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Hemato-Biochemical Indices of Hanwoo Cattle Raised at Different Altitudes

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Abstract : The health status of three groups of Hanwoo steers (n = 157) aged 2-3 years, raised at three different altitudes (600 m, n = 50; 200-400 m, n = 58; plane land, n = 49) and environment with more or less similar management have been evaluated through hematological, biochemical and globulin examinations in order to determine the optimum environment suitable for raising cattle while at the same time minimizing the risk of disease. Five mL of blood samples from each animal were collected by jugular veinipuncture and 2 mL was transferred to a tube containing EDTA for complete blood count (CBC) and 3 mL in lithium heparin for chemistry screening (CS) and immunoassay. Among the CBC parameters a significantly higher white blood cell count (tWBC), total red blood cell count (tRBC), hemoglobin (Hb) and packed cell volume (PCV) were noticed in the high altitude groups, whereas those were lower in the plane land group. In the CS parameters higher levels of aspartate aminotransferase (AST), creatinine, lactate dehydrogenase (LDH) and total bilirubin (TBL) were found in the plane land group, whereas those were lower in the high altitude groups. The total protein (significantly) and globulins were higher in the 600 m group. The results of this study revealed that the overall health status of the Hanwoo cattle based on the hemato-biochemical indices was superior in the highest altitude and inferior in the plane land group but all the parameters were within the reference range in all the groups. Therefore, for recommendation of a suitable environment at an appropriate altitude for raising cattle there need to be further studied along with the hemato-biochemical parameters; considering, breeding, feeding, management, marketing, waste disposal and other factors.

Key words **:** high altitude, hemato-biochemical, Hanwoo cattle.

Introduction

The Korean economy has grown very fast since 1970, which has brought about a change in the Korean life-style. Since 1970, consumption of livestock products such as meat has increased by 4.6 times. Most Korean beef cattle are the traditional brown Korean Native Cattle, the Hanwoo (18). Therefore, defining a suitable environmental condition is of great importance for Hanwoo cattle. The significance of determining hematological and biochemical indices of domestic animals has been well documented (1,20,21,25). The differences of various haematobiochemical parameters of the Hanwoo cattle raised under native husbandry condition at different altitude have further underlined the need to establish appropriate physiological baseline values which could help in realistic evaluation of the management practice, nutrition and diagnosis of health condition.

The complete blood count (CBC) is an important and powerful diagnostic tool as a component of a minimum database. It can be used to monitor response to therapy, to gage the severity of an illness or as a starting point for formulating a list of differential diagnosis (4). It is well known that variables such as breed, stage of growth, age, reproduction status and stage of lactation have an influence on many blood parameters (7).

Hematological values such as total red blood cell count (tRBC) (14) packed cell volume (PCV) (8), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), hemoglobin concentration (Hb) (15) and white blood cells (9), i.e. lymphocyte and monocyte indicate adaptability to adverse environmental condition. As production is closely related to the health and nutrition status of all animals, hematological and plasma biochemical evaluation is considered to be basic aid in this determination (19). However, hematological and biochemical values are also used for indicating stress and welfare (3). Determination of normal values for hematological and biochemical parameters are important for the clinical interpretation of laboratory data. These indices may vary depending on factors such as sex, age, weather, stress, season and physical exercise (13). Hence, the hematological values during different physiological situations should be known for the diagno-

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sis of various pathological and metabolic disorders, which can adversely affect the productive and reproductive performance of cows, resulting in great economic losses to dairy farmers (22).

Raising cattle in a good environmental condition have advantages on production and reproduction due to minimal stress and less chance of infection. The ideal situation is to determine the optimum environment suitable for raising cattle while at the same time minimizing the risk of disease. Therefore, in this study, the health status of three groups of steers, raised at three different altitude and environment having more or less similar management have been evaluated through hematological, biochemical and globulin examinations.

Materials and Methods

Study Period

This study was conducted during the period 23 December 2008 to 02 February 2009 in 3 areas having different altitudes in Imsil, Jeollabukdo Province, Republic of Korea.

Selection of farms

Imsil area is potential for cattle farming because of its suitable weather, feeds and fodder availability, and available veterinary facilities from the local Veterinary Hospital as well as from The College of Veterinary Medicine, Chonbuk National University (CBNU). The cattle farms were selected for conducting the study because of its suitable location, large population, satisfactory record keeping system, more or less similar feeding and management, and also for the kind cooperation from the farm administration.

Selection of experimental animals

One hundred fifty seven healthy fattening Hanwoo steers of 2-3 years of age, raised in three different altitudes were selected for this study. All the steers physically examined and the clinical history from the farm record was reviewed. Fifty steers were selected from the 3 farms situated at an altitude of more than 600 meters (m) whereas 58 from 4 farms at an altitude between 200 to 400m, and 49 steers from 3 farms at the plane land in Imsil.

General management system of selected farms

In all the selected farms the steers are housed in multiple pens with grilled pavement containing 4 to 5 animals in each. There are different sheds for different age population. In the sheds all the animals get sufficient airspace and there is proper drainage facility and quick disposal of animal waste.

Both roughage (straw, 1% of the bodyweight) and commercial concentrates (Chongche Bori®, 2% of the bodyweight) are offered to the animals twice a day and they had access to the water *ad libitum*.

There is a least prevalence of infectious diseases in the farms. When any sign of sickness arises the farmers call a veterinarian from the Local Animal Hospital or communicate with the Professors of the clinical subjects, College of Veterinary Medicine, CBNU. All the animals are regularly immunized and administered anthelmentics.

Collection, preservation and analysis of blood samples

Five mL of blood samples from each animal were collected by jugular veinipuncture and immediately 2 mL of the samples were transferred to sterile screw-capped tubes containing EDTA for hematology and other 3 mL in lithium heparin for biochemistry. Blood samples were transported to the laboratory within one hour keeping in a thermoflask with ice and then fresh blood was examined. The heparinized samples were centrifuged at 1500 g for 15 minutes, the supernatant plasma was collected, divided into two halves and transferred to Eppendorf tubes and stored at −80°C until used for chemistry screening (CS) and immunoglobulin assay.

The blood samples were subjected complete blood count (CBC) which includes total white blood cell count (tWBC), total red blood cell count (tRBC), hemoglobin (Hb), packed cell volume (PCV), platelet count, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC); and CS which includes alkaline phosphatase (AP), aspartate aminotransferase (AST), blood urea nitrogen (BUN), creatinine, glucose, lactate dehydrogenase (LDH), total bilirubin (TBL), total protein, albumin, albumin/globulin (A/G) ratio and to immunoassay for various fractions of globulins (alpha, beta and gamma).

The CBC was analyzed using an automatic hematology analyzer (Scil Vet abc®, Scil Animal Care Company, USA). Briefly, the EDTA-blood sample tubes were placed on a roller mixer and rolled at 33 rpm and 16 mm amplitude (rise and fall) for proper mixing. The sample identity was entered in the automatic hematology analyzer and once the sample needle was down it was put into the blood samples until it touched the bottom of the tube and then the tube was slightly back off the needle. Once the start button was pressed, 20 μL of the blood sample was automatically taken in by the sample needle. The results were available for read by 90 seconds on the LCD and were received as printed form as well.

For CS analysis 90 μL plasma was drawn from the Eppendorf tubes using the automatic pipettor/dilutor. The plasma was then dispensed into a test rotor (VET-16 Veterinary Test Rotor®, Hemagen Diagnostics, Inc., USA) and the rotor cap was snapped into place. The test rotor was loaded in the automatic chemistry analyzer (Hemagen Analyst®, Hemagen Diagnostics, Inc., USA) and a complete result was received in a printed form in about 10 minutes. For immunoassay the plasma samples were sent to the Samkwang Medical Laboratory, Seoul, Korea. The immunoassay for various fractions of globulins (alpha, beta and gamma) was done by a specialist of the laboratory using specific kits for the assay.

Statistical analysis:

The data obtained from the present study was analyzed using analyses of variance (ANOVA) technique and means were sep-

arated by Ducan's multiple range test. The $P < 0.05$ or less was considered as significant. The data is expressed as as mean ± standard deviation (SD).

Results

The complete blood count (CBC) values

In this study the CBC values of all the groups were found within the normal reference range (12). A significantly ($P < 0.05$) higher tWBC was found in the 600 m ($8.01 \pm 1.90 \times 10^3$/mL) and 200-400 m groups ($7.80 \pm 2.77 \times 10^3$/mL) in comparison to that of the plane land group ($6.53 \pm 3.41 \times 10^3$/mL). The highest tRBC value was observed in the 600 m group ($9.36 \pm 1.43 \times 10^3$/mL) in comparison to that of the 200-400 m ($9.06 \pm 1.198 \times 10^3$/mL) and plane land group ($7.98 \pm 1.48 \times 10^3$/mL). The differences of the tRBC values in both high altitude groups were significant ($P < 0.05$) as compared with the plane land group. The Hb and PCV values correspond to the tWBC and tRBC values, which were also found significantly higher ($P < 0.05$) in the 600 m (Hb $11.42 \pm 0.83 \times 10^3$/mL, PCV $42.19 \pm 3.98 \times 10^3$/mL) and 200-400 m groups (Hb $10.88 \pm 0.82 \times 10^3$/mL, PCV $38.67 \pm 7.92 \times 10^3$/mL) in comparison to that of the plane land group (Hb $8.50 \pm 1.37 \times 10^3$/mL, PCV $36.90 \pm 6.34 \times 10^3$/mL). However, the Hb and PCV values were highest in the 600 m group. A significantly higher platelet count ($P < 0.05$) was observed in the 600 m as compared with that of the 400 m and plane land groups. No significant differences were observed in the values of MCV, MCH and MCHC between the groups. The CBC values in the Hanwoo cattle (n = 157) raised in different altitudes in the Imsil is presented in the Table 1.

The immunoassay and chemistry screening (CS) values

The values of the AP, AST, BUN, creatitine, glucose and TBL were within the reference range in all the groups (11). The higher levels of AST and TBL were found in the plane land group (AST 36.50 ± 0.70 U/L, TBL 1.11 ± 0.05 mg/dL) in comparison to those of the 200- 400 m (AST 14.20 ± 4.92 U/L, TBL 0.26 ± 0.10 mg/dL) and 600 m groups (AST 14.00 ± 5.66 U/L, TBL 0.25 ± 0.08 mg/dL), but this differences were not statistically significant. A relatively higher level of creatinine was observed in the plane land group (1.36 ± 0.07 mg/dL) in comparison to that of the 200- 400 m (0.61 ± 0.12 mg/dL) and 600 m groups (0.66 ± 0.06 mg/dL). However, the differences in creatinine level between the groups were not statictically significant. A significantly higher ($P < 0.05$) level of LDH was observed in the plane land group (871.00 ± 72.12 U/L) in comparison to that of the 200- 400 m (241.40 ± 32.97 U/L) and 600 m groups (240.40 ± 34.63 U/L).

A significantly highest ($P < 0.05$) total protein value was found in the 600 m group (7.18 ± 0.40 gm/dL) in comparison to that of the 200-400 m (6.70 ± 0.37 gm/dL) and plane land group (6.90 ± 0.67 gm/dL). A relatively higher globulin values were observed in 600 m (53.47 ± 2.53) and plane land group (51.55 ± 2.75) in comparison to that of the 200-400 m group (46.77 ± 2.29). However, these differences were not statistically significant. Among the various fractions of globulins, the level of γ- globulin was comparatively higher in 600 m and plane land groups that the 200-400 m group, but the differences were not significant. The immunoassay and CS values of the Hanwoo cattle (n = 157) raised in different altitudes are presented in Table 2 and 3.

Discussion

The importance of hematological and biochemical indices in animal husbandry is well acknowledged (2). Metabolic disturbance usually by inappropriate feeding without manifestation of clinical symptoms are important in animal husbandry and may cause insufficiently developed breeding cattle (23). The physiological equilibrium of the body is maintained mainly by the blood, but many physiological conditions may alter this equilibrium. The changes in hematological constituents are important

Table 1. The complete blood count (CBC) values in the Hanwoo cattle (n = 157) raised in different altitudes

Parameters	Plane land (n = 49) (Mean ± SD)	200 t0 400 m altitude (n = 58) (Mean ± SD)	600 m altitude (n = 50) (Mean ± SD)	Reference range (Mean ± SD)
tWBC 10^3/mL	6.53 ± 3.41	7.80 ± 2.77*	8.01 ± 1.90*	4.00-12.00
tRBC 10^6/mL	7.98 ± 1.48	9.06 ± 1.198*	9.36 ± 1.43*	5.00-10.00
Hb g/dL	8.50 ± 1.37	10.88 ± 0.82*	11.42 ± 0.83*	8.00-15.00
PCV %	36.90 ± 6.34	38.67 ± 7.92*	42.19 ± 3.98*	24.00-46.00
Platelet 10^3/mL	626.81 ± 211.25	454.10 ± 211.95	672.42 ± 270.21*	200-800
MCV fL	46.42 ± 3.39	42.68 ± 4.71	45.07 ± 5.01	40.00-60.00
MCH pg	10.06 ± 0.10	12.07 ± 0.12	12.20 ± 0.14	11.00-17.00
MCHC g/dL	23.03 ± 1.37	28.13 ± 0.91	27.06 ± 1.30	28.20-38.00

*Statistically significant $p < 0.05$ or less, SD- Standard deviation, tWBC- Total white blood cell count, tRBC- Total red blood cell count, Hb- Hemoglobin, PCV- Packed cell volume, MCV- Mean corpuscular volume, MCH- Mean corpuscular hemoglobin, MCHC- Mean corpuscular hemoglobin concentration.

Table 2. The blood chemistry screening (CS) values in the Hanwoo cattle (n = 157) raised in different altitudes

Parameters	Plane land (n = 49) (Mean ± SD)	200 t0 400 m altitude (n = 58) (Mean ± SD)	600 m altitude (n = 50) (Mean ± SD)	Reference range (Mean ± SD)
AP U/L	219.50 ± 85.55	153.00 ± 24.54	220.79 ± 220.08	66.00-220.00
AST U/L	36.50 ± 0.70	14.20 ± 4.92	14.00 ± 5.66	7.00-38.00
BUN mg/dL	19.25 ± 1.34	21.02 ± 5.25	21.04 ± 4.55	6.00-23.00
Creatinine mg/dL	1.36 ± 0.07	0.61 ± 0.12	0.66 ± 0.06	0.40-1.40
Glucose mg/dL	79.50 ± 3.53	123.60 ± 14.57	124.00 ± 17.20	75.00-128.00
LDH IU/L	871.00 ± 72.12*	241.40 ± 32.97	240.40 ± 34.63	105.00-260.00
TBL mg/dL	1.11 ± 0.05	0.26 ± 0.10	0.25 ± 0.08	0.20-1.40

*Statistically significant $p < 0.05$ or less, SD- Standard deviation, AP- Alkaline phosphatase, AST- Aspartate aminotransferase, BUN- Blood urea nitrogen, LDH- lactate dehydrogenase, TBL- Total bilirubin.

Table 3. The concentration of albumin and various fractions of globulin in the Hanwoo cattle (n = 157) raised in different altitudes

Parameters	Plane land (n = 49) (Mean ± SD)	200 t0 400 m altitude (n = 58) (Mean ± SD)	600 m altitude (n = 50) (Mean ± SD)
Total Protein g/dL	6.90 ± 0.67	6.70 ± 0.37	7.18 ± 0.40*
Albumin/ Globulin	0.94 ± 0.13	1.14 ± 0.17	0.87 ± 0.10
Albumin %	48.45 ± 3.60	53.22 ± 3.87	46.52 ± 3.15
Globulin %	51.55 ± 2.75	46.77 ± 2.29	53.47 ± 2.53
α- Globulin %	10.2 ± 1.27	11.07 ± 1.24	12.00 ± 1.78
β- Globulin %	11.55 ± 1.06	12.80 ± 1.61	11.07 ± 1.85
γ- Globulin %	29.8 ± 5.93	22.90 ± 4.02	30.40 ± 3.98

*Statistically significant $p < 0.05$ or less, SD- Standard deviation.

indicators of the physiological or pathological state of the animal. Blood examination is also performed for screening procedure to assess general health of the animals. (9,12).

In this study, a significantly higher tWBC, tRBC, Hb and PCV were noticed in the high altitude groups, whereas those were lower in the plane land group. This finding is in agreement with the previous reports (6,10,16). In high altitude, the animals are subjected to relatively lower oxygen saturation in the atmosphere (hypoxia). This physiological hypoxia stimulates hematopoiesis, increase production of Hb and accelerates pulse and respirations rates (5,12,16). In this study, a significantly higher platelet count observed in the 600 m group as compared with that of the 400 m and plane land groups correlates with the reports of Leon-Velarde *et al.*, 2000 (16) and Hyun *et al.*, 2007 (10), who also reported higher level of platelets in the cattle raised at high altitude. In the high altitude, the prolong hypoxia stimulates the erythroid and myeloid activity in the bone marrow which leads to increasing the number of cellular elements of the blood including RBC, WBC and platelets. Therefore the highest tWBC, tRBC, Hb, PCV and platelets noticed in the 600 m group resulted from the physiological compensation by the bone marrow in response to the persistent hypoxic condition at the high altitude.

The values of the AP, AST, BUN, glucose and TBL were within the reference range in all the groups (11,17). The AST and TBL levels are associated with hepatic cellular damage, muscular injury and RBC destruction. However, higher levels of AST and TBL were found in the plane land group in comparison to those of the 200-400 m and 600 m groups. This result revealed that more animals at the ground level have cellular damages by unidentified causes suggesting more stressful environment in lower altitude which in agreement with the earlier report (10).

A significantly higher level of LDH was observed in the plane land group in comparison to those of the 200-400 m and 600 m groups. Lactate dehydrogenase (LDH) catalyzes the conversion of lactate to pyruvate and is present in high concentrations in the cytoplasm of almost all cells. Since LDH is found in every tissue in the body, the diagnostic value of an elevated level without additional laboratory investigation is limited. Increased levels are found in myocardial infarction, hepatic necrosis, skeletal muscle disease, renal disease, pulmonary infarction, malignancy, megaloblastic anemia, hemolysis and numerous other disease processes involving tissue damage increase serum levels of LDH. In liver disease, AST and ALT are usually much more elevated than LDH. Haemolysis during collection, delays in pro-

cessing, or refrigeration of unseparated blood may cause an artefactual increase in LDH activity. Therefore, the higher level of LDH in the ground level group might be due to some nonspecific stress rather than pathologic condition s.

The total protein values were significantly highest in the 600 m group in comparison to that of the 200-400 m and plane land group. A relatively higher globulin values were observed in 600 m and plane land group in comparison to that of the 200-400 m group. However, there were no significant differences among the various fractions of globulins between the groups. This finding cannot be compared and contrasted because of the lack of similar reports. The total plasma protein mirrors the health and nutritional status, whereas the globulin reflects the immune status of the animals.

The results of this study in generally revealed that the cattle raised at high altitude have better health statutes. This might be due to hypoxic stimulation of the cardiopulmonary function and better oxygen utilization at the high altitude. The relatively colder environment there also minimizes the risks of infectious diseases mediated by insect vectors and stress from the environmental pollution. On the contrary, cattle raised at high altitude are prone to high altitude disease which is reported to be severe at an altitude over 1500 m (24).

Though the hemato-biochemical profile was relatively better in the high altitude group, no parameters were beyond the reference range in the plane group also. During this study, it was observed that there are several advantages of raising cattle in the plane land over that in the high altitude in terms of the herd management, convenient for the personnel to attend the herd, easy to carry foods and other tools to the farm even in the adverse climates, easy to dispose the waste and carcass, the manure can be easily used to the nearby crop field as organic fertilizer and so on. On the other hand, the limitation is that it is not known to us what would be the health status of the calves bought from the local market and attempted to raise at high altitude for fattening. The high altitude may aid to the health of the calves born and raised there but may be inconvenient to a newly introduced herd from the plane land. Considering the above things raising cattle in the plane land may be convenient than that in the high altitudes.

The results of this study revealed that the overall health status of the Hanwoo cattle based on the hemato-biochemical indices was superior in the highest altitude and inferior in the plane land group but all the parameters were within the reference range. Therefore, for recommendation of a suitable environment at an appropriate altitude for raising cattle require further study along with the hemato-biochemical parameters; considering, breeding, feeding, management, marketing, waste disposal, cost effectiveness etc.

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서로 다른 고도에서 사육된 비육한우의 혈액-혈청 지수

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요 약 : 서로 다른 고도에서 사육하고 있는 2-3세령의 수컷, 건강한 비육 한우를 세 그룹(600 m, n=5 0; 200-400 m, n=58; 0 m, n=49)으로 나누어 혈액 및 혈청검사 그리고 면역항체가 검사를 실시 하였다. 고도를 제외한 일반 사육환경과 식이 및 축사의 형태는 매우 비슷한 조건이었으며 질병이 확인 된 개체는 통계에서 제외 하였다. 경정맥에서 채취한 혈액은 일반혈액검사 중 WBC, RBC, Hb, PCV 를, 혈청화학검사 중 AST, LDH, TBL를 집중적으로 비교 분석하였으며 또한 면역학적 검사를 실시하였다. 고도가 높은 그룹이 낮은 그룹에 비하여 CBC에서는 조금 높게 나타났으며, 혈청검사에서는 평지에서 사육된 한우가 조금 높게 나왔다. 그러나 모두가 정상범위를 벗어나거나 통계적으로 유의한 수준의 차이를 보이지 않았다. 이러한 결과는 지금까지 알려진 대부분의 연구결과 와는 차이가 있는 것이다. 본 연구와 지금까지의 알려진 연구와의 이러한 차이는 과거의 연구들은 고도의 차이가 현저한 경우가 대부분이었으나 본 연구에서 조사한 지역은 서로 고도가 비슷한 점에서 생긴 결과라고 여겨지며, 특히 고도가 600 m 이하인 지형에서는 서로 큰 영향을 받지 않는 것으로 생각된다. 따라서 한우의 경우 목장 시설의 건축시 지형에 따른 어려움과 주변에서 들여오는 조사료의 수송에 따른 경제적 불리, 축산폐수 처리는 물론 퇴비의 활용 등 전반적인 목장 관리 면에서 고도가 높은 산지 보다는 평지가 권장된다.

주요어 : 고도, 혈액-혈청검사, 비육한우, 목장건축