Post parturient hemoglobinuria in a flock of Akkaraman sheep

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ABSTRACT

Postpartum hemoglobinuria (PPH) is a rare but potentially serious metabolic disorder in small ruminants, and cases in sheep are rarely reported. This case report describes an outbreak of PPH associated with hypophosphatemia in a flock of meat-producing ewes. The case involved a 4-year-old Akkaraman ewe that developed reddish urine, weakness, anorexia, and mild dehydration several weeks after lambing. A review of the herd's history revealed that multiple ewes, particularly those that had produced triplets, had exhibited similar clinical signs, and several had died in the preceding days. The diet was found to be insufficient in terms of mineral content, and signs of pica were noted in the flock, suggesting a chronic imbalance in the diet's mineral content. Diagnostic evaluation included physical examination, hematology, biochemistry, blood smear analysis, and urine testing. Hypophosphatemia along with decreased calcium, magnesium, and potassium levels was detected. Hemoglobinuria was confirmed through urine centrifugation and an ammonium sulphate precipitation test, and other potential causes of pigmenturia were excluded based on clinical and laboratory findings. Together, these findings supported a diagnosis of PPH secondary to dietary phosphorus deficiency. There was no evidence of parasitic or infectious hemolytic disease. Treatment consisted of intramuscular phosphorus supplementation, subcutaneous calcium and magnesium preparations, and supportive therapy. All affected animals responded positively, with complete clinical recovery observed after five days of treatment. A vitamin and mineral supplement was subsequently added to the animals diet to prevent recurrence, and no new cases were reported during the subsequent one-year follow-up period. This case demonstrates that PPH, although rare in sheep, can pose a significant health challenge to flocks with inadequate nutrition. Even moderate reductions in serum phosphorus may predispose susceptible animals to hemolysis, underscoring the variability in individual tolerance. The present report highlights the importance of balanced dietary mineral intake, proper postpartum monitoring, and early therapeutic intervention to prevent losses associated with PPH in sheep.

KEY WORDS

Akkaraman; postparturient hemoglobinuria; sheep flock; phosphorus deficiency; treatment.

INTRODUCTION

Postpartum hemoglobinuria (PPH) is a metabolic disease of dairy cattle and buffaloes characterized by intravascular hemolysis, hemoglobinuria, and anemia. Hypophosphatemia, often caused by dietary phosphorus deficiency (1). Severe hypophosphatemia reduces erythrocyte glycolysis and adenosine triphosphate (ATP) synthesis, leading to erythrocyte fragility, hemoglobinemia, and hemoglobinuria (2). PPH is rarely seen in sheep due to low milk yield, selective grazing habits, and erythrocyte resistance to hypophosphatemic hemolysis (3), with only one case previously reported (1). This report describes PPH in a sheep flock with hypophosphatemia and its treatment.

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MATERIALS AND METHODS

A 4-year-old Akkaraman sheep that had delivered twins was presented reddish urine and anorexia. In a flock of 250 meat-producing ewes, 15 had lambed 4-5 weeks earlier; four ewes with triplets developed hemoglobinuria and died within 4-5 days, while the remaining 11 that twins showed similar signs but survived. Lambs were healthy. Enterprise records show an increase in the rate of multiple lambing compared to the previous season. Pica was observed throughout the entire flock.

RESULTS

Clinical examination revealed weakness, anorexia, dehydration

(8%), and mild mucous membrane pallor, with normal vital signs and no jaundice. Fecal examination was negative for parasites. Blood samples was collected for blood gas analysis, complete blood count (CBC), and biochemical analysis. Mild hemolysis was observed in serum, which was stored at −80 °C until analysis. In all animal species, the renal threshold for myoglobin is so low that the pronounced discoloration of serum observed in hemoglobinemia is not detectable. Biochemical testing revealed hypophosphatemia, hypocalcemia, hypokalemia and hypomagnesemia (Table 1). The blood smear taken from the ear tip did not reveal any parasites (*Anaplasma*, *Babesia*, *Theileria*). The urine was reddish (Figure 1). To differentiate between hematuria and hemoglobinuria, the urine sample was centrifuged at 3500 x *g* for 5 minutes. After centrifugation, no change in urine colour was observed, confirming the presence of hemoglobinuria. Subsequently, an ammonium sulfate precipitation test was performed to differentiate between hemoglobinuria and myoglobinuria. To 5 ml urine sample 2.8 g ammonium sulfate was added slowly with thorough mixing to obtain 80 % saturation of ammonium sulphate in urine. A mixture of ammonium sulfate and urine sample was centrifuged at 3500 x *g* for 10 minutes. Ammonium sulfate precipitates hemoglobin, but not myoglobin (4). Following centrifugation, supernatant is clear, the pigment is hemoglobin. Copper toxicity, leptospirosis, bacillary hemoglobinuria, water intoxication, and brassica and onion toxicity (5) were excluded as possible causes of hemoglobinuria in sheep based on history, clinical examination, and laboratory findings. On the basis of the history, the clinical signs and the laboratory findings, the case was diagnosed as PPH due to hypophosphatemia. Phosphorus, calcium, magnesium, and supportive treatment were administered to the 11 affected ewes. Toldimfos sodium (Phosphotonic® 20%, Topkim) 3 mL, administered once daily by intramuscular injection, and calcium gluconate, calcium acetate, and magnesium hypophosphite hexahydrate (Calject 50®, Teknovet) 20 mL, administered once daily by subcutaneous injection, were used for 5 days. After treatment, all the sheep showing clinical signs recovered completely. Additionally, a vitamin-mineral premix (Royvimin-B Forte) was added to the diet at 1 kg per 100 kg feed. During 1-year follow-up, no recurrence was observed.

Table 1 - Complete blood count, blood gas, and biochemical parameters in a sheep with postparturient hemoglobinuria.

Parameters	Results	Reference interval[a]
WBC (cells/ L)	10.30	4000-12000
Lym (cells/ L)	8.40	2000-9000
Mon (cells/ L)	0.19	0-800
Gra (cells/ L)	1.7	0.7-6
RBC ($x10^6$/ L)	11	9-15
Hb (g/dL)	14.80	9-15
HCT (%)	44.20	27-45
MCV (fL)	40.20	28-40
MCH (pg)	13.40	8-12
MCHC (g/dL)	33.40	31-34
PLT ($x10^3$/ L)	315	205-705
pH	7.35	7.29-7.37
pCO_2 (mmHg)	29.9	41-50
pO_2 (mmHg)	39.3	34-58
sO_2 (%)	56	50-71
K (mmol/L)	2.6	3.9-5.4
Na (mmol/L)	151	139-152
Cl (mmol/L)	113	95-103
Glucose (mg/dL)	164	50-80
Lac (mmol/L)	2.2	1-1.33
Bilirubin total (mg/dL)	0.1	0.1-0.5
BE (mmol/L)	-8.8	+/- 5
HCO_3^- (mmol/L)	17.6	20-25
Total Ca (mg/dL)	9.2	10.5-12.8
P (mg/dL)	2.2	3.1-5.6
Mg (mg/dL)	2.0	2.2-2.8

[a] Reference values obtained from Baird and Pugh (2012)
WBC: white blood cell, Lym: lymphocyte, Mon: monocyte, Gra: Granulocyte, RBC: red blood cell, Hb: hemoglobin, Hct: hematocrit, MCV: mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration, PLT: platelet, pH: power of hydrogen, pCO_2: partial pressure of carbon dioxide, pO_2: partial pressure of oxygen, sO_2: oxygen saturation, K: potassium, Na: sodium, Cl: chlorine, Lac: lactate, BE: base excess, HCO_3^-: bicarbonate, Ca: calcium, P: phosphorus, Mg: magnesium

Figure 1 - Reddish urine.

DISCUSSION

While many PPH cases are reported in cattle and buffalo (6-8), only one prior case has been reported in sheep (1). In this report, all cases occurred postpartum, specifically 4-5 weeks after lambing, consistent with the typical postpartum onset seen in cows and buffaloes (9,10,11). In this case, consistent with previous reports (6,8,12), clinical signs in sheep included weakness, anorexia, dehydration, mild mucous membrane pallor, and hemoglobinuria. In buffaloes with PPH, jaundice is reported to occur in 5% of cases and the increase in total bilirubin correlates clinically with the observed jaundice (6). As a result of erythrocyte hemolysis or degradation in the spleen and liver,

total bilirubin levels may increase and anemia may occur without hemoglobinaemia (13). In this case report, the total bilirubin levels are within the reference ranges. This absence of jaundice in this case can be explained by the acute course of the disease or the mild degree of hemolysis; indeed, no anemia was detected in the animal (13). Additionally, the absence of jaundice may be attributed to the decrease in plasma phosphorus levels, suggesting that it may not reliably indicate intracellular phosphorus levels in erythrocytes and does not affect changes in erythrocyte osmotic resistance (14). PPH may result from primary dietary phosphorus deficiency (3,9). In lactating cows, diets providing ≥40% less than the phosphorus requirement for 5 weeks can rapidly lower plasma phosphorus (14). In this flock, traditional feeding, observed hypophosphatemia, and pica suggest dietary phosphorus deficiency as the main cause. While dietary deficiencies in copper (Cu) and selenium (Se) have been suggested to contribute to PPH (6,15), recovery without copper or selenium supplementation indicates these minerals were likely adequate. The hypophosphatemia observed in the postpartum period has a complex etiology involving reduced feed intake in the periparturient period, increased phosphorus loss from the mammary gland, and a shift of phosphorus from the extracellular to the intracellular compartment (14). Studies have reported that cows with phosphorus levels as high as 1.5 mg/dL (10) and as low as 0.92 mg/dL (9) can develop PPH. Ogawa et al. (1989) observed hemoglobinemia and a decrease in hematocrit in two cows with phosphorus levels falling to 1 mg/dL, but no hemoglobinuria was reported (16). In one flock of sheep, the average phosphorus level in cases of PPH was found to be 1-1.2 mg/dL. In the case report presented, although the phosphorus level was higher (2.2 mg/dL), marked hemoglobinuria was observed. This suggests that clinical signs of PPH may occur even without a severely low phosphorus level, possibly due to individual variation in susceptibility. Decreased RBC, Hb, and HCT are common in PPH, though hemoglobinuria without anemia has also been reported (6,7,12). HCT level reaches its lowest 4-9 days after the onset of hemoglobinuria (15). In this case report, the absence of anemia likely reflects the recent onset of hemoglobinuria, supported by the presence of blood in urine the day before presentation (13,15).

CONCLUSION

This report contributes valuable insights into the etiology, clinical presentation, and treatment of PPH in sheep, emphasizing the need for vigilance in nutritional planning to ensure the health and productivity of sheep flocks.

Authors contribution

Authors 1 and 2 wrote the article. Authors 1, 2 and 3 conducted the initial and follow-up treatments, as well as the laboratory analyses.

Conflict of Interest Statement

The authors declared that there are no conflicts of interest.

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