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Analysis of hematological changes in normal and diarrhea calves

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Abstract

In order to effective treats a sick calf, it is necessary to obtain accurate information about the patient's condition. However, the standard references for Korean cattle claves are not well known. Therefore, this study aims to present useful clinical values by analyzing normal blood and diarrhea blood of Hanwoo calf. Recently, with the advent of a portable blood analyzer, it is possible to immediately analyze the patient's condition and severity in the field, not in the laboratory, and to calculate a suitable dosage for supporting fluid therapy. Therefore, in this study, the distribution of red blood cells (RBC) and white blood cells (WBC) were analyzed in normal and diarrhea Hanwoo calves. As a result, Hematocrit (HCT) levels increase significantly between 1 and 20 days in diarrhea positive calves. Changes in leukocyte composition had similar growth patterns in normal and diarrhea calves. As it grew, the proportion of neutrophils decreased and lymphocytes increased. However, the number of WBCs increased from 1 to 10 and 21 to 30 days in diarrhea positive calves, which is closely related to the increase in neutrophils. Therefore, those data can be used for diagnosis and treatment of diarrhea calf.

Key words : Calf diarrhea, RBC, WBC

INTRODUCTION

Calf diarrhea is the most important factor in the death of newborn calves and can cause serious economic losses to livestock farms. Therefore, the causes of calf diarrhea have been studied a lot, especially studies on the development of rapid diagnostic methods for infectious pathogens and finding out the correlation between such pathogens and diarrhea have been reported. Acres et al. (1977) investigated the prevalence of ETEC and Rota virus in acute calf diarrhea and Don et al. (1980) studied on pathogenicity for the fetal calf following maternal infection. But only epidemiologic investigations of disease outbreaks have been reported in newborn calves of Korean native cattle (Hanwoo) (Kang et al, 2001; Kang et al, 2004; Kim et al, 2015).

On the other hand, the disease causes the loss or accumulation of water and major electrolytes in the body depending on the cause. Especially in newborn calf within 1 month of low weight, these changes can be even more fatal to life (Done et al, 1980; Tzipori et al, 1980). Therefore, accurate and rapid analysis of changes in these components depending on the severity of the disease is required (Kwon et al, 2000; Yimer et al, 2015). In other words, biometric information, which can quickly understand the state of health is also important, as well as the diagnosis of the pathogen, for the effective treatment and management of sick calf.

However, most reports on bovine blood levels and serum biochemistry have been conducted only in adult cattle (Grove-White and White, 1993; Hartmann and Reder, 1995; Constable et al, 2004). According to Bradford P. Smith, detailed normal ranges of bovine blood and electrolytes is presented, but no reference depending on the age of calf is provided. In addition, Hege C. Brun-Hansen

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et al. reported changes of blood levels and major electrolyte values in healthy or diseased newborn calves, but the study target was a dairy breed of the Norwegian Red species (Acres et al, 1977; Hartmann et al, 1997). Therefore, the information whether such data can be directly applied to Korean cattle calves is lacked. So, the standard values of adult cows are inevitably used for health management and disease status investigation of young calves within 1 month of age, the highest mortality rate.

For effective treatment and management of diseased newborn calves, the application of these (adult cattle) references to young calves has its limits. Therefore, in this study, the standard indicators for biometric data such as blood levels and major electrolytes values of healthy Hanwoo calves are divided into growth stages and presented and it will be useful in industrial animal clinical medicine.

MATERIALS AND METHODS

Animals and sample collection

179 newborn Korean native cattle (Hanwoo) calves within 30 days of birth were used. According to the growth stage (age), it was divided into 3 stages: 1 to 10 days (n=51), 11 to 20 days (n=57), and 21 to 30 days (n=71).

Diarrhea was clinically determined by physical examination such as stool status, body temperature, and degree of dehydration (Table 1). The stool obtained by inducing bowel movements by rectal stimulation were not contaminated with the floor environment and were immediately transferred to the laboratory in a frozen state for further fecal examination.

Table 1. Classifications and numbers of calves used in this study divided into groups by age

	1～10 days	11～20 days	21～30 days	Total
Healthy	18	36	46	100
Diarrheic	33	21	25	79
Total	51	57	71	179

Determination of diarrhea calves by fecal examination

Fecal samples were examined with RT-PCR and calf diarrhea 5-point antigen diagnostic kit (BoviD-5 Ag). After the antigen test for Rotavirus, Coronavirus, BVDV, E. coli, Salmonella spp. Cryptosporidium and Giardia, the main pathogens causing calf diarrhea, the negative subjects were selected as control group.

Complete blood cells counts (CBC)

The whole blood collected in the anticoagulant (EDTA) tube was transferred to the laboratory in a refrigerated state, and the CBC test was performed within 3 hours. Blood analysis was performed using IDEXX ProCyte Dx (IDEXX Laboratories). RBC profile tests include RBC counts, hematocrit (HCT), hemoglobin (Hb), MCV, MCHC and red blood cell distribution (RDW) and WBC profile tests include total WBC count, total number of neutrophils, lymphocytes and their percentages.

RESULTS

Identification of pathogens from diarrheic calves feces

From the 79 samples of diarrheic calves tested, the pathogens were detected 36 (45%) to be positive for rotavirus, 13 (16%) for BVDV, 13 (16%) for *E.coli*, 10 (14%) for *Eimeria* spp., 4 (5%) for *C. parvum*, and 3 (4%) for coronavirus, whilst no samples were found to be positive for *Giardia duodenalis* and *Salmonella* spp.

Red blood cells profiles analysis

The Red blood cells profiles of the calves in each group are listed in Table 2. The mean RBC counts, HCT and Hb were significantly increased in most diarrhea positive groups according to age compared with the diarrhea negative group (Table 2). The mean RBC counts ($\times 10^6/\mu L$) was as follows respectively; 8.7 (±1.2) in 1～

10 days old, 8.8 (±1.2) in 11～20 days old and 10.0 (±1.4) in 21～30 days old in diarrhea negative calves group; 11.1 (±2.4) in 1～10 days old, 10.9 (±2.2) in 11～20 days old and 11.1 (±2.5) in 21～30 days old in diarrhea positive calves group. The mean value of total was 9.4 (±1.5) in healthy calves and 11.0 (±2.4) in diarrhea calves. Also, HCT (%) was as follows respectively; 31.7 (±5.9) in 1～10 days old, 29.8 (±5.4) in 11～20 days old and 35.4 (±7.2) in 21～30 days old in negative group; 43.8 (±11.1) in 1～10 days old, 40.1 (±10.8) in 11～20 days old and 39.0 (±8.0) in 21～30 days old in positive group. The mean HCT (%) of total was 32.7 (±6.8) in negative

Table 2. RBCs in diarrhea negative/positive calves according to age

	1~10 days	11~20 days	21~30 days	1 month
RBC ($\times10^6$/μL)				
Diarrhea (−)	8.7±1.2	8.8±1.2	10.0±1.4	9.4±1.5
Diarrhea (+)	11.1±2.4†	10.9±2.2‡	11.1±2.5*	11.0±2.4‡
HCT (%)				
Diarrhea (−)	31.7±5.9	29.8±5.4	35.4±7.2	32.7±6.8
Diarrhea (+)	43.8±11.1‡	40.1±10.8‡	39.0±8.0	41.3±10.2‡
Hb (g/dL)				
Diarrhea (−)	10.6±1.7	9.98±1.4	11.2±1.8	10.6±1.7
Diarrhea (+)	13.6±3.1‡	12.5±2.6‡	12.4±2.4*	12.9±2.8‡
MCV (fL)				
Diarrhea (−)	36.4±3.6	34.2±4.8	35.1±4.8	35.0±4.6
Diarrhea (+)	40.0±4.3†	37±7.2	35.1±3.4	37.6±5.4‡
MCHC (g/dL)				
Diarrhea (−)	33.8±2.0	33.7±3.2	32.0±2.9	32.9±3.0
Diarrhea (+)	30.6±2.3‡	31.8±3.6†	32.0±2.6	31.4±2.8‡
RDW (%)				
Diarrhea (−)	37.7±3.0	42.4±3.6	43.0±3.4	41.8±3.9
Diarrhea (+)	39.6±4.0	41.8±5.7	44.5±5.1	41.7±5.2

Data are presented as mean±SEM.
P-values were obtained using two-tailed independent t-tests ($^{*}P<0.05$, $^{\dagger}P<0.01$, and $^{\ddagger}P<0.001$).
RBC, red blood cells; Hb, hemoglobin; HCT, hematocrit; MCV, mean cell volume; MCH, mean cell hemoglobin; MCHC, mean cell hemoglobin concentration; RDW, red blood cell distribution width.

Table 3. Counts of WBCs in diarrhea negative/positive calves according to age

	1~10 days	11~20 days	21~30 days	1 month
WBC ($\times10^3$/μL)				
Diarrhea (−)	10.0±4.3	12.8±3.3	11.2±2.8	11.6±3.5
Diarrhea (+)	24.5±15.6‡	16.8±9.4*	20.5±11.7‡	21.2±13.2‡
Neutrophil ($\times10^3$/μL)				
Diarrhea (−)	6.3±3.2	7.1±3.0	5.1±1.8	6.1±2.7
Diarrhea (+)	14.5±7.5‡	8.9±3.7	10.2±8.1‡	11.6±7.3‡
Lymphocyte ($\times10^3$/μL)				
Diarrhea (−)	3.6±1.7	5.2±1.5	5.4±1.2	4.9±1.6
Diarrhea (+)	8.9±11.0†	7.1±8.1	8.1±5.7†	8.2±8.8‡
Monocyte ($\times10^3$/μL)				
Diarrhea (−)	0.1±0.2	0.3±0.4	0.5±0.5	0.3±0.4
Diarrhea (+)	0.9±1.4†	0.8±1.6	1.3±1.5	1.0±1.5‡
Eosinophil ($\times10^3$/μL)				
Diarrhea (−)	0.1±0.1	0.3±0.4	0.4±0.4	0.3±0.4
Diarrhea (+)	0.1±0.4	0±0	0.1±0.2‡	0.1±0.3‡
Basophil ($\times10^3$/μL)				
Diarrhea (−)	0±0.1	0±0.2	0.0±0.0	0±0.1
Diarrhea (+)	0.1±0.2	0±0	0.1±0.2†	0.1±0.2†

Data are presented as mean±SEM.
P-values were obtained using two-tailed independent t-tests ($^{*}P<0.05$, $^{\dagger}P<0.01$, and $^{\ddagger}P<0.001$).

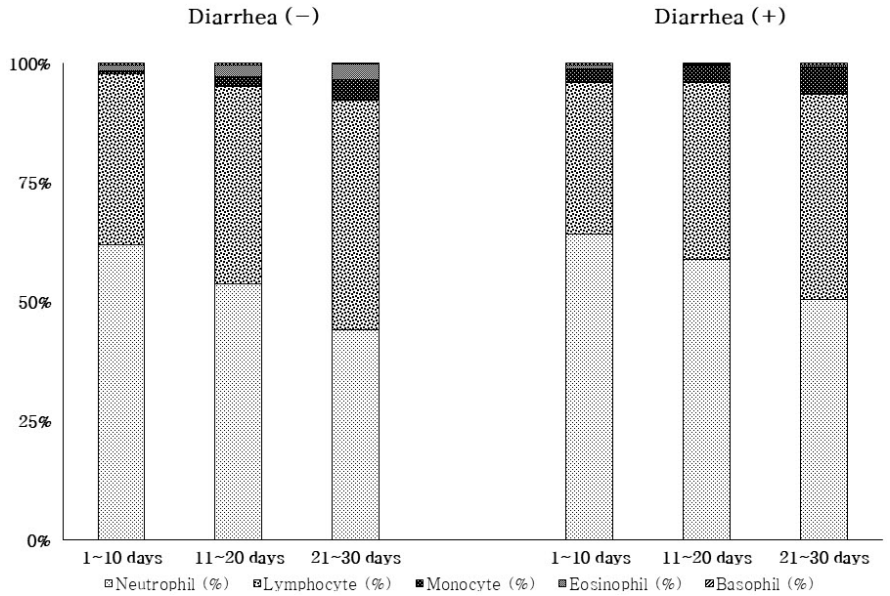

Fig. 1. The ratio of WBC composition (%).

group and 41.3 (±10.2) in positive group. This finding indicates that dehydration occurred due to diarrhea.

MCV (*fl*) was increased in positive groups compared with negative groups. The mean value of total was 35.0 (±4.6) in negative group and 37.6 (±5.4) in positive group respectively ($P<0.001$). And MCHC (g/dL) was decreased in positive groups, the average of total was 32.9 (±3.0) in negative group and 31.4 (±2.8) in positive group ($P<0.001$).

White blood cells profiles analysis

The White blood cells profiles of the calves in each group are listed in Table 3. Total WBC ($\times10^3/\mu L$)count was as follows respectively; 10.0 (±4.3) in 1～10 days old, 12.8 (±3.3) in 11～20 days old and 11.2 (±2.8) in 21～30 days old in negative group; 24.5 (±15.6) in 1～10 days old, 16.8 (±9.4) in 11～20 days old and 20.5 (±11.7) in 21～30 days old in positive group. WBC ($\times10^3/\mu L$)count of total was 11.6 (±3.4) in negative group and 21.2 (±13.2) in positive group ($P<0.001$). And neutrophil, lymphocyte and monocyte counts was increased in diarrhea positive group compared with negative group.

The ratio of neutrophil in both normal and diarrhea calves decreased gradually with age, but the value of diarrhea positive group was higher than that of the negative group (Fig. 1). The proportion of lymphocyte in both groups increased gradually with age, but lymphocyte level in normal calves was higher than that in diarrhea positive group. On the other hand, the level of monocyte increased with age in both groups, but the value in positive group was higher than that in negative group. Eosinophil and basophil levels were not significantly different between normal and diarrhea calves.

DISCUSSION

The purpose of this study was to investigate the change in blood markers with and without diarrhea in 179 Hanwoo calves under 30 days of age. As a result, CBC levels were different in normal and diarrhea calves.

It is well known that HCT increases significantly in animals with diarrhea (Feldman et al, 2000). Usually, an increase in HCT means concentration of blood (dehydration), and a decrease in HCT means anemia in diarrhea. Loss of plasma moisture due to calf dehydration and discharge diarrhea increases the value of HCT. Decreasing water in the plasma increases the viscosity of the plasma, and increasing the viscosity hinders the flow of blood. Therefore, when dehydration is severe, peripheral circulatory disorder occurs and peripheral body temperature decreases. In particular, the largest difference is found between 1 and 20 days of age. Statistical significance decreases between 21 and 30 days. Losing water at the same level can be attributed to relatively young animal with greater weight. MCV has a rather high calves with diarrhea and the difference gradually decreases with age.

WBC counts increased significantly in calves with diarrhea at all ages. It has been reported that neutrophils and monocytes tend to increase with age and in diarrhea calves, and the proportion of lymphocytes tends to decrease. The increase in the total number of WBCs is statistically significant at all ages. However, the change in WBC composition did not differ significantly with age.

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CONFLICT OF INTEREST

No potential conflict of interest relevant to this article was reported.

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REFERENCES

Acres SD, Saunders JR, Radostits OM. 1977. Acute undifferentiated neonatal diarrhea of beef calves: the prevalence of enterotoxigenic *E. coli*, reo-like (rota) virus and other enteropathogens in cow-calf herds. Can Vet J 18: 113-121.

Constable P. 2004. Antimicrobial Use in the Treatment of Calf Diarrhea. Journal of veterinary internal medicine. American College of Veterinary Internal Medicine 18: 8-17.

Done JT, Terlecki S, Richardson C, Harkness JW, Sands JJ, Patterson DS, Sweasey D, Shaw IG, Winkler CE, Duffell SJ. 1980. Bovine virus diarrhoea-mucosal disease virus: pathogenicity for the fetal calf following maternal infection. Vet Rec 106: 473-479.

Feldman BF, Zinkl JG, Jain NC. 2000. Schalm's veterinary hematology. Canadá: 140-142.

Grove-White DH, White DG. 1993. Diagnosis and treatment of metabolic acidosis in calves: a field study. Vet Rec 133: 499-501.

Hartmann H, Berchtold J, Hofmann W. 1997. Pathophysiological aspects of acidosis in diarrhoeic calves. Tierärztl Umsch 52: 568-574.

Hartmann H, Reder S. 1995. Effects of dehydration on functional parameters of fluid balance as well as effectiveness of rehydration using crystalline or colloidal infusion drips in calves. Tierärztliche Praxis 23: 342-350.

Kang SJ, Ryu SJ, Chae JS, Eo SK, Woo GJ, Lee JH. 2004. Occurrence and characteristics of enterohemorrhagic Escherichia coli O157 in calves associated with diarrhoea. Vet Microbiol 98: 323-328.

Kim U, Jung YH, Choe C, Kang SJ, Chang SS, Cho SR, Yang BC, Hur T. 2015. Korean native calf mortality: The causes of calf death in a large breeding farm over a 10-year period. Korean Journal of Veterinary Research 55: 75-80.

Kang MI, Chung YU, Chung DY, Lee CY, Lee CG, Wee SH, Cho JJ. 2001. Survey on Korean-native Calves Diseases and Mortality. Korean journal of veterinary service 24: 223-241.

Kwon OD, Lee SO, Jeong H, Lee JM. 2000. Epidemiological Investigation of Diseases in Korean Native Suckling Calves. Journal of Veterinary Clinics 17: 93-101.

Tzipori S, Campbell I, Sherwood D, Snodgrass DR, Whitelaw A. 1980. An outbreak of calf diarrhoea attributed to cryptosporidial infection. Vet Rec 107: 579-580.

Yimer M, Mamo G, Birhanu B, Dinaol B. 2015. A review on major bacterial causes of calf diarrhea and its diagnostic method. Journal of Veterinary Medicine and Animal Health 7: 173-185.