

SOME CLINICO-PATHOLOGICAL STUDIES OF CATTLE SUFFERING FROM HEMOGLOBINUREA AND ANEMIA IN KAFR-ELGALABTA VILLAGE, MENOFIA GOVERNORATE, EGYPT

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ABSTRACT

This study was carried out to evaluate some clinico-pathological changes in cattle suffering from haematourea and anemia in kafr-Elgalabta village, Menofia Governorate, Egypt. Seven crossbred cows, 3-5 year-old, presented to a private veterinary clinic with a history of parturition one month ago. The clinical signs observed and recorded as passing red colored urine since last 3 days. Animals were dull and depressed, completely anorectic with pale mucous membranes and milk yield reduced from seven liters to less than two liters. The red urine occurred about 4 weeks after parturition. Animals had been fed on a little concentrated diet with hay and Alfalfa. Some affected cows also show fever, diarrhea, and rapid respiration. Blood samples were collected from the seven cases and other samples were collected from 5 healthy cows with the same management program as control group, to determine haematological and biochemical changes during this period. Decreased erythrocyte count and hypophosphatemia were reported in the clinically affected cows. The laboratory results showed that the animals presented low phosphorus concentrations in the plasma. Animals clinically affected by haemoglobiuria showed low number of total red blood cells (TRBC). The values found for hemoglobin in the Control group were 10.7 ± 1.8 g.dL while the levels of the affected by haemoglobinuria were 7 ± 1.5 g.dL. No changes in the erythrocyte, hemoglobin or plasma phosphorus were observed in the control group.

Key words: Hemoglobinurea, Anemia, Cattle, Egypt.

INTRODUCTION

The importance and essentiality of P in ruminant diets worldwide is well established and probably is the nutrient, other than common salt (NaCl), most frequently given as a supplement to grazing ruminants (McDowell 1985). Various criteria have been used as diagnostic aids in assessing the P status of grazing ruminants. The first known response to a dietary deficiency of P is a fall in the inorganic phosphorus fraction of blood plasma (Underwood 1981).

Phosphorus, in the form of phosphate, is an essential mineral required for energy metabolism, maintenance, and function of skeletal structure, reproductive function, and lipid transport (Pond *et al.*, 2005). Phosphorus (P) is used in several body processes including critical energy pathways (ATP), cell signaling, and synthesis of cell membranes,

RNA, DNA, and bone. Because P cannot be synthesized, these needs must be met from dietary sources. Because dietary P cannot be used with perfect efficiency, a portion of dietary P is excreted, where it is subject to runoff and entry into surface waters (Wu *et al.*, 2003).

Post-parturient haemoglobinuria is a non infectious haemolytic syndrome of buffaloes and cattle which is characterized by intravascular haemolysis, anaemia and haemoglobinuria (Akhtar *et al.*, 2007a, b). The exact pathogenesis is not yet known however associated risk factors include ingestion of cruciferous plants, saponin from berseem, dietary phosphorus deficiency, decreased serum copper and selenium whereas increased molybdenum (Pirzada *et al.*, 1989; Radostits *et al.*, 2007; Neto *et al.*, 2007; Brechbuhl *et al.*, 2008).

Excess of molybdenum in soil and fodder reduces phosphorus contents of the body by interfering with its absorption from gastro-intestinal tract and increasing its elimination through urine (Akhtar, 2006). Hypermolybdenosis also induces copper deficiency leading to reduced activity of copper

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containing enzyme superoxide dismutase which is part of erythrocyte protection mechanism against oxidative stress. This process helps in rendering them vulnerable to Heinz body formation as a result of feeding on poisonous plants (Smith *et al.*, 1975; Kahn and Line, 2005 and Radostits *et al.*, 2007).

Post-parturient haemoglobinuria is a sporadic disease in high producing dairy cattle characterised by intravascular haemolysis, haemoglobinuria, and anaemia (Macwillims *et al.*, 1982). The transition between late pregnancy and early lactation, from calving until a 3 to 4 week postpartum, is a high-risk period for the occurrence of the disease in the dairy cow. The risk is especially high around parturition (Jub *et al.*, 1990; Moore *et al.*, 1997 and Whitaker *et al.*, 1999).

The inorganic phosphate level in the blood provided an indication of the dietary phosphate intake (Grunwaldt *et al.*, 2005). The disease occurs within 30 day of calving, and it is usually seen in adult dairy cattle during their third to sixth lactation (Blood *et al.*, 1989).

A variety of risk factors have been reported to be associated with this disease in different parts of the Pakistan (Khan and Akhtar, 2007; Akhtar *et al.*, 2008) the cause of the disease was dietary phosphorus deficiency, ingestion of cruciferous plants, saponin from berseem, mineral deficiency and competition of mineral absorption (Radostits *et al.*, 2007; Neto *et al.*, 2007; Brechbuhl *et al.*, 2008; Mohamed and El-Deeb, 2010). The disease is usually seen in adult dairy cattle during their third to sixth Lactation (Blood *et al.*, 1989). Copper deficiency has been suggested as a possible etiology of PPH Cattle from dairy farms with a high incidence of PPH had low levels of copper in serum and liver (Smith *et al.*, 1973). The acute disease (three to five days) can terminate in death or be followed by prolonged convalescence (two to eight weeks). Ketonuria and a depraved appetite may occur during recovery (Blood *et al.*, 1989). In affected herds, lactating but clinically normal cows have been moderately hypophosphatemic (2-3mg/ dL);

nonlactating cows usually have normal serum phosphorus concentrations (Blood *et al.*, 1989).

Treatment protocol includes the animals received phosphorus supplementation and supportive treatment. The treatment protocol for animals were designated as Tonophosphan as a source of phosphorus 25ml intra venous daily with 500ml normal saline, glucose 5% IV, and 250ml from calphomag (ca, p, mg) preparation, and 200 g of sodium Di basic phosphate powder orally with ration daily, with other supportive treatment animal recovered after one week.

MATERIALS AND METHODS

Animals

A seven crossbred cows, 3-5 year- old, presented to a private Veterinary Clinic with a history of parturition four month ago with heamoglobinurea considered as experimental group. Fife healthy cows with the same management program as control group.

Samples

For confirmative diagnosis blood samples were collected from the jugular vein before treatment. An aliquot of blood was placed into an EDTA-containing plastic tube for haematocytometry (Feldman *et al.*, 2000). The rest of the blood samples were placed into glass tubes for the serum. These tubes were centrifuged and harvested serum was immediately frozen at -20°C until analysis. The serum total protein (TP) (Henary *et al.*, 1974), albumin (Alb) (Doumas *et al.*, 1971), calcium (Ca), inorganic phosphorus (P), magnesium (Mg), glucose (Trincler., 1961), were measured by an autoanalyser. The blood smears were stained by Giemsa's stain and then examined for blood parasites in erythrocytes under a light microscope.

Statistical analyses

Statistical differences were calculated according to the Student t-test with significance level at $P < 0.05$. All results were analyzed using the procedure of (SAS, 2004).

RESULTS

Table 1: Hematological and some biochemical parameters (mean \pm SD) in cattle's with post-parturient haemoglobinuria and control.

Parameters	Clinically affected	Clinically normal (control)	Reference range Radostitis (2007)
P (mg/dl)	3.1 \pm 0.79	5.9 \pm 1.15	5.6 – 6.5 mg/dl
Ca (mg/dl)	7.8 \pm 0.36	11.3 \pm 0.31	9.7 – 12.4 mg/dl
Mg (mg/dl)	0.75 \pm 0.28	1.5 \pm 0.26	1.8 – 2.3 mg/dl
Glucose (mg/dl)	30 \pm 13.2	50 \pm 12.4	45 – 75 mg/dl
Alb (g/L)	2.4 \pm 0.65	2.5 \pm 0.97	2.1 – 3.6 g/dl
TP (g/L)	5.6 \pm 1.0	7.2 \pm 2.5	5.7 – 8.1 g/dl
TRBC ($\times 10^6 / \mu\text{L}$)	2.06 \pm 1.5	6.08 \pm 3.2	5 – 10 ($\times 10^6 / \mu\text{L}$)
Hb (g/dl)	7 \pm 1.5	10.7 \pm 1.8	8 – 15 g/dl

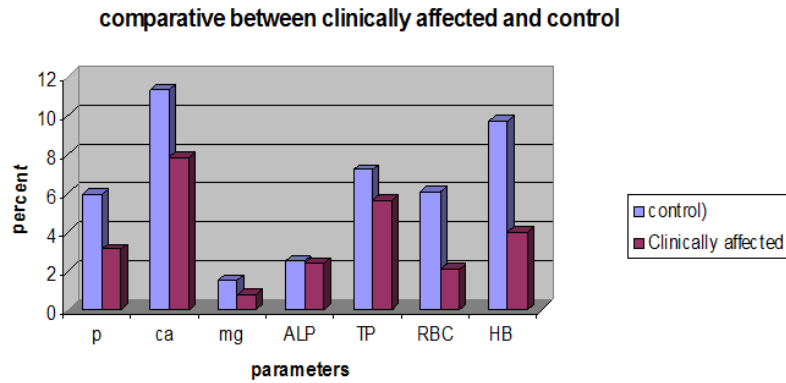


Figure 1: Haematological and some biochemical parameters (mean \pm SD) in the cattle with post-parturient hemoglobinuria and control.

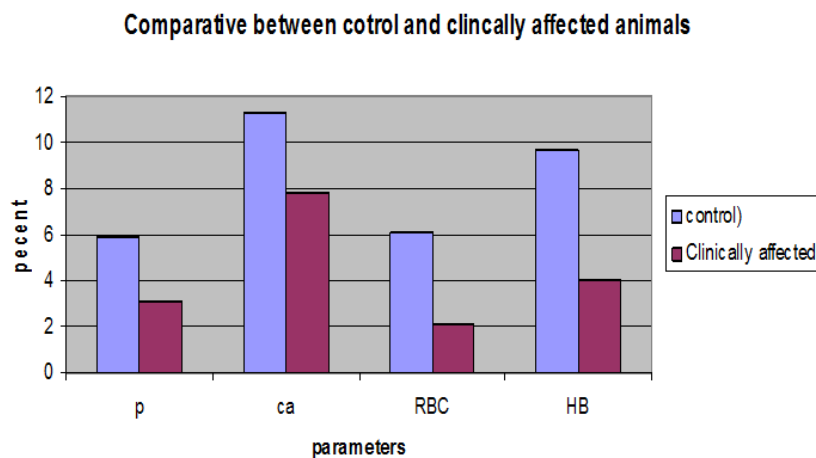


Figure 2: Comparison between recorded value in control and clinically affected animals.

In this cases reports, (**Table 1**) & **Fig (1, 2)** anorexia, depression, decreased ruminal movement, and milk production, moderate tachycardia, pale mucosa membranes, and haemoglobinuria were detected in caws. Moreover, determined that serum phosphorus was very low.

DISCUSSION

It is known that phosphorus deficiency can reduce the adenosine triphosphate content in red blood cells, influencing the structure and function of the cell, thereby increasing fragility and haemolysis, which may lead to acute haemoglobinuria (Wang *et al.*, 1985). In this case, anorexia, depression, decreased ruminal movement, and milk production, moderate tachycardia, pale mucosa membranes, and haemoglobinuria were detected in studied cows. Moreover, we found serum phosphorus was very low. (Jub *et al.*, 1990) reported that circulating oxidants may cause erythrocyte damage, which may be predisposed to hypophosphataemia. The phosphorus deficiency commonly occurs two to four weeks after calving. Stockdale *et al.* (2005) and fat cows had acute haemoglobinuria two to four weeks after

calving. In the current study, detected that post-parturient haemoglobinuria occurred about 6 weeks after calving. No blood parasites were detected in cattle with post-parturient haemoglobinuria. Phosphorus is an essential element for cellular function. It is absorbed in the small intestine and excreted either via the faeces or urine. Factors reducing the flow of saliva, such as anorexia, may cause increased losses of phosphorus via urine Reinhardt *et al.* (1988). Phosphorus is necessary for glycolysis, the formation of adenosine-triphosphate, and is also required for the maintenance of the erythrocyte cell membranes. Low serum inorganic phosphorus is a frequent finding in cattle with postparturient haemoglobinuria McCaughan, (1993). Stockdale *et al.* (2005) reported that marginal dietary phosphorus deficiency may leave cattle predisposed to acute haemoglobinuria. Thompson and Badger

(1999) found that the serum phosphorus level in the cattle with haemoglobinuria was markedly decreased. In addition, Karapınar *et al.* (2006) also detected that the serum phosphorus level in cows with haemoglobinuria was very low (0.5 and 1.5 mg/dL). In this study, the mean serum phosphorus level in 3-5 years old cattle's with post-parturient haemoglobinuria was lower Table (1) and Fig 1 and 2. The cause of postparturient haemoglobinuria in this case may be related to dietary phosphorus deficiency. Since the cattle had been fed on a little concentrated diet of hay, and alfa-alfa which contains low amounts of phosphorus. Moreover, we reported that the serum glucose, Ca, Mg, TP, Alb and capacity concentrations in this case before and after treatment were within the normal range limits.

Decreased total erythrocyte count, hemoglobin concentration in affected cattle's indicating severe anemia which is attributed to intravascular haemolysis in this disease. The findings with respect to haematological parameters of affected cattle are in agreement with Dalir- Naghadeh *et al.* (2005); Akhtar (2006); Khan and Akhtar (2007); Radwan and Rateeb (2007) and Durrani *et al.* (2010).

Decreased serum inorganic phosphorous in affected cattle may be associated with prolonged feeding on cruciferous and or/toxic plants particularly berseem, heavy drainage of phosphorous through milk in high producing animals maintained on low phosphorous diets, low phosphorous rations with high calcium content and hypermolybdenosis. Low phosphorous diets with high calcium content cause hypophosphataemia by decreasing phosphorous absorption from the gastro-intestinal tract due to wider ratio of calcium and phosphorous whereas; excess of molybdenum also decreases phosphorous content by interfering with its absorption and increasing its elimination through urine Bhikane *et al.* (2004); Akhtar, (2006); Khan and Akhtar, (2007) and Dua, (2009).

From present study it can be concluded that addition of sodium dibasic phosphate to the diets of susceptible animals to haemoglobinurea (cattle after parturition), must ensure that the ratio between calcium and phosphorus not more than 2: 1 in the ration, with providing of treatment to clinically affected cases with phosphorus preparation as cafozal, tonophosphan and calgophos, especially in the winter season when the alfalfa (which is of poor in phosphorus) is the main meal for cows.

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بعض الدراسات الاكلينيكية على الحالات المرضية في الماشية التي تعاني من نزول البول مدمم وفقر الدم في قرية
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قد أجريت هذه الدراسة لتقييم بعض التغيرات المرضية الاكلينيكية في الماشية التي تعاني من تدمم البول وفقر الدم في قرية كفر الجلابطة ، محافظة المنوفية، مصر. سبع بقرات هجين، أعمارهم من ٣-٥ سنة قدموا إلى إحدى العيادات البيطرية الخاصة بعد شهر واحد من تاريخ الولادة. وكانت الاعراض العيادية نزول البول مدمم بلون أحمر منذ الثلاثة أيام الماضية. كانت الحيوانات محبطة وقليلة الحركة مع وجود شحوب بالأغشية المخاطية وانخفاض إنتاج الحليب من سبعة لترات إلى أقل من اثنين لتر. مع وجود ظاهرة البول المدمم (haemoglobinuria) حوالي ٤ أسابيع بعد الولادة. كانت تتم تغذية الحيوانات على عليقة مركزة وقليل من القش والبرسيم أظهرت بعض الأبقار المتضررة حمى، وإسهال، وسرعة في التنفس. تم جمع عينة الدم من الحالات السبع وتم جمع عينات أخرى من ٥ أبقار صحيحة اكلينيكية كمجموعة ضابطة مع العلم انهم يعيشون في نفس الظروف المعيشية. تم جمع العينات لتحديد التغيرات الدموية والكيموحيوية خلال هذه الفترة. ومن النتائج تبين نقص عدد كرات الدم الحمراء ونقص فوسفات الدم في الأبقار المتضررة سريريا. أظهرت النتائج المخبرية أن الحيوانات المصابة سريريا انخفاض العدد الكلي لخلايا الدم الحمراء (RBC). وكانت القيم منخفضة في مصل الدم (السيرم) ، أظهرت الحيوانات المصابة سريريا انخفاض العدد الكلي لخلايا الدم الحمراء (RBC). وكانت القيم التي تم تسجيلها للهيموجلوبين في المجموعة الضابطة 10.7 ± 1.8 g.dL في حين أن مستويات الهيموجلوبين في الحيوانات المصابة كانت 7 ± 1.5 g.dL. في حين لم تحدث تغيرات في كرات الدم الحمراء والفوسفور في مصل الدم (السيرم) في المجموعة الضابطة. تم التوصية بإضافة الصوديوم ثنائي الفوسفات إلى علائق الحيوانات المعرضة للإصابة بأمراض نقص الفسفور والحرق على أن لا تزيد النسبة بين الكالسيوم والفسفور عن ٢ : ١ وعلاج الحالات المرضية بمركبات الفسفور كمستحضرات كافوزال والتونوفسفان والكالجوفوس وخصوصا في موسم الشتاء وعندما يكون البرسيم (وهو من الأغذية الفقيرة بالفسفور) هو العلف الرئيسي للأبقار.