Blood oxygen binding in calves with naturally occurring diarrhea

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Objective—To assess blood oxygen binding in calves with diarrhea.

Animals—22 dairy and 26 double-muscled calves with diarrhea, 31 healthy dairy calves and 37 healthy double-muscled calves.

Procedure—Severity of disease, including the ability of affected calves to stand, was evaluated. Hydration and signs of depression were scored. Venous and arterial blood samples were collected, and 2,3-diphosphoglycerate, ATP, chloride, inorganic phosphate, lactate, pyruvate, total protein, albumin, and hemoglobin concentrations, and Hct, pH, P_{CO_2}, and P_{O_2} were determined. Oxygen equilibrium curves (OEC) were constructed under standard conditions, and oxygen extraction ratios were calculated.

Results—Recumbent calves of both breed-types were more dehydrated and had more severe signs of depression than ambulatory affected calves. In both breed-types, hemoglobin oxygen affinity was increased in calves with diarrhea, compared with healthy calves, as indicated by a decrease in standard partial oxygen pressure (P50). Diarrhea induced hypocapnia and hypothermia in the most severely affected calves, which counteracted the acidosis-induced right shift in arterial and venous OEC. Arterial and venous P50 were significantly less in double-muscled calves with diarrhea than healthy calves, whereas P50 for affected dairy calves were similar to those of healthy calves. Except in the most severely affected dairy calves, oxygen extraction ratio was significantly less in calves with diarrhea, compared with healthy calves.

Conclusions and Clinical Relevance—Release of oxygen from blood may be impaired in calves with diarrhea, depending on the effect of the disease on certain blood biochemical variables. (*Am J Vet Res* 2001;62:799–804).

Diarrhea is a common disease in newborn calves. However, although much is known about the physiopathologic events that develop in neonatal calves with gastroenteritis, to our knowledge, the effect of diarrhea on hemoglobin oxygen transport in calves has not been described. In other species, blood oxygen binding may improve or worsen in illness, depending on several modulating factors. For example, in humans with diabetic ketoacidosis, the opposing effects of acidosis and reduced concentrations of **2,3-diphosphoglycerate (2,3-DPG)** result in hemoglobin oxygen affinity that is within or slightly less than reference range.[1] If blood pH is rapidly corrected while 2,3-DPG concentration is still low, blood oxygen binding is impaired because of the marked left shift of the **oxygen equilibrium curve (OEC)**. In others studies, OEC were determined for humans with severe hypoxemia caused by acute respiratory distress syndrome or sleep apnea.[2,3] Under these conditions, hypoxemia stimulated 2,3-DPG synthesis, shifting the OEC to the right. Patients with chronic obstructive pulmonary disease have hemoglobin oxygen affinities that are less than[4] or within reference range[5] and 2,3-DPG concentrations greater than[5,6] or within reference range.[7] The function of RBC has also been studied in dogs with induced endotoxic shock.[8] In affected dogs, blood 2,3-DPG and ATP concentrations decreased, compared with healthy dogs, causing the **standard partial oxygen pressure (P50)** to decrease.

Because marked changes in blood biochemical variables are observed in calves with diarrhea, it may be that mechanisms similar to those observed in humans and dogs influence blood oxygen binding. The physiologic mechanisms controlling blood oxygen binding have been studied in healthy calves[9,10] but not in diseased calves. The purpose of the study reported here was to assess blood oxygen binding in calves with diarrhea by determining OEC, taking into account body temperature, acid-base balance, Pa_{CO_2}, Pa_{O_2}, Pv_{CO_2}, and Pv_{O_2}. We also considered the influence of OEC-regulating factors such as blood 2,3-DPG, ATP, inorganic phosphate, and chloride concentrations. Finally, because of differences in the regulation of blood oxygen binding in dairy and double-muscled calves,[9,10] the influence of breed-type was considered.

Materials and Methods

Animals—Forty-eight 1- to 23-day-old calves with diarrhea were studied. Calves comprised the following 2 breed-types: double-muscled (Belgian White and Blue beef calves; n = 26) and dairy (22). Sick calves were assigned to 4 groups according to breed-type and clinical signs. Calves that were recumbent were classified as severely affected (double-muscled, n = 14; dairy, 10), whereas calves that remained ambulatory were classified as moderately affected (double-muscled, 12; dairy, 12). A control group of healthy calves (double-muscled, n = 37; dairy, 31) in the same age range as affected calves was also studied.

Clinical evaluation—A scoring system was used to determine severity of disease. Dehydration was scored on the

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basis of enophthalmia and skin tenting, according to a previous report.[11] Individual scores ranged from 0 in calves that were 5 to 6% dehydrated to 3 in calves that were 10 to 14% dehydrated. Signs of depression were assessed as described by Kasari and Naylor.[12] For each calf, scores indicating strength of the sucking reflex, menace reflex, and tactile response, ability to stand, and warmth of the oral cavity and extremities were summed to give an overall depression score. Depression scores ranged from 0 in healthy calves to 13 in comatose calves.

Blood sample collection—Heparinized syringes were used to collect 1 ml of venous (jugular vein) and 1 ml of arterial (brachial artery) blood under anaerobic conditions. Syringes were placed on ice, and pH, PO_2, and PCO_2 were measured immediately.[a] Rectal temperature was measured to correct blood gas values and pH. Venous blood samples were also collected into 20-ml syringes containing heparin for determination of OEC and biochemical and hematologic analyses. Blood was immediately stored at 4 C, and OEC were recorded within 1 day of sample collection, a time frame in which we did not observe modifications in OEC for control calves. To determine chloride, inorganic phosphate, total protein, and albumin concentrations, 1 ml of plasma and 1 ml of serum were stored at 4 C after centrifuging venous blood for 15 minutes at 2,600 × g. For determination of pyruvate and lactate concentrations, 2 ml of venous blood was immediately deproteinized in 4 ml of 10% trichloracetic acid and centrifuged at 2,600 × g for 10 minutes.

Determination of oxygen equilibrium curves—The OEC of oxyhemoglobin was measured by use of a dynamic method under standard conditions (pH, 7.4; PCO_2, 40 mm Hg; temperature, 37 C).[13] A 15-ml venous blood sample was deoxygenated in a rotary tonometer with a gas mixture composed of 5.6% CO_2 and nitrogen. Blood was placed in an analyzer and equilibrated with this first gas mixture. For 15 minutes, oxygen tension in blood samples was slowly increased from 0 to 320 mm Hg by introducing a second gas mixture composed of 5.6% CO_2 and oxygen. Oxygen saturation was measured by photometry[b] as a function of PO_2; PO_2 was measured polarographically.[c] Changes in plasma pH were corrected automatically to 7.4 by adding 1N NaOH or 1N HCl. A temperature of 37 C and a PCO_2 of 40 mm Hg were maintained in the analyzer throughout determination of OEC. For each curve, 100 points were automatically measured. Values for PO_2 and oxygen saturation were stored on a computer for data processing and curve generation. The accuracy of our method for measuring OEC, expressed by the SD of P50, was 0.1 mm Hg for 6 curves determined for the same blood sample (ie, analytical error of the analyzer) and 0.3 mm Hg for 11 curves determined for 11 blood samples collected from the same healthy calf over a 30-day period (ie, analytical error associated with intraindividual variations). Oxygen affinity changes were evaluated by measuring PO_2 at 50% hemoglobin saturation under standard conditions (ie, standard P50).

Hematologic and biochemical analyses—Hemoglobin concentration (g/100 ml of blood) was determined with a hemoximeter.[d] Hematocrit was measured by microcentrifugation.[e] Concentrations of DPG and ATP in blood were determined by use of enzymatic methods.[f,g] Concentration of inorganic phosphate in plasma was determined by use of a commercially available kit.[h] Plasma chloride concentration was determined by use of a titrimetric method.[i] L-Lactate and L-pyruvate concentrations in blood were determined by use of an enzymatic method,[j] and the **L-lactate-to-L-pyruvate concentration ratio (lactate:pyruvate)** was calculated. Total protein concentration in serum was determined by use of the biuret method,[14] and albumin concentration was determined by electrophoretic fractionation of serum proteins.[k]

Calculation of oxygen extraction ratio—The oxygen extraction ratio was calculated as the ratio of the difference between arterial and venous oxygen content to arterial oxygen content. Arterial (CaO_2) and venous (CvO_2) blood oxygen content were calculated as follows:

$$CaO_2 = Hb \times BO_2 \times (SaO_2/100) + \alpha(PaO_2)$$

$$CvO_2 = Hb \times BO_2 \times [SvO_2/100] + \alpha(PvO_2)$$

where BO_2 is hemoglobin oxygen capacity (1.39 ml of O_2/g of Hb),[15] Hb is blood hemoglobin concentration (g/100 ml), α is the oxygen solubility coefficient for blood at the temperature of the experiment (0.003 ml × 100 ml^{-1} × mm Hg^{-1}), and SaO_2 and SvO_2 are the percentage of oxygen saturation of hemoglobin in arterial and venous blood, respectively. Percentage of oxygen saturation of hemoglobin was obtained from the measured PO_2 of arterial and venous blood, taking into account the position and shape of the OEC in both compartments. In practice, arterial and venous OEC were calculated for both breed-types from the standard OEC corrected for the effects of pH, body temperature,[16] and PCO_2.

Table 1—Effects of breed type and severity of diarrhea on determinants of the oxygen equilibrium curve in calves

	Healthy		Ambulatory affected		Recumbent affected	
Variable	Double-muscled (n = 37)	Dairy (n = 31)	Double-muscled (n = 12)	Dairy (n = 12)	Double-muscled (n = 14)	Dairy (n = 10)
P50s (mm Hg)	22.5 ± 0.4	21.2 ± 0.4	19.9 ± 0.5*	19.6 ± 0.5*	19.6 ± 0.6*	19.9 ± 0.4*
P50a (mm Hg)	26.8 ± 0.4	25.1 ± 0.4†	24.7 ± 0.4*	23.9 ± 0.8	24.1 ± 0.7*	26.3 ± 1.1‡
P50v (mm Hg)	29.4 ± 0.5	27.0 ± 0.5†	26.5 ± 0.5*	25.8 ± 0.4	26.8 ± 0.9*	29.4 ± 1.5‡
pHa	7.34 ± 0.01	7.37 ± 0.01†	7.30 ± 0.02	7.32 ± 0.02*	7.20 ± 0.03*‡	7.16 ± 0.04*‡
pHv	7.29 ± 0.01	7.34 ± 0.01†	7.27 ± 0.03	7.30 ± 0.02*	7.16 ± 0.03*‡	7.12 ± 0.05*‡
$PaCO_2$ (mm Hg)	45.8 ± 0.6	46.6 ± 0.7	43.5 ± 2.2	41.4 ± 1.2*	34.0 ± 2.0*‡	33.3 ± 1.7*‡
$PvCO_2$ (mm Hg)	56.8 ± 1.0	54.8 ± 0.9	51.2 ± 2.2*	50.1 ± 1.9*	45.8 ± 2.4*	46.0 ± 1.9*‡
PaO_2 (mm Hg)	86.4 ± 1.9	85.1 ± 2.2	88.8 ± 4.5	86.4 ± 3.8	90.1 ± 4.7	85.8 ± 2.8
PvO_2 (mm Hg)	37.5 ± 1.3	37.2 ± 1.2	44.1 ± 2.5*	43.7 ± 2.6*	42.1 ± 2.2*	38.4 ± 2.0
OER	0.33 ± 0.02	0.28 ± 0.02	0.18 ± 0.03*	0.19 ± 0.03*	0.22 ± 0.03*	0.30 ± 0.03†‡

Data reported as mean ± SEM.

*Significantly ($P < 0.05$) different from value for healthy calves of the same breed-type. †Significantly ($P < 0.05$) different from value for double-muscled calves. ‡Significantly ($P < 0.05$) different from value for ambulatory calves of the same breed-type.

P50s = PO_2 at 50% hemoglobin saturation, measured under standard conditions (pH, 7.4; PCO_2, 40 mm Hg; temperature, 37 C). P50a = PO_2 at 50% hemoglobin saturation in the arterial compartment. P50v = PO_2 at 50% hemoglobin saturation in the venous compartment. a = Arterial. v = Venous. OER = Oxygen extraction ratio.

Statistical analyses—Data were expressed as means ± SEM. Values were tested for normal distribution by use of the Omnibus test.[17] Influences of breed-type and diarrhea on OEC were assessed by use of 2-way ANOVA. Normally distributed data were compared by use of a Student *t*-test or an Aspin-Welch test, depending on whether the variances were equal. When data were not normally distributed, a nonparametric test (Mann-Whitney test) was performed. For all analyses, values of $P < 0.05$ were considered significant.

Results

Recumbent calves had significantly higher dehydration and depression scores than ambulatory calves, confirming that disease was more severe in the former group. However, severity of dehydration and depression were independent of breed-type.

Age distribution was not significantly different among groups (healthy double-muscled, 9.8 ± 1.1 days; healthy dairy 8.7 ± 0.8 days; ambulatory affected double-muscled, 11.1 ± 1.6 days; ambulatory affected dairy 7.5 ± 1.0 days; recumbent affected double-muscled, 9.5 ± 1.4 days; recumbent affected dairy 7.8 ± 1.1 days). In addition, breed-related differences were not detected for standard P50. Standard P50 for calves with diarrhea were significantly less than for healthy calves. However, severity of disease did not affect standard P50 (**Table 1**).

Mean concentration of 2,3-DPG in blood was significantly higher in healthy double-muscled calves than in healthy dairy calves (14.1 ± 0.8 vs 10.0 ± 0.7 µmol/g of Hb). Breed-related differences, however, were not detected in affected calves, because 2,3-DPG concentration decreased significantly in double-muscled calves with diarrhea (ambulatory, 6.5 ± 1.0 µmol/g of Hb; recumbent, 6.1 ± 1.3 µmol/g of Hb) but did not change significantly from the control value in affected dairy calves. Concentrations of ATP, inorganic phosphate, and chloride were also significantly greater in healthy double-muscled calves, compared with healthy dairy calves, and chloride concentration was greater in ambulatory affected double-muscled calves, compared with ambulatory affected dairy calves (**Table 2**). We did not detect significant changes in ATP and chloride concentrations in affected calves of either breed-type, but concentrations of inorganic phosphate were significantly increased in calves of both breed-types with diarrhea.

Breed-related differences in rectal temperature were not detected in healthy or affected calves; rectal temperatures of healthy calves and ambulatory calves were within reference range. However, recumbent calves of both breed-types were hypothermic (double-muscled, 38.2 ± 0.4 C; dairy, 38.2 ± 0.4 C).

Healthy double-muscled calves had a relative acidosis, compared with healthy dairy calves (Table 1). This breed-related difference disappeared in calves with diarrhea, where, except among the ambulatory double-muscled group, blood was more acidic in affected calves than in healthy calves. Acidosis was significantly more severe in recumbent calves, compared with ambulatory calves. Breed-related differences were not detected for Pa_{CO_2} or Pv_{CO_2}. In calves with diarrhea, Pa_{CO_2} and Pv_{CO_2} were less, compared with healthy calves; Pa_{CO_2} was also significantly less in recumbent affected calves, compared with ambulatory affected calves.

Arterial and venous P50 were significantly higher in healthy double-muscled calves than in healthy dairy calves (Table 1). Arterial and venous P50 decreased significantly in double-muscled calves with diarrhea, but no change was observed in affected dairy calves. Hence, breed-related differences were not detected in arterial and venous P50 values for calves with diarrhea.

In healthy calves, breed-related differences were not observed for Pa_{O_2}, Pv_{O_2}, or oxygen extraction ratio (Table 1). Diarrhea did not significantly affect Pa_{O_2} but did result in a significantly increased Pv_{O_2}. Consequently, mean oxygen extraction ratios were significantly less in calves with diarrhea, compared with healthy calves, except for recumbent dairy calves in which the oxygen extraction ratio was similar to the control value. Hemoglobin concentration was significantly greater in healthy double-muscled calves, compared with healthy dairy calves. Hemoglobin concentrations increased in calves with diarrhea, especially in the most affected calves (Table 2).

Breed-related differences were not detected in L-lactate and L-pyruvate concentrations (Table 2). However, lactate:pyruvate was significantly higher in ambulatory double-muscled calves with diarrhea, compared with affected dairy calves. Mean L-lactate and L-pyruvate concentrations were greater in calves with

Table 2—Effects of breed type and severity of diarrhea on variables affecting oxygen equilibrium curves and related indexes in calves

	Healthy		Ambulatory affected		Recumbent affected	
Variable	Double-muscled (n = 37)	Dairy (n = 31)	Double-muscled (n = 12)	Dairy (n = 12)	Double-muscled (n = 14)	Dairy (n = 10)
ATP (µmol/dl)	27.1 ± 1.0	21.7 ± 1.3*	24.2 ± 2.3	24.7 ± 1.7	26.4 ± 2.8	29.0 ± 3.0
Chloride (mmol/L)	105.5 ± 0.9	101.8 ± 1.0*	106.2 ± 1.6	101.8 ± 1.6*	107.4 ± 2.5	102.6 ± 2.3
Pi (mmol/L)	2.5 ± 0.1	2.3 ± 0.1*	2.3 ± 0.1†	2.3 ± 0.2	3.5 ± 0.4†‡	3.6 ± 0.4†‡
Hb (g/dl)	10.6 ± 0.2	9.7 ± 0.4*	10.8 ± 0.5	11.3 ± 0.5†	12.1 ± 0.7†	13.2 ± 0.9†‡
L-lactate (mmol/L)	2.0 ± 0.2	1.5 ± 0.1	1.9 ± 0.5	1.9 ± 0.4	3.4 ± 0.9	5.5 ± 1.8*
L-pyruvate (mmol/L)	0.18 ± 0.01	0.18 ± 0.01	0.14 ± 0.02	0.18 ± 0.02	0.19 ± 0.03	0.22 ± 0.13
Lactate:pyruvate	10.5 ± 0.4	8.9 ± 0.6	12.3 ± 1.0	10.3 ± 1.1*	15.5 ± 1.5†‡	20.6 ± 3.3†‡

Pi = Inorganic phosphate concentration. Hb = Hemoglobin concentration. Lactate:pyruvate = L-Lactate-to-L-pyruvate concentration ratio.

*Significantly ($P < 0.05$) different from value for healthy calves of the same breedtype. †Significantly ($P < 0.05$) different from value for double-muscled calves. ‡Significantly ($P < 0.05$) different from value for ambulatory calves of the same breed-type.

diarrhea, compared with healthy calves. However, this difference was only significant between healthy and severely affected dairy calves. Lactate:pyruvate was significantly higher in recumbent calves, compared with both ambulatory affected calves and healthy calves.

Breed-related differences were not detected in Hct and serum total protein and albumin concentrations. Moreover, total protein and albumin concentrations did not differ significantly between healthy calves and calves of either breed-type with diarrhea. Hematocrit increased significantly in dairy calves with diarrhea, regardless of severity of disease (healthy, 32.3 ± 1.5%; ambulatory affected, 38.8 ± 1.9%; recumbent affected, 45.2 ± 3.2%); Hct was also significantly greater in recumbent affected dairy calves, compared with ambulatory affected calves. For double-muscled calves, Hct was significantly increased only in the recumbent affected group (healthy, 35.0 ± 0.8%; ambulatory affected, 36.9 ± 2.0%; recumbent affected, 41.1 ± 2.5%).

Discussion

In a previous study,[10] blood oxygen binding was compared between healthy dairy and double-muscled calves. Results of that study indicated that, to maintain a level of oxygen consumption similar to that of dairy calves, double-muscled calves must resort to compensatory mechanisms such as greater blood concentrations of 2,3-DPG, inorganic phosphate, and hemoglobin and a relative acidosis. For this reason, we considered the influence of breed-type in addition to the effects of clinically severe and moderate diarrhea on blood oxygen binding in calves. Our results indicate that blood oxygen binding can be impaired by diarrhea depending on how the pathologic process affects certain blood biochemical variables.

As illustrated by the decrease in standard P50, hemoglobin oxygen affinity was enhanced in calves with diarrhea, especially in affected double-muscled calves. This may be related to the decrease in 2,3-DPG concentration in affected calves, compared with healthy calves, which was significant only in double-muscled calves. The role of 2,3-DPG has been described in calves[9]; adult cattle do not have detectable concentrations of 2,3-DPG in erthryocytes.[9,18] The decrease in 2,3-DPG concentration likely resulted because affected calves developed acidosis. 2,3-Diphosphoglycerate is a known glucose metabolite in erythrocytes. By inhibiting phosphoglycerate mutase and activating phosphoglycerate phosphatase, acidosis can result in a mobilization of 2,3-DPG stores.[19] However, a decrease in glucose concentration could also be involved, as glucose is the precursor of 2,3-DPG. A relationship between acidosis and 2,3-DPG concentration was identified in humans and dogs.[1,8] Our results indicate that a similar relationship exists in calves, at least in double-muscled calves.

Similar to results of previous studies,[9,10] we found that healthy double-muscled calves had higher chloride and ATP concentrations than dairy calves. Concentrations of these OEC-regulating factors did not change in calves with diarrhea. However, concentration of inorganic phosphate, another regulatory factor, significantly increased in calves with diarrhea, especially in the severely affected groups. This increase in inorganic phosphate concentration as a result of diarrhea was also demonstrated by Groutides and Michell[20] and may be related to dehydration-linked prerenal failure. Because high inorganic phosphate concentrations can shift the OEC to the right,[21] an increase in concentration may limit the effects of low 2,3-DPG concentrations on hemoglobin oxygen affinity. The changes in inorganic phosphate concentration in calves with diarrhea, however, are probably too slight to have clinical relevance.[9]

The recorded changes in standard P50 were insufficient to enable us to assess the influence of diarrhea on blood oxygen binding, because factors such as blood pH, body temperature, and P_{CO_2} shift the OEC and alter related variables such as the oxygen extraction ratio. Acidosis develops in animals with diarrhea[22] and enhances blood oxygen release in all species studied, including calves, by shifting the OEC to the right. When severe, however, metabolic acidosis affects many organs, especially the pulmonary and cardiovascular systems. In humans, arterial pH < 7.2 is associated with increased pulmonary vascular resistance and decreased myocardial contractility, because hydrogen ions act as a negative inotrope.[23] In this context, the acidosis-induced shift of the OEC can be viewed as a mechanism limiting the negative effects of acidosis on the cardiovascular system. Yet acidosis also causes 2,3-DPG concentrations to decrease, which counteracts the Bohr effect on blood oxygen transport. Moreover, in calves with diarrhea, the left shift in the OEC induced by hypocapnia and hypothermia, which was most evident in the severely affected calves, also counteracted the effects of acidosis on blood oxygen transport. We attributed hypocapnia in affected calves to an increase in alveolar ventilation caused by stimulation of peripheral and medullary chemoreceptors in response to an increase in hydrogen ion concentration.[23] Hypothermia in the severely affected calves may have been related to poor tissue perfusion as a result of dehydration. However, results of other studies[24-26] evaluating calves with experimentally induced noninfectious diarrhea indicate an increase in rectal temperature associated with dehydration.

In both breed-types of calves that we studied, the combined effects of all regulating factors on blood oxygen binding rendered the position of the OEC dependent on severity of disease. Diarrhea caused arterial and venous P50 to decrease in double-muscled calves but not in dairy calves. This was primarily a result of the more severe acidosis in dairy calves. Recumbent dairy calves had higher venous and arterial P50 than ambulatory dairy calves (ie, a position of the OEC more in favor of oxygen release at the tissue level).

Oxygen release is determined not only by the position of the OEC but also by Pa_{O_2} and Pv_{O_2}. Similar to results of Fayet and Overwater,[27] we detected an increase in Pv_{O_2} in calves with diarrhea, compared with healthy calves. Because of the significant increase in Pv_{O_2} recorded in affected calves of both breed-types and the left shift of the OEC, the oxygen extraction ratio decreased significantly in all groups of calves with diarrhea except in the severely affected dairy calves. In this latter group, the oxygen extraction ratio was not significantly different from the control value. The primary reason for this finding was the normal positions of the OEC for arterial and venous blood in severely affected dairy calves, which was a result of more severe acidosis in this group. Oxygen extraction was thus impaired in double-muscled calves and moderately affected dairy calves but not in severely affected dairy calves. These differences between breed-types are more likely attributable to differences in disease severity than genetic differences. The increase in Pv_{O_2} and the decrease in oxygen extraction ratio in calves with diarrhea were surprising. Indeed, it has been extensively described that, over a wide range of decreased cardiac output, as may occur in calves with diarrhea, systemic oxygen consumption is independent of systemic oxygen delivery. This relationship reflects the compensatory properties of the oxygen transport system, in which extraction of oxygen from the blood by tissues is increased.[28-32] The increase in Pv_{O_2} and the decrease in the oxygen extraction ratio that we detected in the present study could be attributable to an increase in tissue perfusion, which seems unlikely in calves with diarrhea, or to increased shunting of blood. When perfusion mismatching develops, as described in animals with bacteremia, Pv_{O_2} increases, and oxygen extraction ratio decreases.[33] The increase in lactate:pyruvate that we detected in calves with diarrhea supports this hypothesis. This ratio was increased in affected calves, compared with healthy calves, which may be attributable to a decrease in intracellular pH and not to a decrease in cell redox state (ie, a decrease in the NAD-to-NADH concentration ratio). Indeed, lactate:pyruvate was significantly (r = 0.61) correlated to pH of venous blood.

The increase in hemoglobin concentration that we detected in calves with diarrhea was attributable to hemoconcentration, which is potentially deleterious to affected calves, as it can lead to reduced peripheral circulation.[34] Recumbent calves appeared more severely dehydrated than ambulatory calves. Degree of dehydration can also be estimated from Hct and serum concentrations of total protein and albumin. However, several authors report[35] that Hct is highly variable in healthy calves and calves with diarrhea. This is probably why mean Hct was not correlated with dehydration score in the present study. Total serum protein concentration in other animals is commonly used as an index of dehydration, but it is unreliable in calves, because it varies with the amount of colostral immunoglobulin absorbed.[35] Moreover, in calves with diarrhea, total protein concentration can decrease as a result of fecal protein loss, decreased food intake, and decreased absorption.[14] This may explain why total protein concentration did not differ significantly between healthy calves and calves with diarrhea.

Our results indicate that a left shift in the OEC occurs in calves with diarrhea as a result of hypothermia and hypocapnia and, at least in double-muscled calves, a decrease in blood concentration of 2,3-DPG. This left shift is partially or totally counterbalanced by the development of acidosis, depending on its intensity. In addition, an increase in Pv_{O_2} in affected calves can contribute to limiting hemoglobin desaturation and oxygen extraction at the tissue level.

[a]AVL, Biomedical Instruments, Graz, Austria.
[b]LED (660 nm), Monsanto, St Louis, Mo.
[c]P_{O_2} electrode, Eschweiler, Kiel, Germany.
[d]OSM3 Hemoximeter, Radiometer Medical A/S, Copenhagen, Denmark.
[e]Universal 30 RF, Hettich, Tuttlingen, Germany.
[f]DPG kit No. 35A, Sigma Chemical Co, St Louis, Mo.
[g]ATP kit No. 366, Sigma Chemical Co, St Louis, Mo.
[h]Phosphorus, inorganic kit No. 670, Sigma Chemical Co, St Louis, Mo.
[i]Merckotest, Merck & Co, Dormstad, Germany.
[j]Lactate kit No. 826A and pyruvate kit No.726, Sigma Chemical Co, St Louis, Mo.
[k]Cliniscan 2, Helena Laboratories, Beaumont, Tex.

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