Regional Tolerance to Acute Normovolemic Hemodilution: Evidence That the Kidney May Be at Greatest Risk

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Objective: **To evaluate the regional tolerance to acute normovolemic hemodilution (ANH).**

Design: **Prospective animal study.**

Setting: **University research laboratory.**

Participants: **Nine anesthetized (isoflurane) dogs.**

Interventions: **Hematocrit reduced in 10% decrements using dextran-for-blood exchange until cardiac insufficiency observed.**

Measurements and Main Results: **Cardiac index (CI) was measured using thermodilution and regional blood flow (RBF) in myocardium, brain, spinal cord, kidney, liver, duodenum, pancreas, spleen, skeletal muscle, and skin with radioactive microspheres. Oxygen delivery (DO_2) was calculated from the product of respective blood flow and arterial oxygen content. Systemic oxygen extraction (EO_2) and oxygen consumption (VO_2) were calculated. Increases in CI during ANH were inadequate to prevent decreases in systemic DO_2; however, an increased systemic EO_2 maintained VO_2 during graded ANH to hematocrit < 10%. In the myocardium, brain, and spinal cord, increases in RBF were sufficient to maintain DO_2 across the entire range of hematocrits, but this was not the case in the other organs studied. Of note, renal DO_2 first decreased at a hematocrit of 30% and was only 25% of baseline at a hematocrit of 10%.**

Conclusions: **During graded ANH, increases in RBF were sufficient to maintain DO_2 in only the heart, brain, and spinal cord. The especially marked decrease in DO_2 in the kidney, combined with previous physiologic studies demonstrating its inability to augment EO_2, suggest that this organ may be the most at risk of hypoxic damage during ANH.**

KEY WORDS: hemodilution, oxygen transport, blood conservation, regional blood flow, radioactive microspheres, canine model

ACUTE NORMOVOLEMIC HEMODILUTION (ANH) is a method used to minimize the need for allogeneic blood transfusions.[1] The body has 2 mechanisms available to satisfy its oxygen demand during the attendant decrease in oxygen-carrying capacity of the blood: An increase in cardiac output and an increase in oxygen extraction.[2] Studies were performed in healthy, anesthetized, adult dogs during a progressive colloid-for-blood exchange to determine the hematocrit at which systemic oxygen uptake becomes dependent on systemic oxygen delivery; that is, the so-called critical hematocrit. The findings indicated that this value was in the range of 8%-12%.[3–5] A corresponding value for critical hematocrit has not been obtained in humans. However, a study in healthy conscious humans has demonstrated that systemic oxygen delivery remained adequate during a decrease in hematocrit to 15%, as indicated by no change in either systemic oxygen uptake or plasma lactate concentration.[6]

Systemic indices of oxygen supply and demand are an aggregate of responses in the individual body tissues. A constancy of systemic oxygen uptake during ANH does not necessarily indicate that all organs are adequately perfused and oxygenated. Critical decreases in oxygen delivery in some organs could be masked by increases in oxygen delivery in others. Although previous studies have evaluated the changes in regional blood flow and oxygen delivery during ANH, the studies were limited by a narrow range of hematocrits, a small number of organs, and the use of a research anesthetic, such as pentobarbital.[2,7–9]

The current study was conducted in isoflurane-anesthetized dogs to assess regional blood flow and oxygen delivery while hematocrit was reduced step-wise via an isovolemic blood-for-dextran exchange until cardiac insufficiency was observed (defined as 20% reduction in mean aortic pressure accompanied by an elevated pulmonary capillary wedge pressure). With the use of the radioactive microsphere technique,[10] it was possible to obtain measurements in a wide assortment of organs, including the left and right ventricles, brain, spinal cord, kidney, splanchnic organs, skeletal muscle, and skin. The distribution of blood flow and oxygen delivery in regions within the ventricular walls, brain, and spinal cord also could be assessed. Systemic hemodynamic and metabolic variables were obtained to provide a context for interpreting the regional responses. The current study tested the hypothesis that the body tissues were heterogenous in their tolerance to ANH.

MATERIALS AND METHODS

The study was conducted in compliance with the Institutional Animal Research Committee. Experiments were performed on 9 mongrel adult dogs of either sex (weight range 21-24 kg). Before experimental use, each dog was evaluated thoroughly by the veterinary staff of the Biologic Resources Laboratory at the University of Illinois at Chicago and confirmed to be heartworm-free and in good overall health. Anesthesia was induced with an intravenous bolus injection of thiopental in a dose of 30 mg/kg. After tracheal intubation, anesthesia was maintained by ventilation with 1.4% isoflurane in oxygen, which is the 1 minimum alveolar concentration dose in the dog.[11] The volume and rate of the ventilator were set to maintain arterial carbon dioxide tension (PCO_2) at 35-40 mmHg. Oxygen tension (PO_2), PCO_2, and pH of arterial and mixed venous blood samples were measured electrometrically (model 413, Instrumentation Laboratories, Lexington, MA). Sodium bicarbonate solution was given as necessary to correct

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This project was supported by intramural funding from the Department of Anesthesiology, Advocate Illinois Masonic Medical Center, Chicago, IL.

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1053-0770/2601-0001$36.00/0

http://dx.doi.org/10.1053/j.jvca.2014.06.014

metabolic acidosis. Hematocrit of blood samples was determined with a microcentrifuge. Core body temperature was monitored and maintained at 38° C with a heating pad, warmed intravenous fluids, and warming lights. Heparin, 400 U/kg, was administered postsurgically to prevent blood coagulation.

Polyethylene cannulae were inserted into 1) the thoracic aorta via the right brachial artery for monitoring aortic blood pressure and for obtaining samples of arterial blood for analysis, and 2) the left femoral vein and left femoral artery for isovolemic exchange of whole blood with dextran solution. A 7-Fr Swan-Ganz catheter, equipped with multiple ports and a thermistor at the tip, was introduced into the right femoral vein and flow-directed into the pulmonary artery using pressure monitoring for guidance. This catheter was used to measure pulmonary capillary wedge pressure, to obtain samples of mixed venous blood, and to measure CO by thermodilution. Cardiac index was calculated by dividing CO by body weight.

A left thoracotomy was performed in the fourth intercostal space and the pericardium was incised to expose the heart. A small polyethylene cannula was inserted into the left atrium via the left atrial appendage for injecting radioactive microspheres. Aortic pressure and pulmonary artery pressure were measured with Statham transducers (model P23ID, Gould, Cleveland, OH) and averaged electronically. A permanent record of monitored hemodynamic parameters was obtained with a Gould recorder (model 2800S).

Experimental Measurements

Measurement of regional blood flow. Regional blood flow was measured with 15 ± 3μm labeled with γ-emitting radionuclides, ^{141}Ce, ^{51}Cr, ^{46}Sc, ^{85}Sr, and ^{113}Sn (New England Nuclear Corp, Newton, MA). This technique is validated and well accepted.[8,10] It is based on the following propositions: 1) Microspheres that are pumped out of the left ventricle follow the distribution of the cardiac output, and 2) The microspheres are trapped completely in the microcirculation of the body tissues; thus, the amount of radioactivity in a tissue sample obtained at the termination of an experiment reflects the level of blood flow at the time of microsphere administration. Before administration, microspheres labeled with a specific radionuclide were dispersed in a solution of 10% dextran and agitated in a vortex mixer and in an ultrasonic bath. Approximately 1×10^6 microspheres were injected into the left atrium for each flow determination. The microspheres were flushed into the left atrium over 30 seconds with 5 mL of body-temperature isotonic saline. Neither administration of the microspheres nor withdrawal of reference samples had detectable effects on monitored hemodynamic parameters. Beginning simultaneously with each microsphere injection, duplicate reference blood samples were collected at a rate of 6 mL/min for 3 minutes through 2 cannulae of different lengths threaded into the arch of the aorta via the right femoral artery. Radioactivity of the duplicate reference samples differed by less than 10%, indicating satisfactory mixing of the microspheres in the left ventricular output. Autologous blood was infused into the dog to compensate for the reference blood samples withdrawn for the microsphere technique.

After the final injection of microspheres, the heart was stopped by intravenous injection of potassium chloride, excised, and frozen, to facilitate transmural sampling. Full-thickness samples were obtained from the right and left ventricular walls. The right ventricular samples were cut into halves transmurally and the left ventricular samples were cut into thirds transmurally to yield regional sections. The brain was removed and sectioned using anatomic landmarks: Cerebral cortex, cerebellum, pons, and medulla. The entire spinal cord from C1 to the cauda equina was excised and divided into cervical, thoracic, and lumbar segments. Samples also were obtained from peripheral tissues (kidney cortex, liver, duodenum, pancreas, skeletal [temporalis] muscle, and skin). The tissue samples were transferred to tared counting tubes and weighed. The tissue and reference blood samples were analyzed for radioactivity with a gamma scintillation counter equipped with a multichannel analyzer (model 1282-002, LKB, Turku, Finland). Isotope separation was accomplished by standard techniques of γ-spectroscopy. Values for regional blood flow (RBF; in mL/min/100 g) were calculated from the equation:

$$RBF = ABF \times \left(MC/AC\right) \times 100$$

where ABF is the rate of arterial reference sampling (mL/min), MC is microsphere radioactivity (counts/min/g) in the tissue samples, and AC is the total microsphere radioactivity (counts/min) in the arterial reference samples. The values for regional blood flow within each ventricular wall were averaged to compute a value for mean transmural blood flow. The value for regional blood flow in the subendocardial sample was divided by that in the subepicardial sample to yield a value for endocardial/epicardial flow ratio.

Systemic oxygen uptake. At specified times in the study, 1-mL blood samples were collected anaerobically from the aorta and the pulmonary artery to determine the systemic arteriovenous oxygen difference (a-v O_2 diff). Hemoglobin concentration and percent hemoglobin oxygen saturation of the blood samples were measured with a CO-oximeter (model 482, Instrumentation Laboratories, Lexington, MS), and used to calculate oxygen bound to hemoglobin assuming an oxygen carrying capacity for hemoglobin of 1.39 mL O_2/g. The oxygen dissolved in blood was computed (O_2 dissolved = (0.003 mL O_2/100 mL blood/mmHg) and added to the bound component to compute total oxygen content. Systemic oxygen extraction was determined by dividing the a-v O_2 difference by the oxygen content of the aortic blood sample.

Systemic oxygen uptake (VO_2; mL/min/100 g) was calculated from the Fick equation

$$VO_2 = CI \times \left[(a{-}v)\ O_2\ diff/100\right]$$

where CI is cardiac index at the time that blood samples were obtained. Systemic oxygen delivery and regional oxygen delivery were calculated by multiplying the respective values for blood flow and arterial oxygen content. A 1-mL blood sample was obtained from the aorta and analyzed for plasma lactate concentration using a commercial enzymatic method (Paramax Analytical System, Baxter, Irvine, CA).

Experimental Protocol

The dogs were permitted to stabilize for at least 30 minutes following surgical preparation before baseline measurements were obtained (hematocrit approximately 40%). ANH then was produced by removing blood from the left femoral artery at a rate of 20 mL/min while replacing it with an equal volume of 5% dextran (molecular weight 40,000; American McGaw, Irvine, CA) pumped into the left femoral vein at the same rate, which was intended to maintain isovolemic conditions, as verified by unchanged values for pulmonary capillary wedge pressure. Steady-state measurements were repeated as hematocrit was reduced in 10% absolute steps (30%, 20%, 10%) and then, finally, when cardiac insufficiency was observed, as defined by 20% reduction in mean aortic pressure accompanied by an elevated pulmonary capillary wedge pressure. This combination of hemodynamic changes is associated with myocardial ischemia, as evidenced by lactate production.[12] The current protocol, which involved 5 sequential measurements, was used because it was compatible with the number of differently labeled radioactive microspheres that were available, and because it was used in previous studies,[2] which facilitated a comparison of the results.

Statistical Analysis

A power analysis based on the decrease in systemic oxygen delivery when hematocrit was reduced to 20% in pentobarbital-

Table 1. Systemic Hemodynamic Changes During Graded Acute Normovolemic Hemodilution

	Baseline	HD (30)	HD (20)	HD (10)	Cardiac Insufficiency
MAP, mmHg	111 ± 8	105 ± 8	106 ± 7	98 ± 6*	88 ± 5*
HR, beats/min	149 ± 13	176 ± 6	187 ± 4*	195 ± 8*	169 ± 10*
PCWP, mmHg	3.5 ± 0.5	4.4 ± 0.6	5.1 ± 0.5*	5.9 ± 1.1*	12.3 ± 1.2*
Arterial Variables					
Hct, %	42 ± 1	30 ± 1*	20 ± 1*	10 ± 1*	7 ± 1*
Hb, g/100 mL	14.3 ± 0.4	10.2 ± 0.2*	7.1 ± 0.3*	3.7 ± 0.3*	2.2 ± 0.3*
PO_2, mmHg	333 ± 30	300 ± 37	303 ± 45	290 ± 45	368 ± 30
PCO_2, mmHg	34 ± 1	37 ± 2	37 ± 1	37 ± 1	37 ± 1
pH	7.39 ± 0.02	7.36 ± 0.02	7.38 ± 0.02	7.36 ± 0.01	7.34 ± 0.02
O_2 content, vol. %	20.4 ± 0.6	14.8 ± 0.4*	10.5 ± 0.5*	5.8 ± 0.3*	4.1 ± 0.5*
Lactate, meq/L	1.6 ± 0.3	2.1 ± 0.4	2.0 ± 0.3	2.4 ± 0.4	4.0 ± 0.8*

NOTE. Values are means ± SE.

Abbreviations: Hb, hemoglobin; Hct, hematocrit; HD, hemodilution (number in parentheses is hematocrit level); HR, heart rate; MAP, mean aortic pressure; PCWP, pulmonary capillary wedge pressure.

*$p < 0.05$ *v* baseline.

anesthetized dogs[8] indicated that a sample size of 8 provided 80% power at a two-sided 0.05 significance level. The primary comparison in the study was the difference between the value at each hematocrit level during ANH and the respective baseline value. This difference was assessed using the Student's t test for paired samples. Regression analysis was used to describe the relationship between regional blood flow in the heart, brain, and spinal cord and hematocrit over the range in which hemodynamic conditions were essentially stable (baseline to 10%). A $P < 0.05$ was considered statistically significant. Data are presented as mean ± standard error of the mean.

RESULTS

Systemic hemodynamic variables and arterial blood gases during graded ANH are presented in Table 1. Graded ANH caused parallel decreases in hematocrit, hemoglobin concentration, and arterial oxygen content. Mean aortic pressure and pulmonary capillary wedge pressure were essentially constant until hematocrit was reduced to an average of 7 ± 1%, when a decrease in mean arterial pressure (-21%) and an increase in pulmonary capillary wedge pressure (+251%) were evidence of cardiac insufficiency. These adverse hemodynamic changes coincided with an increase in arterial lactate concentration (+150%). The corresponding values for systemic oxygen extraction, mixed venous hemoglobin saturation, and mixed venous PO_2 were 64.3 ± 3.8 %, 44.9 ± 3.6 %, and 30.2 ± 2.6 mmHg, respectively.

Graded ANH caused increases in cardiac index that were less than proportional to the imposed reductions in arterial oxygen content; thus, systemic oxygen delivery decreased (Table 2). However, systemic oxygen extraction increased sufficiently to maintain systemic oxygen uptake. The increases in cardiac index during ANH paralleled those in heart rate.

Graded ANH caused progressive increases in blood flow within the left and right ventricular walls, brain, and spinal cord (Table 3, Fig 1). The responses in the ventricular walls were transmurally uniform, as reflected in unchanging endo:epi flow ratios. Over the hematocrit range 30%-10%, the increases in myocardial blood flow were sufficient to increase oxygen delivery in the left ventricle and to maintain oxygen delivery at the baseline level in the right ventricle (Table 4). Oxygen delivery was maintained in all regions of the brain and spinal cord over the entire ANH protocol.

Graded ANH caused heterogenous changes in blood flow in the other organs (Table 3). Blood flow in the kidney and skin did not change over the course of ANH. Blood flow in the remaining organs did not change during moderate ANH (hematocrit of 30%), and it either increased (liver, duodenum, pancreas, and skeletal muscle) or decreased (spleen) during more severe ANH. Oxygen delivery was reduced at all levels of ANH in the kidney, but was maintained until hematocrit was reduced to 20% in the spleen, skeletal muscle, and skin and to 10% in the liver, duodenum, and pancreas (Table 4). Of note was that renal oxygen delivery was only 25 ± 5% of baseline at a hematocrit of 10% (Fig 2).

DISCUSSION

The main findings from this study were: (1) Increases in cardiac index and oxygen extraction maintained systemic oxygen uptake during graded ANH to a hematocrit <10%.

Table 2. Systemic Changes in Oxygen Metabolism During Graded Acute Normovolemic Hemodilution

	Baseline	HD (30)	HD (20)	HD (10)	Cardiac Insufficiency
CI, mL/min/kg	67.5 ± 5.7	75.3 ± 6.0	82.0 ± 3.1*	107.1 ± 7.6*	98.6 ± 6.4*
DO_2, mL/min/kg	14.3 ± 1.3	11.5 ± 1.1*	8.7 ± 0.3*	6.1 ± 0.5*	4.1 ± 0.5*
VO_2, mL/min/kg	3.0 ± 0.3	3.8 ± 0.2	3.6 ± 0.2	3.3 ± 0.2	2.5 ± 0.1
EO_2, %	20.8 ± 1.3	34.2 ± 3.2*	41.5 ± 2.0*	54.3 ± 1.4*	64.3 ± 3.8*

NOTE. Values are means ± SE.

Abbreviations: CI, cardiac index; DO_2, systemic oxygen delivery; EO_2, systemic oxygen extraction; HD, hemodilution (number in parentheses is hematcrit level); VO_2, systemic oxygen uptake.

*$P < 0.05$ *v* baseline.

Table 3. Regional Blood Flow (in mL/min/100 g) During Graded Acute Normovolemic Hemodilution

	Baseline	HD (30)	HD (20)	HD (10)	Cardiac Insufficiency
Heart					
Left Ventricle					
Subepicardium	83 ± 5	143 ± 15*	223 ± 21*	410 ± 45*	421 ± 52*
Midmural	74 ± 6	136 ± 17*	216 ± 21*	373 ± 26*	454 ± 65*
Subendocardium	76 ± 6	136 ± 17*	208 ± 18*	357 ± 43*	422 ± 64*
Mean Transmural	78 ± 5	139 ± 16*	216 ± 20*	386 ± 46*	432 ± 62*
Endo:epi ratio	0.9 ± 0.1	0.9 ± 0.1	0.9 ± 0.1	0.9 ± 0.1	1.0 ± 0.1
Right Ventricle*					
Subepicardium	59 ± 7	88 ± 14*	135 ± 11*	219 ± 20*	270 ± 47*
Subendocardium	49 ± 7	83 ± 16*	138 ± 9*	232 ± 27*	306 ± 27*
Mean Transmural	54 ± 6	86 ± 14*	137 ± 7*	225 ± 21*	288 ± 35*
Endo:epi ratio	0.9 ± 0.1	1 ± 0.1	1.1 ± 0.1	1.1 ± 0.1	1.2 ± 0.2
CNS					
Cerebral Cortex	41 ± 5	54 ± 6*	87 ± 10*	131 ± 13*	175 ± 15*
Cerebellum	39 ± 4	50 ± 6*	74 ± 9*	121 ± 13*	165 ± 14*
Pons	22 ± 3	28 ± 3*	46 ± 7*	72 ± 9*	98 ± 15*
Medulla	23 ± 2	32 ± 3*	46 ± 7*	74 ± 8*	115 ± 20*
Cervical Spinal Cord	20 ± 3	27 ± 3*	39 ± 8*	63 ± 9*	95 ± 19*
Thoracic Spinal Cord	12 ± 2	15 ± 2	20 ± 3*	37 ± 4*	63 ± 8*
Lumbar Spinal Cord	14 ± 2	18 ± 2	23 ± 3*	40 ± 6*	68 ± 7*
Peripheral Organs					
Renal Cortex	449 ± 66	502 ± 92	494 ± 82	445 ± 96	587 ± 129
Liver (Hep. Artery)	32 ± 7	47 ± 8	46 ± 12	62 ± 22*	87 ± 2*
Duodenum	43 ± 5	48 ± 7	65 ± 7*	83 ± 13*	87 ± 26*
Pancreas	17 ± 2	23 ± 5	34 ± 6*	35 ± 12*	39 ± 8*
Spleen	146 ± 32	152 ± 32	101 ± 12	68 ± 11*	81 ± 30*
Skeletal Muscle	3.9 ± 0.9	3.6 ± 0.5	4.2 ± 0.7	7.7 ± 1.2	13.9 ± 3.8*
Skin	3.5 ± 0.5	3.4 ± 0.6	4.0 ± 0.9	4.2 ± 0.4	4.7 ± 1.0

NOTE. Values are means ± SE.

Abbreviations: CNS, central nervous system; HD, hemodilution (number in parentheses is hematcrit level); Hep, heparin.

*$p < 0.05$ *v* baseline.

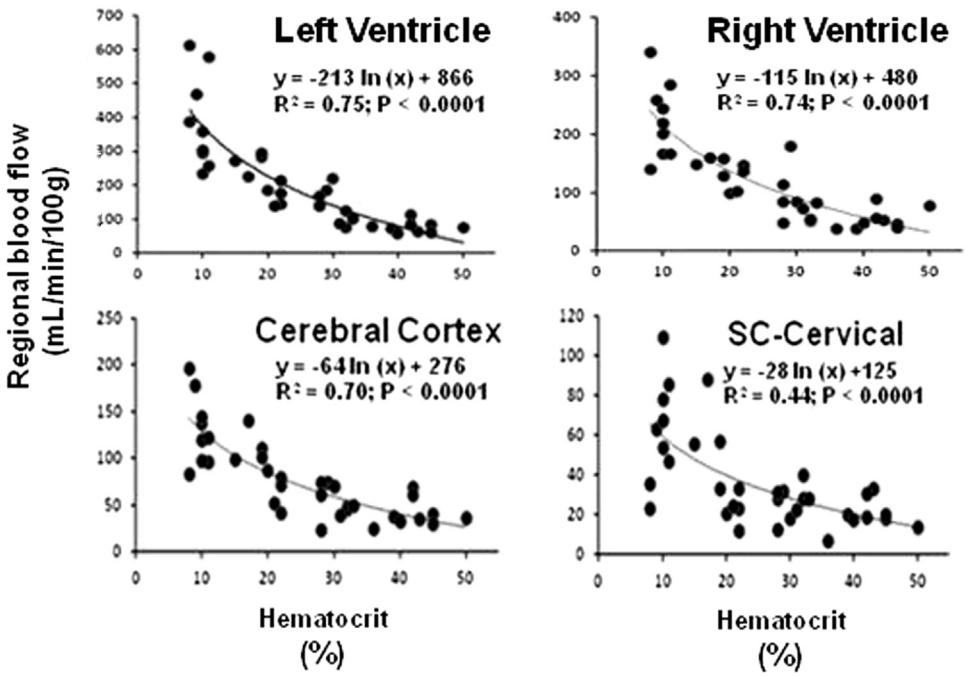

Fig 1. Blood flow to the heart, brain, and spinal cord (SC) had a strong inverse correlation to hematocrit during graded hemodilution.

Table 4. Regional Oxygen Delivery (in mL/min/100 g) During Graded Acute Normovolemic Hemodilution

	Baseline	HD (30)	HD (20)	HD (10)	Cardiac Insufficiency
Heart					
Left Ventricle					
Subepicardium	16.9 ± 1.1	21.0 ± 2.1*	22.9 ± 1.7*	23.5 ± 2.4*	16.9 ± 2.2
Midmural	15.8 ± 1.3	20.0 ± 2.2*	22.2 ± 1.9*	23.1 ± 2.7*	18.2 ± 3.0
Subendocardium	15.5 ± 1.3	19.9 ± 2.4*	21.6 ± 1.8*	20.5 ± 2.2*	16.7 ± 2.2
Mean Transmural	15.9 ± 1.2	20.3 ± 2.2*	22.2 ± 1.7*	22.4 ± 2.4*	17.3 ± 2.7
Right Ventricle					
Subepicardium	12.2 ± 1.6	13.1 ± 2.1	14.1 ± 1.2	13.7 ± 1.3*	11.1 ± 2.4
Subendocardium	10.1 ± 1.5	12.4 ± 2.4	14.5 ± 1.1*	14.5 ± 1.3*	12.4 ± 1.5
Mean Transmural	11.2 ± 1.4	12.7 ± 2.1	14.3 ± 0.9*	14.1 ± 1.2*	11.7 ± 1.9
CNS					
Cerebral Cortex	8.5 ± 1.1	8.1 ± 0.9	9.0 ± 1.0	7.6 ± 0.7	7.3 ± 1.2
Cerebellum	7.9 ± 0.9	7.4 ± 0.5	7.6 ± 0.9	7.0 ± 0.7	6.9 ± 1.0
Pons	4.6 ± 0.7	4.2 ± 0.9	4.7 ± 0.7	4.2 ± 0.6	4.0 ± 0.6
Medulla	4.8 ± 0.4	4.7 ± 0.5	4.8 ± 0.7	4.3 ± 0.5	4.4 ± 0.5
Cervical Spinal Cord	4.2 ± 0.6	4.0 ± 0.5	3.9 ± 0.8	3.7 ± 0.6	3.7 ± 0.7
Thoracic Spinal Cord	2.5 ± 0.5	2.3 ± 0.2	2.1 ± 0.4	2.1 ± 0.2	2.5 ± 0.3
Lumbar Spinal Cord	3.0 ± 0.4	2.6 ± 0.4	2.4 ± 0.3	2.3 ± 0.3	2.8 ± 0.4
Peripheral Organs					
Renal Cortex	92.7 ± 14.3	75.0 ± 13.5*	52.0 ± 8.9*	25.2 ± 4.9*	25.6 ± 7.8*
Liver (Hep. Artery)	6.6 ± 1.5	7.1 ± 1.3	4.7 ± 1.3	3.5 ± 22.0*	3.6 ± 1.2*
Duodenum	8.8 ± 1.2	8.2 ± 1.5	6.8 ± 0.8	4.8 ± 13.0*	4.7 ± 1*
Pancreas	3.4 ± 0.5	3.5 ± 0.8	3.6 ± 0.7	2.0 ± 12.0*	1.7 ± 0.6*
Spleen	30.1 ± 7.0	22.7 ± 5.0	10.9 ± 1.5*	4.0 ± 11.0*	3.6 ± 1.9*
Skeletal Muscle	0.8 ± 0.2	0.5 ± 0.1	0.4 ± 0.1*	0.4 ± 0.1*	0.5 ± 0.2*
Skin	0.7 ± 0.1	0.5 ± 0.1	0.4 ± 0.1*	0.3 ± 0.1*	0.2 ± 0.1*

NOTE. Values are means ± SE.

Abbreviation: HD, hemodilution (number in parentheses is hematcrit level).

*$P < 0.05$ *v* baseline.

(2) These systemic responses were associated with heterogenous changes in regional blood flow and oxygen delivery. The increases in regional blood flow in the myocardium, brain, and spinal cord were sufficient to maintain or even increase oxygen delivery. However, in the other organs evaluated, regional blood flow either did not change (kidney and skin), decreased (spleen), or increased modestly (liver, duodenum, pancreas, skeletal muscle). This ultimately produced decreases in oxygen delivery in all these organs during graded ANH, with that in the kidney occurring at the highest hematocrit and being the most pronounced.

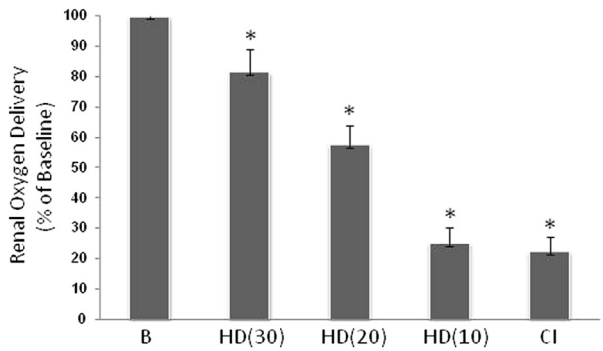

Fig 2. Change in renal oxygen delivery during progressive hemodilution (HD). Values in parentheses represent hematocrit level. CI, cardiac insufficiency. Mean and SE are shown. *p < 0.05 versus baseline (B).

Because of relatively modest increases in cardiac index, systemic oxygen delivery declined during ANH. However, systemic oxygen uptake was maintained by an increase in oxygen extraction, reflecting decreases in both mixed venous hemoglobin saturation and PO_2. This pattern of response has been shown in various animal models[2,4,7,13,14] and in humans.[6,15,16] A reduction in mixed venous PO_2 implies a reduction in average PO_2 in the body tissues.[17] However, arterial lactate concentration remained unchanged across a wide hematocrit range of 40% to 10%, suggesting the absence of widespread anaerobic metabolism. The decline in oxygen delivery to the kidney, splanchnic organs, skeletal muscle, and skin suggested that these organs may be the sites of compensatory increases in oxygen extraction, although previous physiologic studies suggested that the involvement of this mechanism may vary from organ to organ (see below).

A hematocrit of 7 ± 1% was associated with an increase in arterial lactate concentration, and with cardiac insufficiency as evidenced by an increased pulmonary capillary wedge pressure and a decreased arterial pressure. This value for critical hematocrit, as well as the accompanying values for systemic oxygen extraction, mixed venous hemoglobin saturation, and mixed venous PO_2, were similar to those reported previously.[2,12,18,19]

Two factors may decrease regional vascular resistance, and, thus, increase regional blood flow during ANH. These factors are: (1) vascular relaxation in response to reduced arterial oxygen content, and (2) decreased blood viscosity.[7] The limited blood flow responses during ANH in the kidney, splanchnic

organs, skeletal muscle, and skin are consistent with a countervailing vasoconstrictor mechanism. A role for an arterial chemoreceptor/ sympathetic vasoconstrictor nerve pathway has been suggested.[7,20,21] The engagement of this pathway would support aortic pressure, while redistributing blood flow towards the vital organs, the heart and brain.

Under baseline conditions, the left ventricle extracts approximately 70%-75% of the oxygen that is delivered to it; thus, it has a limited oxygen extraction reserve.[22] Consequently, during ANH the myocardium is critically dependent on increases in blood flow to offset the decreases in arterial oxygen content. The current findings confirmed previous studies[2,12] indicating a remarkable capacity of the coronary circulation to increase its blood flow during ANH. Blood flow was increased nearly 400% at a hematocrit of 10%, and was sufficient to maintain oxygen delivery and contractile function. The increased oxygen delivery to the left ventricle during ANH reflected a greater oxygen demand, which was attributable, at least in part, to the elevated heart rate. The increases in myocardial blood flow during ANH were accompanied by a reduction in the vasodilator reserve ratio (assessed from reactive hyperemic responses), implying a prominent role for coronary vasodilation in the hemodilution-induced blood flow increases.[2,12]

Previous studies have demonstrated that blood flow to the brain increases progressively during graded ANH.[7,23] The current results extended this response to regions within the brain (cerebral cortex, cerebellum, pons, medulla), and spinal cord (cervical, thoracic, lumbar regions). In all of these tissues, regional blood flow increased sufficiently to maintain regional oxygen delivery. A recent study demonstrated that hypercapnia-induced increases in regional blood flow were blunted in the brain and spinal cord during ANH, which suggested that vasodilation was integral to the increases in blood flow during ANH.[24]

Because of inadequate blood flow responses, oxygen delivery eventually decreased in the kidney, splanchnic organs, skeletal muscle, and skin during graded ANH. The decrease in oxygen delivery appeared first in the kidney after only a reduction in hematocrit to 30%. A subsequent reduction in hematocrit to the "critical level" produced a decrease in renal oxygen delivery to approximately one-fourth of the baseline value. A susceptibility of the kidney to reductions in oxygen delivery during ANH was consistent with findings in dogs anesthetized with pentobarbital[7,8] or halothane.[25]

A reduction in oxygen delivery during ANH will not result in a decrease in oxygen uptake if it is accompanied by a proportional increase in oxygen extraction. Because of a primary function in filtering the blood, renal blood flow (and oxygen delivery) is normally well in excess of that required to meet its basal oxygen demand. This results in a very modest baseline oxygen extraction of less than 10%.[26] Thus, it generally has been assumed that increases in oxygen extraction would occur to offset the decreases in renal oxygen delivery during ANH.[7,8] However, previous experimental evidence raised serious questions about this assumption. First, a canine study demonstrated that oxygen uptake of the kidney varied directly with induced decreases in blood flow, implying no increases in oxygen extraction.[27] Furthermore, a recent rat study showed that both renal tissue PO_2 and oxygen uptake declined in parallel with progressive decreases in hematocrit.[28] The inability of the kidney to increase oxygen extraction has been explained by a diffusive shunt for oxygen made possible by the interlobar arteries lying in close proximity to the interlobular veins.[29] This oxygen bypasses the tissue and is responsible for the disparity between renal venous PO_2 and microvascular PO_2, which widens when hematocrit is lowered.[28] The current findings, when viewed in the context of this previous laboratory work, were highly suggestive of renal hypoxia during ANH. However, this conclusion must remain speculative until confirmed by further studies demonstrating decreases in renal oxygen uptake or renal lactate production. A decline in renal function or the appearance of a biomarker of renal injury, eg, urinary neutrophil gelatinase-associated lipocaline,[30] would provide indirect evidence for inadequate renal oxygenation during ANH. Several clinical studies have suggested a vulnerability of hemodiluted patients to renal dysfunction and injury.[31–33]

Animal studies have demonstrated that the liver[34,35] and skeletal muscle[36] have appreciable oxygen extraction reserves that can be recruited during ANH, but that this capability may be more limited in the intestine.[37–39] The impediment to oxygen extraction in the intestine occurs because of anatomic features of its microcirculation, which promote convective shunting of blood, countercurrent shunting of oxygen, and plasma skimming of the blood supplying the intestinal mucosa.[37,39,40] Although blood flow to resting skeletal muscle is low on a per-weight basis, skeletal muscle constitutes more than 40% of body mass, and, thus, in the aggregate, it likely made a large contribution to the increase in systemic oxygen extraction observed during ANH.

The open-chest dogs of the current study were ventilated with oxygen to ensure that hemoglobin in arterial blood remained well saturated. The resultant arterial PO_2 values were approximately 300 mmHg, indicating that the dogs were hyperoxic. Because the amount of oxygen dissolved in the plasma remained elevated during ANH, this component represented a greater portion of the total oxygen content of the arterial blood. Whether hyperoxia extended the safe range of hematocrit reduction in the current study is an open question.

The extended duration of the dextran-for-blood exchange introduced the possibility that deterioration of the preparation affected the findings. This possibility was refuted by previous results from a canine study indicating that systemic hemodynamic variables and myocardial blood flow and oxygen consumption were constant during an equally prolonged blood-for-blood exchange.[12]

The current findings pertained strictly to healthy, instrumented, and open-chest dogs and therefore should not be extrapolated directly to human patients. Isoflurane has been demonstrated to have direct negative inotropic and vasodilating effects[41,42] and to cause depression of the sympathetic nervous system[43,44] actions, which could have altered the regional blood flow responses during ANH. Dextran-40 was used as the plasma expander in the current study because it was economical and tended to remain in the intravascular space, which promoted maintenance of isovolemic conditions.[1] Furthermore, low-molecular-weight dextran was used in previous relevant studies,[2] which facilitated comparisons. The authors could not exclude the possibility that results may have differed

if another anesthetic or a diluent with a different ionic composition or viscosity, such as saline or plasma, was used.

In 1999, the Transfusion Requirement in Critical Care trial demonstrated that implementation of a restrictive transfusion protocol to a transfusion trigger of hemoglobin below 7 g/dL reduced the range of allogeneic blood transfusion by 33% without increases in mortality rates and hospital length of stay.[45] On the basis of these findings and those in subsequent studies, a transfusion trigger of 7 g/dL was proposed and is now widely accepted.[46] It is noteworthy that the authors observed a decrease in renal oxygen delivery at a hematocrit of 30%, a value that exceeds this transfusion trigger. A recent multicenter pilot trial suggested that a conservative transfusion trigger may not be safe in patients with symptomatic coronary artery disease.[47] These findings were consistent with animal data demonstrating an enhanced vulnerability to myocardial ischemia and cardiac dysfunction during ANH when coronary reserve was compromised.[12]

It has been proposed that a marker of systemic oxygen supply/demand balance, such as mixed venous or central venous oxygen saturation, be used as a physiologic transfusion trigger.[48] A shortcoming of this approach is that it would be insensitive to inadequate oxygen delivery in especially vulnerable organ systems, such as the kidney. The adequacy of renal oxygenation may be a limiting factor in how low hematocrit can be reduced safely during ANH in the surgical patient.

In conclusion, the current findings suggested that the heart and the brain (including the spinal cord) were the most adaptive organs and the kidney was the least adaptive organ during ANH. On the basis of what is currently known, it seems prudent to consider the adequacy of renal oxygen delivery, perhaps as reflected in indices of renal function, eg, serum creatinine and blood urea nitrogen,[49] in deciding whether a blood transfusion is necessary in a hemodiluted patient, especially one with pre-existing renal disease.

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