Venous Blood Pressure in Broilers During Acute Inhalation of Five Percent Carbon Dioxide or Unilateral Pulmonary Artery Occlusion

ROBERT F. WIDEMAN, JR.,[1] PHILIP MAYNARD, and WALTER G. BOTTJE

Department of Poultry Science, University of Arkansas, Fayetteville, Arkansas 72701

ABSTRACT We evaluated the hypothesis that venous congestion (increased venous volume), as reflected by venous hypertension (increased venous pressure), can arise when the right ventricle is unable to elevate the pulmonary arterial pressure sufficiently to propel the cardiac output through an anatomically inadequate or inappropriately constricted pulmonary vasculature. Changes in venous pressure were evaluated in clinically healthy broilers during modest increases in pulmonary vascular resistance induced by inhalation of 5% CO_2 and during large increases in pulmonary vascular resistance accomplished by acutely tightening a snare around one pulmonary artery. Inhalation of 5% CO_2 induced a pronounced respiratory acidosis, as reflected by increases the partial pressure of CO_2 and the hydrogen ion concentration in arterial blood. Inhalation of 5% CO_2 also increased pulmonary arterial pressure by approximately 3 mm Hg and increased venous pressure by approximately 1 mm Hg when compared with the pre-inhalation venous pressure. Tightening the pulmonary artery snare increased the pulmonary arterial pressure by approximately 10 mm Hg, and this degree of pulmonary hypertension was sustained until the snare was released. When compared with the pre- and post-snare intervals, tightening of the pulmonary artery snare induced a sustained increase in venous pressure of ≥1 mm Hg. Veins have highly compliant walls that permit an approximate doubling in volume with only small (4 to 6 mm Hg) increases in central venous pressure. Presumably the apparently modest 1 mm Hg increase in venous pressure measured after CO_2 inhalation or unilateral pulmonary artery occlusion reflects a large increase in venous volume and, thus, substantial venous congestion. These observations support the hypothesis that increases in pulmonary vascular resistance can initiate increases in venous pressure by challenging the capacity of the right ventricle to propel all of the returning venous blood through the lungs. Central venous congestion predisposes broilers to the onset of cirrhosis and ascites by impeding the outflow of hepatic venous blood and increasing the hydrostatic pressure within hepatic sinusoids.

(*Key words*: pulmonary hypertension, respiratory acidosis, pulmonary arterial restriction, broilers)

1999 Poultry Science 78:1443–1451

INTRODUCTION

In formulating a hypothesis to explain the pathophysiological progression leading from the initiation of pulmonary hypertension (elevated pulmonary arterial pressure) to the development of terminal ascites in broiler chickens, it has been proposed that an increase in central venous pressure occurs as a secondary consequence of right-sided congestive heart failure. According to this hypothesis, venous congestion (increased venous volume) and hypertension (increased venous pressure) must arise whenever the right ventricle cannot propel all of the returning blood through an anatomically inadequate or inappropriately constricted pulmonary vasculature. Blood that cannot readily be pumped through the lungs must accumulate within the chamber of the right ventricle (increases in end systolic ventricular volume cause ventricular dilation) and in the systemic veins (venous congestion). As the right ventricle progressively dilates and undergoes work hypertrophy in the process of sustaining an elevated pressure, the right atrio-ventricular valve eventually becomes distorted and rendered incompetent. The ensuing valvular regurgitation should partially attenuate further right ventricular dilation and may serve as a relief mechanism to protect the noncompliant pulmonary vasculature from exposure to excessive hydrostatic pressures. However, valvular regurgitation would further exacerbate the incipient descending venous congestion and hypertension, thereby predisposing broilers to the onset of cirrhosis and ascites by impeding the outflow of hepatic venous blood and increasing the hydrostatic pressure within hepatic sinusoids (Wideman and Bottje, 1993; Wideman 1997). Indeed, when large increments in pulmonary vascular resistance are induced experimentally in

Received for publication March 8, 1999.
Accepted for publication June 18, 1999.
[1]To whom correspondence should be addressed: rwideman@comp.uark.edu

Abbreviation Key: HCT = hematocrit.

broilers, the maximum pulmonary arterial pressure acutely attainable by the right ventricle is inadequate to propel all of the returning venous blood through the lungs. Consequently, the impeded venous return to the left ventricle causes dependent reductions in stroke volume, cardiac output, and mean systemic arterial pressure, and the cumulative hemodynamic perturbations rapidly evolve into ascites when sustained chronically (Wideman and Kirby, 1995a,b, 1996; Wideman *et al.*, 1996a,b, 1997, 1998a,b, 1999; Wideman, 1999). The present study was designed to evaluate changes in venous pressure during modest increases in pulmonary vascular resistance induced by inhalation of 5% CO_2 and during large increases in pulmonary vascular resistance accomplished by acutely tightening a snare around one pulmonary artery. Changes in venous pressure had not previously been measured in relation to the onset of pulmonary hypertension, although broilers with terminal ascites have obviously distended systemic veins (Wideman and Bottje, 1993). Venous congestion also had been evaluated as a potential seasonal factor contributing to bleedout problems during the processing of broilers grown under cool temperatures that increase the incidence of ascites (Kranen *et al.*, 1998).

MATERIALS AND METHODS

Male broiler chicks were reared on fresh wood shavings litter in an environmental chamber (8 m^2 floor space). They were brooded at 32 and 30 C during Weeks 1 and 2, respectively, and, thereafter, the temperature was maintained at 24 C. They were fed a 23% CP corn-soybean meal-based broiler ration formulated to meet or exceed the minimum NRC (1984) standards for all ingredients. Feed and water were provided for *ad libitum* consumption.

Experiment 1

Sixteen broilers, 35 to 42 d of age, were prepared using a surgical protocol described previously (Wideman and Kirby, 1995b; Wideman *et al.*, 1996a,b, 1998a,b). Clinically healthy males were anesthetized to a light surgical plane with intramuscular injections of allobarbital (5,5-diallylbarbituric acid; 25 mg/kg BW), fastened in dorsal recumbency, and the thoracic inlet was opened. A Transonic ultrasonic flowprobe[2] was positioned on the left pulmonary artery, the probe was connected to a Transonic T206 blood flow meter[2] to confirm signal acquisition; then, the skin of the thoracic inlet was sealed with surgical wound clips. Silastic® tubing[3] filled with heparinized saline was inserted through the left ulnar vein, and was advanced into the right pulmonary artery as judged by pressure tracings (Guthrie *et al.*, 1987; Owen *et al.*, 1995). The distal end of the cannula was attached to a BLPR blood pressure transducer[4] interfaced through a Transbridge™ preamplifier[4] to a Biopac MP 100 data acquisition system using AcqKnowledge software[5]. The left brachial artery, the right ulnar vein, and an anterior tibial vein were cannulated with PE-50 polyethylene tubing filled with heparinized saline. The arterial cannula was advanced to a position near the descending aorta and was attached to a blood pressure transducer for continuous monitoring of systemic arterial pressure. The cannula inserted in the right ulnar vein was attached to a blood pressure transducer for continuous monitoring of venous pressure. The cannula was advanced past the proximal (shoulder) joint of the humerus bone and into the right subclavian vein or cranial vena cava as judged by the length of the inserted cannula and characteristic pulsatile pressure tracings when the cannula approached the right atrium. This cannula was not advanced into the sinus venosus or right atrium to prevent interference with the sino-atrial and atrio-ventricular valves, respectively. Pressure transducers attached to the venous and pulmonary arterial cannulae were positioned level with the heart. Throughout each experiment, 2.5% mannitol was infused through the anterior tibial cannula at a constant rate of 0.1 mL/min per kilogram BW to hydrate the birds.

When surgical preparations were complete and a stabilization period of 30 min had elapsed, control data were collected for 20 min, and two arterial blood samples were collected 5 and 15 min into the control period (Samples A and B). A polyethylene inhalation mask was positioned to cover the front of the bird's head, directing incoming gas toward the beak and nostrils and allowing ample open space for excess gas to escape freely around the back of the head. Gas[6] consisting of compressed air alone (control group; n = 7) or compressed air containing 5% CO_2 (CO_2 group; n = 9) was released through the inhalation mask at 8 psi at a rate of approximately 12 L/min for 17 min, and two arterial blood samples were collected 5 and 15 min into the gas inhalation period (Samples C and D). The gas flow was shut off, the inhalation mask was removed, and two arterial samples were collected 10 and 20 min into the recovery period (Samples E and F). Arterial blood samples (1 mL each) were withdrawn anaerobically and were injected within 30 s into a Radiometer ABL 330 Acid-Base Laboratory[7]. Appropriate function of the blood gas analyzer was assessed by periodically injecting Blood Gas Qualicheck® reference standards[7]. The primary arterial blood values for pH, Pa_{O2}, and Pa_{CO2} were generated by the ABL330 operating at a sample chamber temperature of 37 C and were recalculated by the ABL330 for a temperature of 41 C to match the normal body temperature of domestic fowl (Fedde, 1986). A portion of the arterial blood was used for duplicate hematocrit (HCT) determinations using heparinized capillary tubes and a microhematocrit centrifuge. Birds were assigned to the control or CO_2 groups in a random order. At the end of the experiment, they were killed with a 10-

[2]Transonic Systems Inc., Ithaca, NY 14850.
[3]Dow Corning Corp., Midland, MI 48686-0994.
[4]World Precision Instruments, Sarasota, FL 34230.
[5]Biopac Systems, Inc., Goleta, CA 93117.
[6]Air Products, Inc., Fayetteville, AR 72701.
[7]Radiometer America Inc., Westlake, OH 44145.

mL i.v. injection of 0.1 *M* KCl. Necropsies were not conducted.

The Biopac MP 100 data acquisition system recorded four primary data channels, including systemic arterial pressure in millimeters of mercury (mm Hg), pulmonary arterial pressure (mm Hg), venous pressure (mm Hg), and blood flow through the left pulmonary artery (mL/min). Average values for these parameters were measured electronically during representative intervals immediately preceding (sample intervals A1, B1, C1, etc.) and following (sample intervals A2, B2, C2, etc.) withdrawal of each arterial blood sample. Values were similarly averaged over a 15- to 20-s interval coincident with the maximum (peak) pulmonary arterial pressure attained within the first 5 min after the start of the gas inhalation period. The protocol used for data averaging previously was demonstrated to compensate accurately for the influences of pulse pressure and respiratory cycles on pulmonary and systemic arterial pressures (Wideman *et al.*, 1996a,b). These primary peak and pre- and post-blood sampling values were used to calculate cardiac output, stroke volume, pulmonary vascular resistance, and total peripheral resistance. Based on the assumption that cardiac output (mL/min) normally is divided approximately equally between two lungs of equal size, cardiac output was calculated as 2× blood flow. The cardiac output is the product of heart rate × stroke volume (mL/beat); consequently, stroke volume was calculated as cardiac output divided by heart rate. Heart rate (beats/min) was obtained by counting systolic peaks over time in the pulmonary arterial pressure recording, coincident with each sample interval. Assuming the pressure gradients across the pulmonary and systemic circulations are essentially equal to pulmonary arterial pressure and systemic arterial pressure, respectively (Wideman *et al.*, 1996a,b, 1998a,b), then the relationships among pressure gradients, flow rates, and resistances are summarized by the respective equations: pulmonary arterial pressure = cardiac output × pulmonary vascular resistance, and mean systemic arterial pressure = cardiac output × total peripheral resistance. Thus, pulmonary vascular resistance was calculated in relative resistance units as pulmonary arterial pressure (mm Hg) divided by cardiac output (mL/min), and total peripheral resistance was calculated in relative resistance units as mean systemic arterial pressure (mm Hg) divided by cardiac output (mL/min) (Besch and Kadono, 1978; Sturkie, 1986; Wideman *et al.*, 1996a,b, 1998a,b). Respiratory rate (breaths/min) was obtained by counting the wave cycles associated with respiratory movement that constitute an integral part of the pulmonary arterial pressure recordings (Sturkie, 1986).

Experiment 2

Seven male broilers, 36 to 39 d of age (2,371 ± 48 g BW; mean ± SEM), were anesthetized and surgically prepared as described for Experiment 1, with the exception that a snare was placed loosely around the left pulmonary artery as described previously (Wideman and Kirby, 1995b; Wideman *et al.*, 1996a,b, 1998a) and no flowprobe was used. The Biopac MP 100 data acquisition system recorded three primary data channels including systemic arterial pressure (mm Hg), pulmonary arterial pressure (mm Hg), and venous pressure (mm Hg). Following a stabilization period of 30 min, average values for these parameters were measured electronically at 5-min intervals during a 20-min control period (sample intervals A to D). The snare was tightened to occlude the left pulmonary artery fully, and average values were measured coincident with the maximum pulmonary arterial pressure attained during the first 5 min (peak) and at 3-min intervals thereafter (sample intervals E to H). The snare was released, and average values were measured beginning 10 min into a 30-min recovery period (sample intervals I to L).

Statistical Analysis

Data were analyzed across sample intervals within a group using the SigmaStat® Repeated Measures Analysis of Variance procedure, and means were differentiated by the Student-Newman-Keuls method (Jandel Scientific, 1994). Within a single sample interval across groups, the SigmaStat® T test was used to assess differences ($P < 0.05$) among means. The SigmaStat® linear regression procedure was used to evaluate relationships among cardiopulmonary variables for Experiment 1.

RESULTS

Experiment 1

The control and CO_2 groups did not differ in BW (2,468 ± 54 g *vs* 2,116 ± 154 g, respectively), nor were differences detected between the groups for comparisons of initial arterial blood values (sample intervals A and B; Figure 1; Table 1) or initial cardiopulmonary variables (sample intervals A1 to B2; Figures 2 to 4; Table 1). Inhalation of 5% CO_2 increased the partial pressure of CO_2, the hydrogen ion concentration, and the partial pressure of O_2 in the arterial blood of the CO_2 group (Figure 1). The blood values for the control group remained unchanged throughout the course of the experiment, and, in the CO_2 group, the respiratory rate, HCT, bicarbonate concentration of arterial blood, and saturation of hemoglobin with oxygen in arterial blood remained unchanged (Table 1). Tidal volume was not quantified in this study. Visual evaluations of sternal excursions failed to detect consistent changes in amplitude during the period of 5% CO_2 inhalation.

Pulmonary arterial pressure gradually declined over the course of the experiment in the control and CO_2 groups, and exposure to compressed air alone did not affect pulmonary arterial pressure, pulmonary vascular resistance, or venous pressure in the control group (Figure 2). Inhalation of 5% CO_2 increased pulmonary arterial pressure by approximately 3 mm Hg during the peak and C1 sample intervals when compared with sample

TABLE 1. Consecutive physiological values during inhalation of ambient air (sample intervals A and B), during inhalation of compressed air or 5% CO_2 (control and CO_2 groups, respectively; sample intervals C and D), and after the return to inhalation of ambient air (sample intervals E and F), Experiment 1[1]

		Experimental protocol (sample intervals)					
		Ambient air		Air or 5% CO_2		Ambient air	
Variable	Group	A	B	C	D	E	F
Respiratory rate, breaths/min	Control	47.1 ± 2.1	46.1 ± 1.9	47.1 ± 1.4	47.1 ± 1.5	50.0 ± 2.1	50.0 ± 2.1
	CO_2	50.8 ± 1.8	50.8 ± 1.8	49.2 ± 2.4	48.5 ± 2.7	40.7 ± 2.3	47.8 ± 2.5
Hematocrit, %	Control	30.7 ± 0.9	30.1 ± 0.9	29.1 ± 0.8	28.0 ± 0.7	26.7 ± 0.9	26.0 ± 0.9
	CO_2	30.8 ± 0.7	30.1 ± 0.8	29.0 ± 0.7	27.9 ± 0.8	27.5 ± 0.8	26.8 ± 0.8
Bicarbonate, m*M*	Control	23.0 ± 0.6	23.1 ± 0.5	23.0 ± 0.5	22.9 ± 0.6	22.5 ± 0.7	22.8 ± 0.8
	CO_2	24.3 ± 0.6	24.0 ± 0.6	24.2 ± 0.5	24.2 ± 0.5	23.8 ± 0.7	22.8 ± 0.7
Hemoglobin O_2 saturation, %	Control	95.4 ± 0.6	94.6 ± 1.4	94.9 ± 1.1	94.3 ± 1.8	95.0 ± 1.5	95.7 ± 1.2
	CO_2	94.7 ± 0.6	94.8 ± 0.6	94.8 ± 0.5	93.0 ± 2.1	94.1 ± 2.1	93.3 ± 2.5

[1]Data are means ± SEM; n = 7 for control group; n = 9 for CO_2 group.

intervals A2 and B2 for the CO_2 group and when compared with contemporaneous peak and C1 sample intervals for the control group (Figure 2). Pulmonary vascular resistance during 5% CO_2 inhalation was not elevated above the initial values for the CO_2 group nor above the contemporaneous values for the control group. The 5% CO_2 inhalation increased the venous pressure by approximately 1 mm Hg during the peak, C1, and D1 sample intervals when compared with sample intervals A1 to B2 for the CO_2 group but not when compared with the contemporaneous peak, C1, and D1 sample intervals for the control group (Figure 2).

As shown in Figure 3, inhalation of 5% CO_2 increased cardiac output during sample interval D1 when compared with sample intervals A2 and B2 for the CO_2 group but not when compared with sample interval D1 for the control group. Stroke volume increased during sample interval D1 when compared with sample interval B1 in the CO_2 group but not when compared with sample interval D1 of the control group. Throughout the experiment, cardiac output and stroke volume remained unchanged in the control group, and heart rate was unchanged in both groups (Figure 3). Mean arterial pressure gradually declined over the course of the experiment in both groups and was higher in the CO_2 group than in the control group during sample intervals D1 and D2 (Figure 4). Total peripheral resistance did not change over time nor did differences exist ($P = 0.088$ for sample interval E2) between the groups (Figure 4).

Linear regression analysis was used to evaluate the possibility that inhalation of 5% CO_2 triggered subtle relationships among the cardiopulmonary variables over sample intervals A1 through D2 in the CO_2 group (Table 2). Increases in pulmonary arterial pressure were correlated with increases in pulmonary vascular resistance but not with increases in cardiac output. Increases in cardiac output were correlated with increases in stroke volume and not in heart rate. Venous pressure was positively correlated with pulmonary arterial pressure, cardiac output, and stroke volume but not with pulmonary vascular resistance (Table 2).

Experiment 2

The hemodynamic responses of a representative individual broiler to tightening the snare around the left pulmonary artery are shown in Figure 5. The large 4- to 5-mm Hg amplitude excursions with a frequency of approximately 0.8/s in the pulmonary arterial pressure and venous pressure recordings are the wave cycles associated with respiration (respiratory rate = 48 per min), whereas the superimposed smaller amplitude fluctuations with a frequency of approximately 6/s are the systolic and diastolic pressures associated with each heart beat (heart rate = 360 beats per min). Within 10 s after the snare was tightened, the mean arterial pressure decreased from 103 to 84 mm Hg, the mean value for pulmonary arterial pressure increased from 19 to 29 mm Hg, and the mean value for venous pressure increased from 2.6 to 3.6 mm Hg. Taking into account the respiratory waves in the venous pressure recording over the same measurement interval (1,240 *vs* 1,255 s), tightening the snare increased the minimum diastolic value at the trough of expiration from 0.54 to 2.1 mm Hg and increased the maximum systolic pressure at the peak of inspiration from 4.8 to 5.4 mm Hg (Figure 5). For the group as a whole, tightening the snare around the left pulmonary artery increased the pulmonary arterial pressure by approximately 10 mm Hg, and this degree of pulmonary hypertension was sustained until the snare was released (Figure 6). In comparison with the respective pre- and post-snare sample intervals, tightening the pulmonary artery snare produced a sustained increase in venous pressure of ≥1 mm Hg and a sustained decrease in mean arterial pressure of ≤30 mm Hg (Figure 6).

DISCUSSION

Respiratory acidosis induced by ventilation with CO_2 and metabolic acidosis induced by gradual acid infusion tend to increase pulmonary vascular resistance and pulmonary arterial pressure in mammals modestly, particularly when the starting pH of the blood or lung perfusate

is within a neutral to alkaline range of ≥7.40 (Barer *et al.*, 1967; Farrukh *et al.*, 1989; Brimioulle *et al.*, 1990; Marshall and Marshall, 1992). Evidence of an analogous modest pulmonary hypertensive response to metabolic acidosis has been reported previously for broiler chickens (Owen *et al.*, 1994; Wideman *et al.*, 1998b). In the present study, 5% CO_2 inhalation induced a pronounced respiratory acidosis (reduced pH and increased partial pressure of CO_2 in arterial blood) that triggered a modest and transient increase in pulmonary arterial pressure. Regression analysis, supported by comparing the time course of changes in group means, indicated that the higher pulmonary

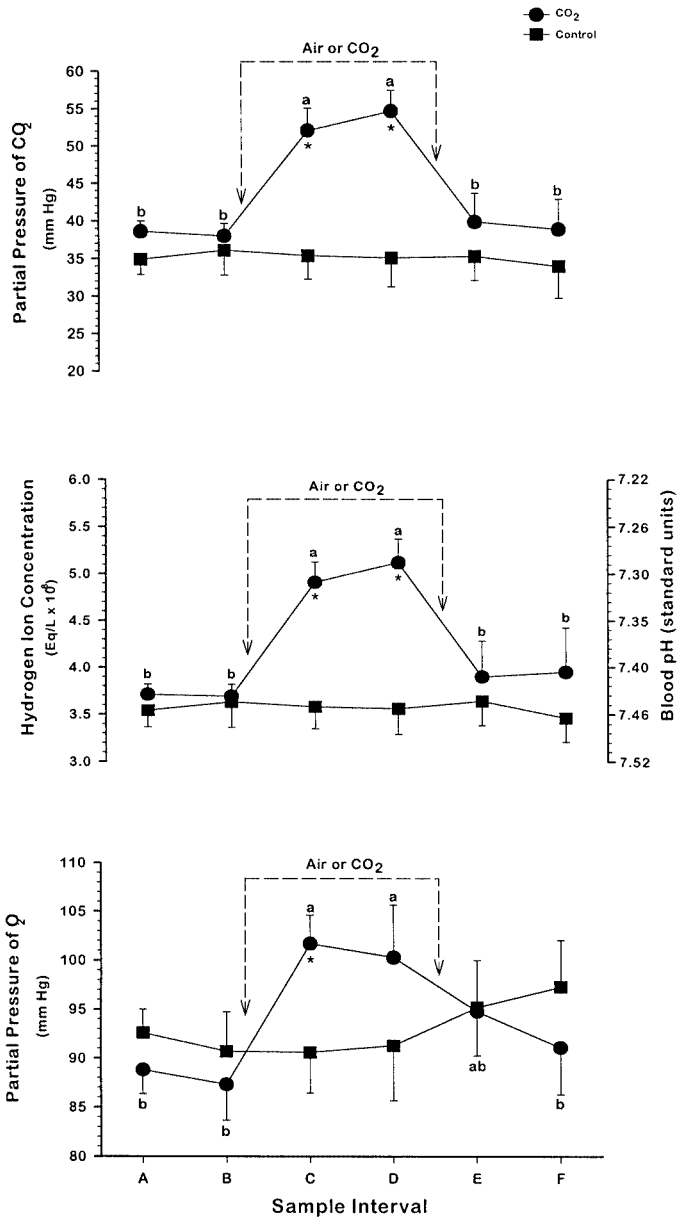

FIGURE 1. Experiment 1: Partial pressure of CO_2 (upper panel), hydrogen ion concentration (middle panel), and partial pressure of O_2 (lower panel) in the arterial blood of control (squares; mean ± SEM; n = 7) and CO_2 groups (circles; mean ± SEM; n = 9) during an initial 20-min period when all broilers breathed ambient air (sample intervals A and B), during a 17-min period when either compressed air (control group) or compressed air containing 5% CO_2 (CO_2 group) was supplied (sample intervals C and D), and during a 30-min recovery period when all broilers again breathed ambient air (sample intervals E and F). Different letters (a,b) designate differences ($P \leq 0.05$) within the CO_2 group over time. Asterisks (*) designate differences ($P \leq 0.05$) between groups within a sample interval.

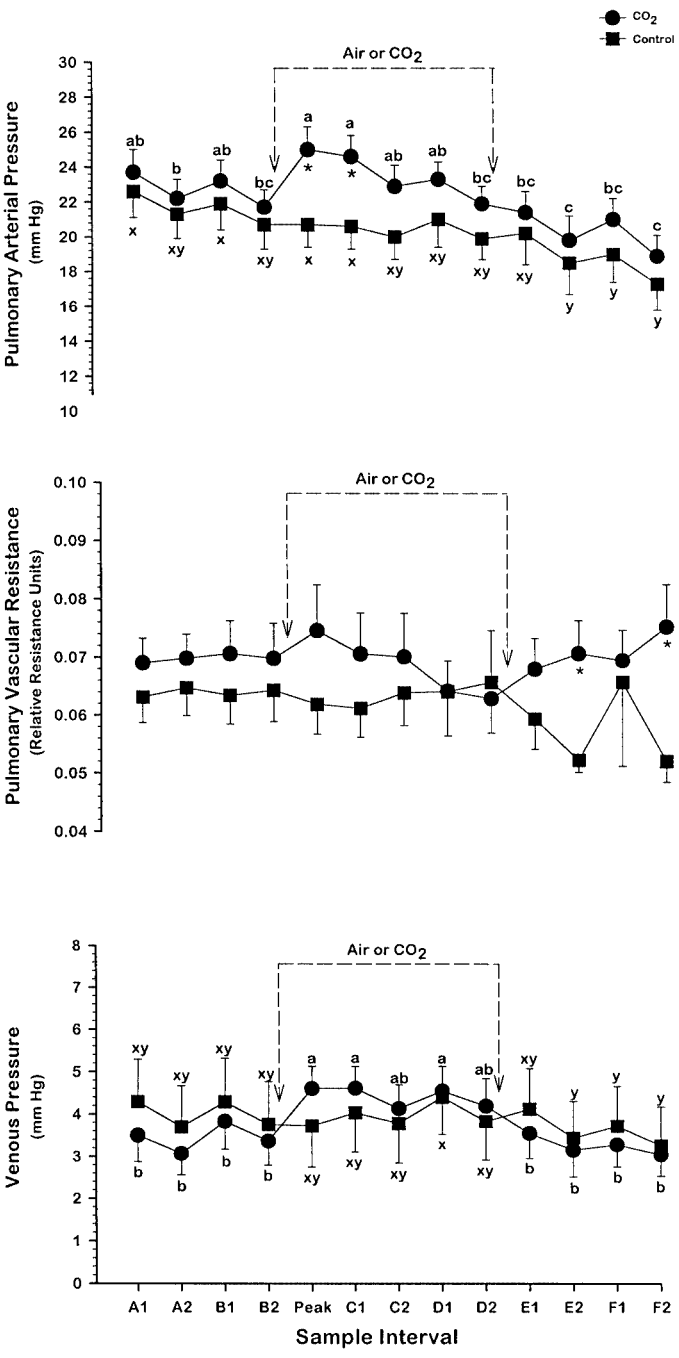

FIGURE 2. Experiment 1: Pulmonary arterial pressure (upper panel), pulmonary vascular resistance (middle panel), and venous pressure (lower panel) for control (squares; mean ± SEM; n = 7) and CO_2 groups (circles; mean ± SEM; n = 9) during an initial 20-min period when all broilers breathed ambient air (sample intervals A1 to B2), during the maximum increase in pulmonary arterial pressure attained within the first 5 min after the start of gas inhalation (peak), during a 17-min period when either compressed air (control group) or compressed air containing 5% CO_2 (CO_2 group) was supplied (sample intervals C1 to D2), and during a 30-min recovery period when all broilers again breathed ambient air (sample intervals E1 and F2). Different letters (x,y for control group; a,b,c for CO_2 group) designate differences ($P \leq 0.05$) within a group over time. Asterisks (*) designate differences ($P \leq 0.05$) between groups within a sample interval.

arterial pressure during the first two sample intervals after the start of CO_2 inhalation was primarily associated with increases in pulmonary vascular resistance rather than with increases in cardiac output. Cardiac output tended to increase later during the CO_2 inhalation period,

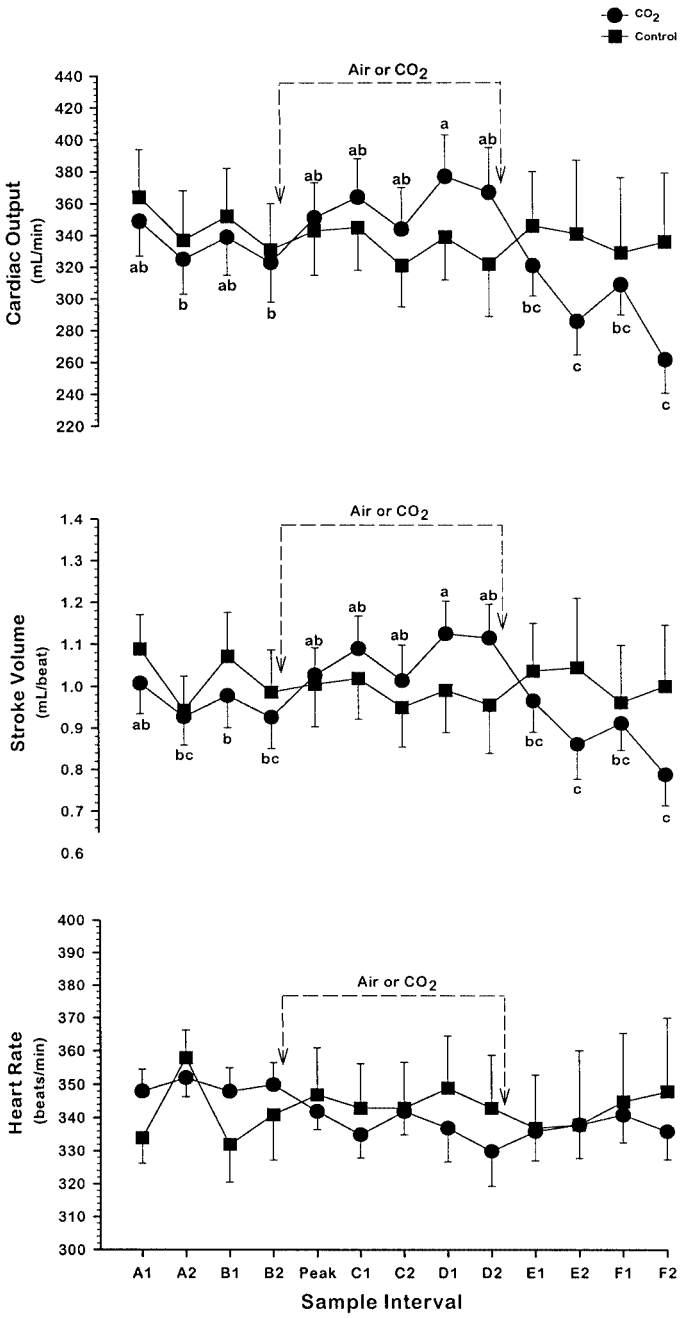

FIGURE 3. Experiment 1: Cardiac output (upper panel), stroke volume (middle panel), and heart rate (lower panel) for control (squares; mean ± SEM; n = 7) and CO_2 groups (circles; mean ± SEM; n = 9) during an initial 20-min period when all broilers breathed ambient air (sample intervals A1 to B2), during the maximum increase in pulmonary arterial pressure attained within the first 5 min after the start of gas inhalation (peak), during a 17-min period when either compressed air (control group) or compressed air containing 5% CO_2 (CO_2 group) was supplied (sample intervals C1 to D2), and during a 30-min recovery period when all broilers again breathed ambient air (sample intervals E1 and F2). Different letters (a,b,c) designate differences ($P \leq 0.05$) within the CO_2 group over time. Asterisks (*) designate differences ($P \leq 0.05$) between groups within a sample interval.

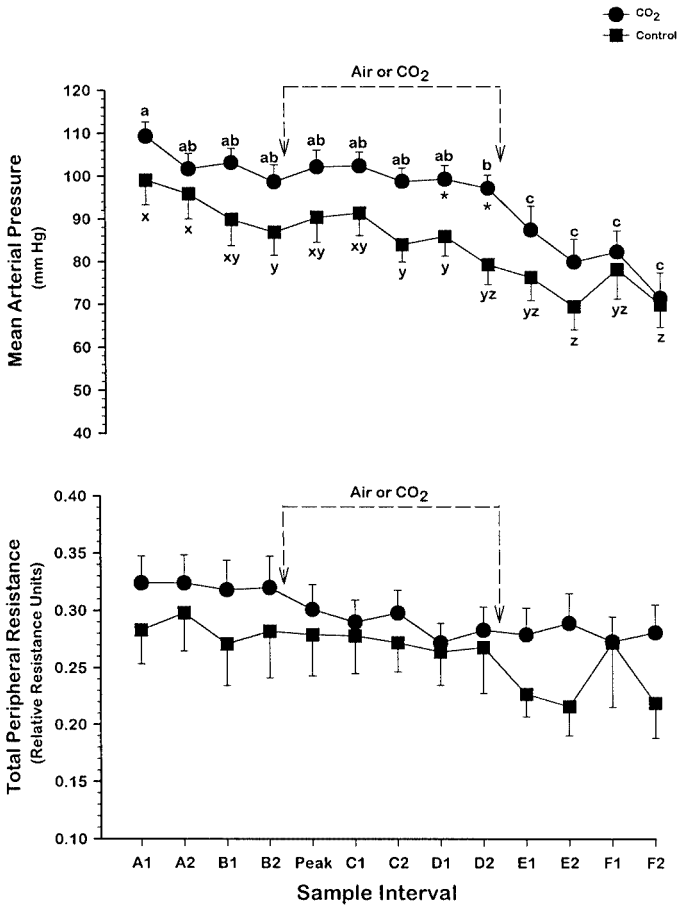

FIGURE 4. Experiment 1: Mean systemic arterial pressure (upper panel) and total peripheral resistance (lower panel) for control (squares; mean ± SEM; n = 7) and CO_2 groups (circles; mean ± SEM; n = 9) during an initial 20-min period when all broilers breathed ambient air (sample intervals A1 to B2), during the maximum increase in pulmonary arterial pressure attained within the first 5 min after the start of gas inhalation (peak), during a 17-min period when either compressed air (control group) or compressed air containing 5% CO_2 (CO_2 group) was supplied (sample intervals C1 to D2), and during a 30-min recovery period when all broilers again breathed ambient air (sample intervals E1 and F2). Different letters (x,y,z for control group; a,b,c for CO_2 group) designate differences ($P \leq 0.05$) within a group over time. Asterisks (*) designate differences ($P \leq 0.05$) between groups within a sample interval.

after the initial pulmonary hypertensive response began to subside. Applying Poiseuille's relationship (Sturkie, 1986) to the pulmonary vasculature, small increments in resistance should more readily contribute to the onset of pulmonary hypertension than modest increases in cardiac output (Wideman, 1999). As predicted based on hemodynamic considerations (see Introduction), venous pressure in the vicinity of the right atrium was elevated modestly but consistently in response to the increase in afterload (increased pulmonary arterial pressure) and increase in preload (increased venous return caused by the increase in cardiac output) that challenged the right ventricle during CO_2 inhalation. The increase in venous filling pressure and consequent increase in end diastolic ventricular volume (increased ventricular stretch) should serve to maintain stroke volume and cardiac output in the face of the elevated afterload (increased pulmonary arterial pressure) during CO_2 inhalation (Sturkie, 1986).

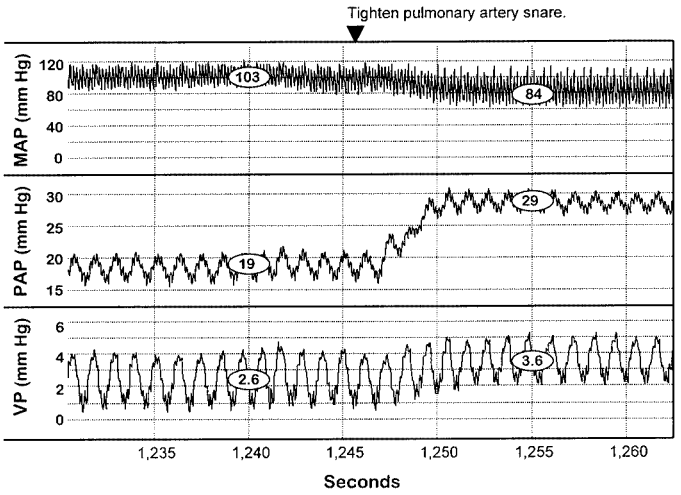

FIGURE 5. Experiment 2: Physiograph recording of mean systemic arterial pressure (MAP; upper tracing), pulmonary arterial pressure (PAP; middle tracing), and venous pressure (VP; lower tracing) for an individual broiler immediately before and during tightening of a snare around the left pulmonary artery. Circled numbers superimposed on each tracing indicate the mean value in millimeters Hg recorded at the times shown.

Venous pressure also increased and was sustained at an elevated level when the pulmonary vascular resistance was doubled by tightening a snare around one pulmonary artery. Previous studies consistently demonstrated that large increments in pulmonary vascular resistance cause dependent reductions in stroke volume and cardiac output (Wideman *et al.*, 1996a,b, 1998a,b, 1999; Wideman, 1999). The present observations confirm our hypothesis that, in addition to the influence of increased afterload on venous pressure, large increases in pulmonary vascular resistance can initiate venous congestion in broilers because of the inability of the right ventricle to elevate pulmonary arterial pressure sufficiently to propel all of the returning venous blood through the lungs (see Introduction). Veins have highly compliant walls that permit an approximate doubling in volume with only small (4 to 6 mm Hg) increases in central venous pressure. This feature permits the venous system to serve as the "volume reservoir" of the circulatory system (Akester, 1971; Langille, 1983; Sturkie, 1986). Presumably, the apparently modest 1-mm Hg increase in venous pressure we measured after CO_2 inhalation or unilateral pulmonary artery occlusion reflects a large increase in venous volume and, thus, substantial venous congestion, rather than generalized venous constriction mediated by sympathetic discharge (Wideman, 1999).

Tightening a snare around one pulmonary artery in clinically healthy broilers dramatically increases the rate of blood flow through the unoccluded lung and creates an immediate and fully reversible reduction of arterial blood oxygenation (hypoxemia) accompanied by an increase in the arterial partial pressure of CO_2 (hypercapnia) (Wideman and Kirby, 1995b; Wideman *et al.*, 1996a). The hyperperfusion hypoxemia and hypercapnia persist when the entire respiratory minute volume also is directed toward the left lung (Wideman *et al.*, 1996b). Similar hypoxemic and hypercapnic changes in blood gas occur in ascitic broilers. Indeed, the presence of a mild cyanosis, indicative of arterial blood that is less than 80% saturated with oxygen, is one of the earliest visible symptoms that an apparently healthy bird will develop ascites (Peacock *et al.*, 1989, 1990; Reeves *et al.*, 1991; Julian and Mirsalimi, 1992; Wideman and Bottje, 1993; Wideman,

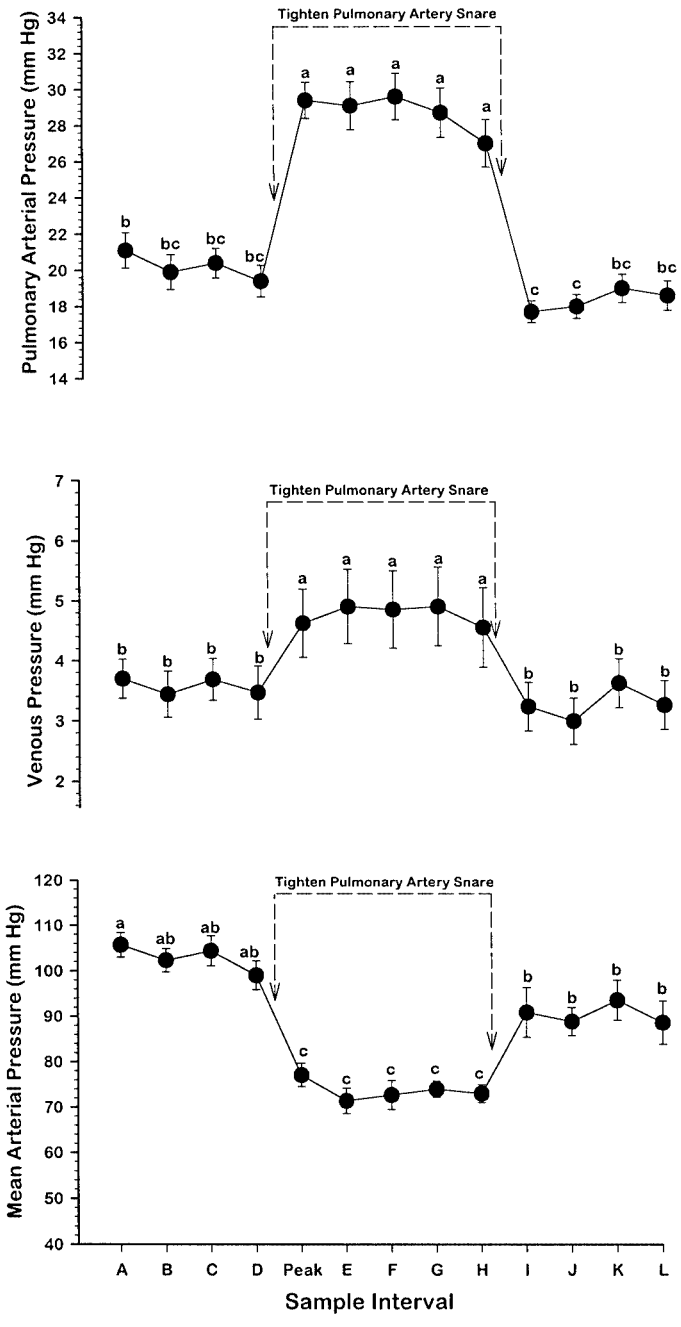

FIGURE 6. Experiment 2: Pulmonary arterial pressure (upper panel), venous pressure (middle panel), and mean systemic arterial pressure (lower panel) for clinically healthy broilers (mean ± SEM; n = 7) before tightening a pulmonary artery snare (sample intervals A to D), during the maximum increase in pulmonary arterial pressure attained within the first 5 min after tightening the snare (peak), throughout the period when the snare remained tightened (sample intervals E to H), and during the recovery period after the snare was released (sample intervals I to L). Different letters (a,b,c) designate differences ($P \leq 0.05$) over time.

TABLE 2. Linear regression equations, Pearson correlation coefficients (r), coefficients of determination (r^2), and probability (*P*) values for relationships between pulmonary arterial pressure (PAP) and pulmonary vascular resistance (PVR) or cardiac output (CO), between CO and stroke volume (SV) or heart rate (HR), and between venous pressure (VP) and cardiopulmonary variables during sample intervals A1 to D2 for the CO_2 group in Experiment 1[1]

PAP, CO, or VP *vs* variables	Equation	r	r^2	*P*
PAP *vs* PVR	PAP = 123.8PVR + 14.6	0.626	0.392	0.0001
PAP *vs* CO	PAP = 0.002CO + 22.7	0.031	0.0009	0.7848
CO *vs* SV	CO = 301.8SV + 39.8	0.954	0.909	0.0001
CO *vs* HR	CO = −0.389HR + 482	0.122	0.015	0.2814
VP *vs* PAP	VP = 0.218PAP − 1.07	0.438	0.192	0.0001
VP *vs* PVR	VP = 3.54PVR + 3.74	0.036	0.001	0.7512
VP *vs* CO	VP = 0.007CO + 1.51	0.292	0.085	0.0086
VP *vs* SV	VP = 3.11SV + 0.800	0.405	0.164	0.0002

[1]n = 80.

1997). This preascitic cyanosis cannot be attributed to low atmospheric oxygen, anemia, intracardiac right to left shunts, hypoventilation, or impaired respiratory function *per se*. Instead, the onset of cyanosis apparently is caused by a diffusion:perfusion mismatch that develops when blood flows too rapidly through the lungs to allow sufficient time for adequate oxygen diffusion across the pulmonary gas exchange surfaces or when blood perfuses under-ventilated regions of the parabronchi (Wideman and Kirby, 1995b; Wideman *et al.*, 1996a,b; Fedde *et al.*, 1998). The same mechanisms presumably prevent adequate diffusion of CO_2 out of the blood in susceptible broilers, leading to the onset of respiratory acidosis that, as shown in the present study, can modestly contribute to an overall elevation in the pulmonary vascular resistance and the pulmonary arterial pressure.

The partial pressure of O_2 in systemic arterial blood increased without causing a contemporaneous change in the percentage saturation of hemoglobin with O_2 when clinically healthy broilers inhaled 5% CO_2. Previously, bolus i.v. injections of 1.2 *N* HCl also increased the arterial partial pressure of O_2 without changing the percentage saturation of hemoglobin with O_2 or the partial pressure of CO_2 (Wideman *et al.*,1998b). In both experiments, the increased partial pressure of O_2 could not be attributed to an increase in respiratory minute volume (respiratory rate × tidal volume) and, therefore, may reflect a more efficient matching of parabronchial ventilation and pulmonary blood flow (improved diffusion:perfusion matching) following the induction of acidosis. Increases in the partial pressure of O_2 should attenuate pulmonary hypertension by reducing the cardiac output and by vasodilating the pulmonary vasculature (reduced pulmonary vascular resistance) in domestic fowl (Besch and Kadono, 1978; Sillau and Montalvo, 1982; Peacock *et al.*, 1989; Owen *et al.*, 1995). However, the increase in the arterial partial pressure of O_2 during CO_2 inhalation did not prevent the modest increase in pulmonary vascular resistance and pulmonary arterial pressure induced by the concurrent respiratory acidosis. Therefore, in agreement with previous studies, the level of arterial blood oxygenation can be dissociated from increases in pulmonary vascular resistance known to contribute to the onset of pulmonary hypertension in broilers (Owen *et al.*, 1994; Wideman *et al.*, 1996b, 1997, 1998b).

Respiratory acidosis reduces the affinity of hemoglobin for O_2 (Bohr effect), causing O_2 to be released more readily to the tissues but also requiring a higher partial pressure of O_2 at the gas exchange surfaces of the lungs to maintain a similar percentage saturation of hemoglobin with O_2 (Fedde, 1986). Increases in the partial pressure of O_2 during 5% CO_2 inhalation (present study) or i.v. injections of 1.2 *N* HCl (Wideman *et al.*, 1998b) were sufficient to maintain a constant percentage saturation of hemoglobin with O_2 in acidotic arterial blood. Given the stability of the HCT, this capacity to maintain a constant percentage saturation of hemoglobin with O_2, despite the imposed respiratory acidosis, served to maintain a relatively constant arterial O_2 content. In turn, a constant arterial O_2 content should maintain O_2 delivery to the tissues with no need to increase in cardiac output, thereby minimizing the contribution of cardiac output to the development of pulmonary hypertension during acidosis. Apparently, the factors that tend to inhibit the onset of pulmonary hypertension (increased partial pressure of O_2, constant arterial O_2 content, relatively stable cardiac output) were unable to counteract the pulmonary vasoconstrictive response to acidosis during the initial interval of 5% CO_2 inhalation.

ACKNOWLEDGMENTS

This research was supported by a US-Israel Binational Agricultural Research and Development grant (BARD US-2736-96) and by a grant from Hubbard ISA, Walpole, NH 03608.

REFERENCES

Akester, A. R., 1971. The blood vascular system. Pages 783–839 *in*: Physiology and Biochemistry of the Domestic Fowl. Volume 2. D. J. Bell and B. M. Freeman, ed. Academic Press, New York, NY.

Barer, G. R., H. McCurrie, and J. R. McCurrie, 1967. The effect of carbon dioxide and changes in blood pH on the pulmonary vascular resistance in cats. Clin. Sci. 32:361–376.

Besch, E. L., and H. Kadono, 1978. Cardiopulmonary responses to acute hypoxia in domestic fowl. Pages 71–78 *in*: Respiratory Function in Birds Adult and Embryonic. J. Piiper, ed. Springer-Verlag, New York, NY.

Brimioulle, S., P. Lejeune, J.-L. Vachiery, M. Leeman, C. Melot, and R. Naeije, 1990. Effects of acidosis and alkalosis on hypoxic pulmonary vasoconstriction in dogs. Am. J. Physiol. 258:H347–H353.

Farrukh, I. S., G. H. Gurtner, P. B. Terry, W. Tohidi, J. Yang, N. F. Adkinson, Jr., and J. R. Michael, 1989. Effect of pH on pulmonary vascular tone, reactivity, and arachidonate metabolism. J. Appl. Physiol. 67:445–452.

Fedde, M. R., 1986. Respiration. Pages 191–220 *in*: Avian Physiology, 4th ed. P. J. Sturkie, ed. Springer-Verlag, New York, NY.

Fedde, M. R., G. E. Weigle, and R. F. Wideman, 1998. Influence of feed deprivation on ventilation and gas exchange in broilers: Relationship to pulmonary hypertension syndrome. Poultry Sci. 77:1704–1710.

Guthrie, A. J., J. A. Cilliers, F. W. Huchzermeyer, and V. M. Killeen, 1987. Broiler pulmonary hypertension syndrome. II. The direct measurement of right ventricular and pulmonary artery pressures in the closed chest domestic fowl. Onderstepoort J. Vet. Res. 54:599–602.

Jandel Scientific, 1994. SigmaStat® Statistical Software User's Manual. Jandel Scientific Software, San Rafael, CA.

Julian, R. J., and S. M. Mirsalimi, 1992. Blood oxygen concentration of fast-growing and slow-growing broiler chickens, and chickens with ascites from right ventricular failure. Avian Dis. 36:730–732.

Kranen, R. W., C. H. Veerkamp, E. Lambooy, T. H. Van Kuppevelt, and J. H. Veerkamp, 1998. The effect of thermal preslaughter stress on the susceptibility of broiler chickens differing with respect to growth rate, age at slaughter, blood parameters, and ascites mortality, to hemorrhages in muscles. Poultry Sci. 77:737–744.

Langille, B. L., 1983. Role of venoconstriction in the cardiovascular responses of ducks to head immersion. Am. J. Physiol. 244:R292–R298.

Marshall, B. E., and C. Marshall, 1992. Acidosis and the pulmonary circulation. Pages 99–115 *in*: Hypoxia, Metabolic Acidosis, and the Circulation. A. I. Arieff, ed. Oxford University Press, New York, NY.

National Research Council, 1984. Nutrient Requirements of Poultry. 8th rev. ed. National Academy Press, Washington, DC.

Owen, R. L., R. F. Wideman, and B. S. Cowen, 1995. Changes in pulmonary arterial and femoral arterial blood pressure upon acute exposure to hypobaric hypoxia in broiler chickens. Poultry Sci. 74:708–715.

Owen, R. L., R. F. Wideman, R. M. Leach, B. S. Cowen, P. A. Dunn, and B. C. Ford, 1994. Effect of age of exposure and dietary acidification or alkalinization on mortality due to broiler pulmonary hypertension syndrome. J. Appl. Poult. Res. 3:244–252.

Peacock, A. J., C. Pickett, K. Morris, and J. T. Reeves, 1989. The relationship between rapid growth and pulmonary hemodynamics in the fast-growing broiler chicken. Am. Rev. Respir. Dis. 139:1524–1530.

Peacock, A. J., C. Pickett, K. Morris, and J. T. Reeves, 1990. Spontaneous hypoxaemia and right ventricular hypertrophy in fast growing broiler chickens reared at sea level. Comp. Biochem. Physiol. 97A:537–541.

Reeves, J. T., G. Ballam, S. Hofmeister, C. Pickett, K. Morris, and A. Peacock, 1991. Improved arterial oxygenation with feed restriction in rapidly growing broiler chickens. Comp. Biochem. Physiol. 99A:481–485.

Sillau, A. H., and C. Montalvo, 1982. Pulmonary hypertension and the smooth muscle of pulmonary arterioles in chickens at high altitude. Comp. Biochem. Physiol. 71A:125–130.

Sturkie, P. D., 1986. Heart and circulation: Anatomy, hemodynamics, blood pressure, blood flow. Pages 130–166 *in*: Avian Physiology, 4th ed. P. J. Sturkie, ed. Springer-Verlag, New York, NY.

Wideman, R. F., 1997. Understanding pulmonary hypertension syndrome (ascites). Hubbard Farms Technical Report, Walpole, NH, June 1997:1–6.

Wideman, R. F., 1999. Cardiac output in four, five, and six week old female broilers, and hemodynamic responses to intravenous injections of epinephrine. Poultry Sci. 78:392–403.

Wideman, R. F., and W. G. Bottje, 1993. Current understanding of the ascites syndrome and future research directions. Pages 1–20 *in*: Nutrition and Technical Symposium Proceedings. Novus International, Inc., St. Louis, MO.

Wideman, R. F., M. F. Forman, J. D. Hughes, Y. K. Kirby, N. Marson, and N. B. Anthony, 1998a. Flow-dependent pulmonary vasodilation during acute unilateral pulmonary artery occlusion in jungle fowl. Poultry Sci. 77:615–626.

Wideman, R. F., and Y. K. Kirby, 1995a. A pulmonary artery clamp model for inducing pulmonary hypertension syndrome (ascites) in broilers. Poultry Sci. 74:805–812.

Wideman, R. F., and Y. K. Kirby, 1995b. Evidence of a ventilation-perfusion mismatch during acute unilateral pulmonary artery occlusion in broilers. Poultry Sci. 74:1209–1217.

Wideman, R. F., and Y. K. Kirby, 1996. Electrocardiographic evaluation of broilers during the onset of pulmonary hypertension initiated by unilateral pulmonary artery occlusion. Poultry Sci. 75:407–416.

Wideman, R. F., Y. K. Kirby, M. F. Forman, N. Marson, R. W. McNew, and R. L. Owen, 1998b. The infusion rate dependent influence of metabolic acidosis on pulmonary vascular resistance in broilers. Poultry Sci. 77:309–321.

Wideman, R. F., Y. K. Kirby, R. L. Owen, and H. French, 1997. Chronic unilateral occlusion of an extra-pulmonary primary bronchus induces pulmonary hypertension syndrome (ascites) in male and female broilers. Poultry Sci. 76:400–404.

Wideman, R. F., Y. K. Kirby, C. D. Tackett, N. E. Marson, and R. W. McNew, 1996a. Cardio-pulmonary function during acute unilateral occlusion of the pulmonary artery in broilers fed diets containing normal or high levels of arginine-HCl. Poultry Sci. 75:1587–1602.

Wideman, R. F., Y. K. Kirby, C. D. Tackett, N. E. Marson, C. J. Tressler, and R. W. McNew, 1996b. Independent and simultaneous unilateral occlusion of the pulmonary artery and extrapulmonary primary bronchus in broilers. Poultry Sci. 75:1417–1427.

Wideman, R. F., P. Maynard, and W. G. Bottje, 1999. Thromboxane mimics the pulmonary but not systemic vascular responses to bolus HCl injections in broiler chickens. Poultry Sci. 78:714–721.