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## Clinical and Laboratory Studies on Diarrhea Problem in Newborn Calves

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### **Abstract**

The study was carried on a total number of 130 calves. From them 53 buffalo calves were held in the animal farm station of Faculty of Veterinary Medicine, Suez Canal University and 77 Frisian cattle calves were held in El-Salhya private farm, Ismailia, Egypt. Two control groups 9 from the faculty farm and 11 for El-Salhya farm were used from each group. All groups were clinically examined for presence of fever, signs of diarrhea and dehydration. Blood with and without anticoagulant and fecal samples were collected from both diseased and control calves at the same time. Heparinized blood was used for blood picture. Clear serum samples were used for biochemical analysis of sodium, potassium, chloride, total proteins, albumin and globulins as well as Protein electrophoresis pattern was performed to detect the level of each protein fraction. Fecal samples were examined microscopically to detect the presence of parasitic infestation. According to the severity of dehydration the diseased calves were divided into three groups mild, moderate and severe one. Diseased calves showed mild to severe diarrhea and few cases showed signs of dehydration. Hematological findings revealed no significant variations in hemoglobin values and leukocyte counts but RBCs counts showed a significant decrease in mild cases of diarrheic calves; however PCV showed a highly significant increase in severe diarrheic calves in both farms. Biochemical analysis of serum showed no significant differences in sodium and potassium, while chloride showed highly significant increase in severe diarrheic buffalo calves. Total proteins showed a significant increase in severe diarrheic buffalo calves in the faculty farm, while albumin showed a significant increase in severe group of El-salhya farm, also globulin showed a significant increase in all diseased buffalo calves. Protein electrophoresis revealed that only  $\alpha_1$  globulins showed highly significant increase in diseased calves. No severe blood changes were recorded. The obtained result support the idea that mild diarrhea does not affect blood parameters in early stages of the disease.

**Keywords:** Diarrhea, Newborn calves, hematological findings, proteinelectrophoresis, electrolytes and dehydration.

### Introduction

Diarrhea is a serious problem and considered the most common disease inducing high morbidity and mortality rates in newborn calves and responsible for severe economic losses. Moreover, *Williamson (2002)* stated that diarrhea is a common complaint in calves and other young ruminants, particularly in the first few months of life.

Signs of diarrhea include anorexia, loss of weight, and hemorrhagic and/ or mucoid diarrhea (*Georgi, 1985*). *Radostits, et al (2007)* recorded that in severe cases, feces are liquid, bloody and may contain strands of intestinal mucosa, and animals may become emaciated, dehydrated, weak, and listless.

*Guzelbektes et al (2007)* reported a decrease of blood plasma sodium and an increase of blood plasma potassium concentration in calves between one to five months of age affected by diarrhea. However, a decrease in blood plasma chloride concentration in these calves was observed only when animals were highly dehydrated. *El-Sangary et al (2008)* recorded different variations in total serum protein and serum protein electrophoresis between healthy and diarrheic enteric dehydrated buffalo calves. The variations included a significant decrease of serum total protein, albumin and albumin

globulin ratio, a non significant decrease of  $\beta$  globulin, a non significant increase of  $\alpha$ , gamma and total globulins. *ElSheikh et al (2012)* recorded hyponatremia, hypochloremia, hyperkalaemia and hyperproteinemia in dehydrated diarrhaic newborn Friesian calves. *Malik et al (2012)* reported that there was a significant increase in hematological parameters like packed cell volume and total leukocyte counts in diarrheic calves in comparison to apparently healthy calves which was suggestive of dehydration. *Malik et al (2013)* reported that the effects of diarrhea on fluid loss, imbalance of electrolyte and acid base are always governed by the type, duration and severity of diarrhea. However, different variations in clinical symptoms, blood, serum analysis of total serum protein fractions were previously recorded (as mentioned before) so that the aim of the current study is to identify the variations in the clinical manifestation of diarrhea in cattle and buffalo calves and exploring the effect of diarrhea on some hematological and serum parameters especially the changes in serum electrolytes levels and total serum proteins and protein electrophoretic pattern.

### Material and methods

The total number of diseased calves used in this investigation was 130 calves, 53 buffalo calves in the faculty farm and 77 Frisian calves in El-Salhya farm. They are divided into 3 groups, mild, moderate and severe diarrhea. Their age varied from birth up to 3 months. The number of control clinically healthy calves in faculty farm was 9 buffalo calves and in El- Salhya farm was 11 frisian calves.

Clinical Examination of the animals and Skin fold test was applied according to *Radostits et al (2006)*. Blood and fecal samples were taken from each calf of diseased and control groups. A blood sample was collected with anticoagulant (Potassium EDTA) for complete blood picture (*Jain, 1986*). Another blood sample was taken without anticoagulant to obtain serum for the biochemical analysis (*Coles, 1986*). The hematological procedures were carried out according to (*Jain, 1986*) and hemoglobin (HB) was colourimetrically determined according to *Young (2001)*.

Serum levels of sodium and potassium were estimated colorimetrically according to *Henry et al (1974)* for sodium and *Tietz (1976)* for potassium. Serum levels of chloride, total proteins and albumin levels were estimated colorimetrically according to *Young (2001)*. The globulin concentration was calculated by subtracting albumin from total proteins. Serum

protein electrophoretic pattern was done according to *Woolf et al (1973)*.

Fecal samples were collected in a plastic vials directly from the rectum and examined microscopically by direct and simple flotation methods to detect presence of parasitic infestation.

The statistical analysis was carried out according to *Snedecor and Cochran (1975)*, t-test in pairs was used for detection of significant differences.

### Results

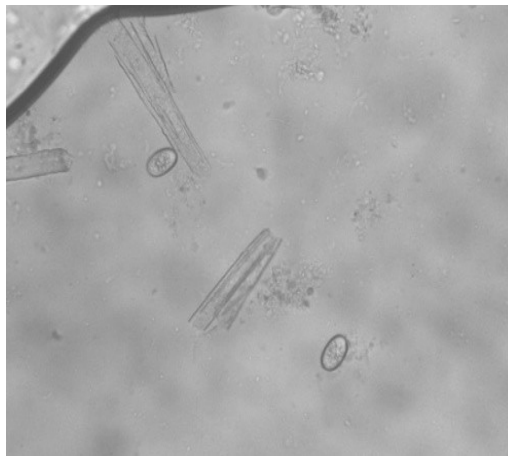
The study revealed that the clinical signs of infected calves showed mild to severe diarrhea, few cases of dehydration. Tenesmus was observed on some cases and fever was evident in a few numbers. The state of appetite was different according to the severity of illness. Feces was varied in consistency from pasty to watery, and varied in color from yellow to yellowish brown or green and sometimes contain blood or mucus. Body temperature, respiratory and heart rates in both farms showed no significant differences between diseased and control groups (table 1).

The laboratory investigations of fecal samples by direct and flotation revealed that only five cases infected with *Eimeria* spp. (picture. 1).

Blood pictures, serum analysis of total protein fractionation are tabulated in table 2, 3 and 4.

**Table 1:** Mean± S.E of clinical signs of healthy control and diarrheic of buffalo and Frisian calves:

Farm	Measures	Control	Diseased calves		
			Mild	Moderate	Severe
Faculty farm(buffalo calves)	Temperature (°C)	38.7± 0.1	38.8± 0.1	38.8± 0.22	39.2± 0.23
	Respiratory rate (No./minute)	24± 0.7	23.8± 0.34	22.42± 0.7	22.1± 1.1
	Heart rate(No./minute)	81± 1.2	71.5± 0.51	83.5± 0.49	75.2± 1.68
El – Salhya farm(Frisian calves)	Temperature(°C)	38.8± 0.12	38.9± 0.08	38.9± 0.15	38.7± 0.6
	Respiratory rate(No./minute)	24.3± 1.1	23.3± 0.86	24.11± 0.7	23.6± 0.73
	Heart rate(No./minute)	64.4±0.86	72.1± 1.8	83.7± 0.62	83.4± 0.67



**Figure 1:** *Eimeria* oocyst under microscope after examination of fecal sample of diarrheic buffalo calves by simple flotation.

**Table 2:** Mean± S.E of hematological results of healthy control and diarrheic of buffalo and Frisian calves:

Farm	Measures	Control	Mild	Moderate	Severe
<b>Faculty farm(buffalo calves)</b>	RBCs( $\times 10^6/\mu\text{l}$ )	7.6± 0.3	6.2*± 0.2	7.4± 0.2	6.6± 0.4
	Hb (g/dl)	10.26± 0.36	9.64± 0.23	11.39± 0.41	9.59± 0.58
	PCV (%)	31.9± 0.63	25.2± 0.58	31.5± 0.3	36.9**± 0.98
	MCV(fl)	42.59± 1.6	41.87± 2.1	44.19± 1.2	56.17**± 4.39
	MCH (pg)	13.75± 0.77	15.87± 0.57	15.94± 0.70	14.98± 1.07
	MCHC (g/dl)	32.3± 1.38	38.49**± 1.03	36.16± 1.27	27.14± 2.20
	Total leukocyte ( $\times 10^3/\mu\text{l}$ )	10.03± 0.5	10.01± 0.64	15.2± 4.6	10.5± 0.7
<b>EI – Salhya farm(Frisian calves)</b>	RBCs( $\times 10^6/\mu\text{l}$ )	7.3± 0.4	5.8± 0.3	6.7± 0.3	9.4± 0.3
	Hb (g/dl)	10.15± 0.72	9.5± 0.15	9.79± 0.54	11.92± 0.52
	PCV (%)	30± 0.73	26.2± 0.53	30.7± 0.26	36.7**± 0.52
	MCV (fl)	42.95± 2.90	47.2± 1.99	46.92± 2.01	53.29**± 2.22
	MCH (pg)	14.21± 0.81	17.19*± 0.71	14.85± 0.94	16.55*± 0.72
	MCHC (g/dl)	33.68± 1.82	36.81± 1.09	31.82± 1.67	31.31± 1.21
	Total leukocyte ( $\times 10^3/\mu\text{l}$ )	7.7± 0.6	9.57± 0.56	11.3± 5.7	9.4± 0.5

\* Significant

\*\* highly significant

**Table 3:** Mean± S.E of biochemical results of healthy control and diarrheic of buffalo and Frisian calves:-

Farm	Measures	Control	Diseased calves		
			Mild	Moderate	Severe
Faculty farm(buffalo calves)	Sodium (mEq/l)	135.4± 7.4	131.7± 4.25	130± 3.7	136.3± 2.8
	Potassium(mEq/l)	5.05± 0.3	5.2± 0.2	5.3± 0.17	5.5± 0.3
	Chloride (mEq/l)	114.4± 1.8	117.8± 1.99	118.4± 2.24	123.7**± 1.98
	Totalprotein(g/dl)	6.4± 0.36	7.1± 0.2	6.62± 0.24	7.3*± 0.41
	Albumin (g/dl)	5.6± 0.44	4.49± 0.18	4.26± 0.2	4± 0.43
	Globulin (g/dl)	0.83± 0.71	2.56*± 0.25	2.37*± 0.28	3.81*± 0.64
El – Salhya farm(Frisian calves)	Sodium (mEq/l)	141.36± 3.5	132.6± 3.4	135± 3.2	130.06± 4.6
	Potassium (mEq/l)	4.7± 0.29	5.2± 0.19	5.2± 0.2	4.9± 0.2
	Chloride (mEq/l)	104± 1.15	105.5± 0.58	105.9± 1.32	102.5± 1.59
	Total proteins(g/dl)	5.73± 0.22	5.55± 0.16	5.61± 0.14	5.81± 0.21
	Albumin(g/dl)	3.29± 0.1	3.36± 0.09	3.14± 0.08	3.58*± 0.08
	Globulin(g/dl)	2.45± 0.28	2.17± 0.14	2.2± 0.15	2.23± 0.2

\* Significant

\*\* highly significant

**Table 4:** Mean± S.E of protein electrophoresis results of healthy control and diarrheic of buffalo and Frisian calves:-

Farm	Measures	Control	Diseased
Faculty farm(buffalo calves)	Alpha-1- (%)	0± 0	0.841**± 0.841
	Alpha-1- (g/dl)	0± 0	0.057± 0.057
	Alpha-2- (%)	14.4± 3.3	15.2± 2.12
	Alpha-2-(g/dl)	1.03± 0.18	1.05± 0.16
	Beta (%)	11.3± 0.14	16.06± 1.33
	Beta (g/dl)	0.83± 0.051	1.08± 0.09
	Gamma( %)	30.4± 8.2	20.5± 1.5
	Gamma (g/dl)	2.3± 0.79	1.38± 0.11
El – Salhya farm(Frisian calves)	Alpha-1- %	5.41± 3.3	2.2**± 0.86
	Alpha-1- (g/dl)	0.31± 0.18	0.123± 0.04
	Alpha-2- (%)	14.6± 3.2	13.8± 1.2
	Alpha-2-(g/dl)	0.88± 0.21	0.84± 0.07
	Beta (%)	18.7± 0.97	17.5± 0.63
	Beta (g/dl)	1.12± 0.11	1.07± 0.04
	Gamma %	20.4± 3.9	21.8± 1.59
Gamma (g/dl)	1.26± 0.29	1.34± 0.11	

\*\*highly significant

## Discussion

In this investigation, it was observed that mild to severe diarrhea was constant signs. Fever, anorexia, dehydration were observed in some cases. Fecal consistency was varied from pasty to watery and the color was varied from pale yellow to dark brown and in some cases contain blood or mucus. Soiling of hind quarters and tail obviously detected in most cases. Sunken eyes was observed in some cases as a result of dehydration. These results coincided with those reported by *Malik, et al (2012)*. Emaciation was observed among diseased calves and emaciation signs were observed such as sunken eyes, prominent bony structures as ribs. Similar results were obtained by *Georgi (1985) and Ernst and Benz (1986)*. Skin of diseased calves was dry and wrinkled in some cases and this finding was similar to those reported by *Radostits et al (2007)*. The hematological results (table 2) were varied from group to another according to the severity of diarrhea. Leukocyte and hemoglobin showed no significant variations but RBCs and PCV showed a significant increase. The obtained results were agreed with *Hafez (1979)* who found that there were no significant variations in Hb and WBCs between diarrheic parasitized and healthy calves. However, *Deshpande et al (1993)* observed a significant increase in total

erythrothitic count, hemoglobin and PCV values in diarrheic calves.

The obtained results of blood indexes indicate macrocytic anemia in severe groups in both farms according to *Jain (1986)*.

Serum levels of sodium (table 3) showed no significant variations between control and diseased calves in both buffalo and cattle calves. These results agreed with those reported by *Seifi et al (2006)* who did not observe any change in serum sodium concentration in two-weeks old calves with diarrhea but disagreed with mean values obtained by *Ali (1987)* who found a significant decrease in both values of serum sodium and serum chloride ( $136.3 \pm 15.2$  mmol/L and  $91.3 \pm 16.16$  mmol/L respectively) in a group of calves showing symptoms of enteritis. *Dalton et al (1965) and Fisher (1965)* reported also a fall in serum sodium level in prolonged diarrhea that may explain the non significant decrease in sodium level among the observed diseased calves in both farms, where the case lasted for few time before treatment.

Serum potassium results (table 3) revealed no significant differences between diseased and healthy calves. Similar findings were previously reported by *Mcscherry and Grinyer (1954) ; Fisher (1965) and Coles (1980)*. These findings may be explained on the bases that most body potassium is present in

the intracellular fluid (ICF) and there is non constancy of relationship between extracellular fluid potassium and that of intracellular one, there for the level of serum potassium may not reflect the true status of the body potassium concentration (*Coles, 1980 and Duncan and Prase, 1986*).

The result of serum chloride levels (table 3) showed no significant differences between diarrheic and control calves in all groups except in the severe group of the faculty farm, it recorded highly significant increase in chloride values when compared with healthy control calves, and these results disagreed with those obtained by *Ali (1987)* who found a significant decrease in both values of serum sodium and chloride ( $136.3 \pm 15.2$  mmol/l and  $91.3 \pm 16.16$  mmol/l respectively) in calves showing symptoms of enteritis. The author noticed that the reduction of serum chloride levels in most cases of diarrhea was associated with a decreased level of sodium. This reduction was explained early by *Tasker (1969)* who stated that serum chloride is usually follow that of sodium because serum chloride is usually found in a form of sodium chloride. Regarding the hematological and serum biochemical findings, it was observed that minor differences were observed which were not statistically significant in most parameters. This was explained by *Radostits et al (2007)* who stated

that a loss of fluid due to diarrhea is usually started from the intercellular spaces in the early period of diarrhea in calves, however, loss of fluid from circulation starts in late stages of diarrhea especially in subacute and chronic diarrhea. That means the circulation keeps its fluid longer periods than intercellular fluid. Concequently, hematological and biochemical changes could be neglected in the early stages as it was supported by the results of present study.

Statistically the results of total proteins (table 3) showed no significant differences in serum total proteins in all groups except the severe group of the faculty farm there were significant increase when compared with healthy control calves. This disagreed with results of *Dawood (1993) and Connell et al (1969)* who reported that calves with diarrhea lost more serum proteins via the intestinal tract resulting in lowering of total serum proteins, while *Varely (1976)* recorded an increase in total serum proteins in dehydration, and both albumin and globulins levels are increased because of hemoconcentration. An increase in total serum proteins was recorded also by *Zayed (1998)*.

It was observed that results of serum albumin (table 3) values showed no significant variations except the severe group of El-Salhya farm there were a significant increase which agreed those of *El-kabbani et al (1987)* who stated that



, there was a significant increase in albumin as a result of dehydration. And disagreed those reported by *Abdel-Mottelib (1972)* who observed hypoproteinemia and hypoalbuminaemia associating diarrhea in calves.

Serum Globulin (table 3) in all groups of the faculty farm revealed a significant increase in comparison to control ones. And this agreed with *Varely (1976)* who reported that both albumin and globulins levels are increased because of hemoconcentration in case of dehydration. However, the obtained results disagreed with those reported by *Connell et al (1969)* who stated that in enteric calves the significant drop of globulin values is due to loss of serum protein via the intestinal tract.

The electrophoretic pattern (table 4) revealed that alpha -1- globulin recorded a highly significant increase in diseased group in comparison to control ones in the faculty buffalo farm . The obtained results agreed those reported by *El-Kabbani et al (1987)* who attributed the significant increase in serum  $\alpha$  - globulins in diarrheic calves to severe dehydration. This increase was explained by (*Jain, 1986*) who reported that acute phase protein such as alpha globulin is increased in acute inflammatory condition. Serum beta-globulins recorded no significant variations between healthy and diseased calves in both farms. These results agreed those obtained by *Dawood (1993)* and

disagreed with those reported by *Kishtwaria et al (1983)*. Serum gamma -globulins recorded no significant differences between diseased and control buffalo and cattle calves. The obtained result disagreed those recorded by *Affonso et al (1960) and coles (1980)* who mentioned an increase of gamma globulins in infectious diarrhea due to invasion of body by bacterial or viral agent.

It could be concluded that diarrhea is still a major problem facing buffalo and cattle calves in both farms. The effect of diarrhea was minor or neglected in early stages on the studied hematological picture and blood serum constituents. Depending on the obtained results, the early interfere by the suitable methods of treatment in both farms prevent the complication of diarrhea.

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### الملخص العربي

## دراسات اكلينيكية ومختبرية على مشكلة الإسهال في العجول حديثي الولادة

ثروت نافع ، اسماء عمر على ، دينا امين عبد الخالق

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اجريت هذه الدراسة على عدد اجالى ١٣٠ عجل منهم ٥٣ فحل جاموس في مزرعة كلية الطب البيطري -جامعة قناة السويس و٧٧ عجل بقرى حديثو الولادة في مزرعة الصالحة بمحافظة الاسماعيلية. تم اختيار مجموعتين ضابطين من كلا المزرعتين من العجول السليمة اكلينيكيًا. كل المجموعتين فحصت اكلينيكيًا لوجود اعراض الحمى او الاسهال او الجفاف. تم أخذ عينات دم على مائع للتجلط ودم بدون مائع للتجلط لفصل السيرم وعينات براز من كل الحيوانات المريضة و السليمة . وقد استخدمت عينات الدم لعمل صورة دم واستخدم سيرم الدم لاجراء بعض التحاليل البيوكيميائية مثل الصوديوم و البوتاسيوم و الكلوريد و البروتين الكلي والالبومين والجلوبولين . وايضا تم الفصل الكهربى للبروتينات. اما عينات البراز فقد استخدمت في اجراء الفحص المجهرى للكشف عن بويضات بعض الطفيليات. وقد اظهرت النتائج وجود بعض الاعراض الاكلينيكية و اوضحت الدراسة ان العجول المريضة تعانى من اسهالات خفيفة الي شديدة وبعض الحالات تعانى من الجفاف . وقد وجد ارتفاع طفيف فى درجات الحرارة فى بعض الحالات . وقد لوحظ اختلاف فى قوام البراز من عجيني الى مائى واختلف اللون من اصفر الى اصفر مخضر مصاحب احيانا بالمخاط او الدم. واطهرت الدراسة انه لا يوجد اختلاف معنوى بين العجول المريضة والمصابة بالاسهال فى كل من درجات الحرارة ومعدل التنفس ومعدل ضربات القلب. يفحص عينات البراز تم الكشف عن وجود خمس عجول مصابة بالاييميريا. وبالنظر الى نتائج فحص الدم اوضحت الدراسة عدم وجود اختلاف معنوى فى كرات الدم البيضاء والهيموجلوبين مع وجود اختلاف معنوى فى كرات الدم الحمراء وحجم خلايا الدم الكلى ومتوسط حجم الخلايا ومتوسط تركيز الهيموجلوبين ومتوسط وزن الهيموجلوبين لكل خلية بين مجموعات الحيوانات المريضة والمجموعة الضابطة. كما اوضح تحليل سيرم الدم عدم وجود فروق معنوية في عناصر الصوديوم والبوتاسيوم ولكن توجد بعض الفروق المعنوية في الكلورايد و البروتين الكلى والالبومين ويوجد اختلاف معنوى فى الجلوبيولين بين مجموعات الحيوانات المريضة والسليمة. وبخصوص الفصل الكهربى للبروتين لم يلاحظ اى فروق معنوية الا فى حالة الالف-١ -جلوبيولين بين المجموعات المريضة والمجموعة الضابطة.