finally, the similar values for arterial and coronary venous hemoglobin indicated that the samples were not diluted by saline in the venous catheter.

We thank Min Fu, Torel Patel, and Rebecca Campos for technical assistance.

This study was supported by National Heart, Lung, and Blood Institute Grant HL-35027.

Address for reprints requests: X. Bian, Dept. of Integrative Physiology, Univ. of North Texas Health Science Center at Fort Worth, 3500 Camp Bowie Blvd., Fort Worth, TX 76107-2699.

Received 26 August 1998; accepted in final form 26 October 1998.

REFERENCES


