NOTE *Internal Medicine*

Blood biochemical values in Japanese Black breeding cows in Kagoshima Prefecture, Japan

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ABSTRACT. To obtain blood biochemical basic data of Japanese Black breeding cows in Kagoshima Prefecture, Japan, blood samples were obtained from 857 clinically healthy multiparous cows on 71 farms. Cows were divided into three stages: the prepartum stage (between 20 and 80 days before parturition, n=290), lactation stage (between 20 and 80 days after parturition, n=283) and maintenance stage (between 120 and 200 days after parturition, n=284). The mean concentration of total protein and total cholesterol, and the mean activities of aspartate aminotransferase in the lactation stage tended to be higher than those in the prepartum and maintenance stages. The mean concentration of glucose in the prepartum stage tended to be lower than that in the lactation and the maintenance stages. The mean concentration of nonesterified fatty acids and beta-hydroxybutyrate in the prepartum stage tended to be higher than those in the lactation and maintenance stages. The mean concentration of serum retinol was approximately 30 μg/d*l* in all stages. These results suggest that the blood biochemical values of multiparous Japanese Black cows vary with breeding stages, and it is considered that the blood parameters obtained in this study are useful as indices for health management of Japanese Black breeding cows.

KEY WORDS: blood biochemical value, breeding stage, Japanese Black cow, retinol

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The Japanese Black is a breed of beef cattle that originated in and is mainly distributed in Japan [8]. An animal's health can be defined as the absence of diseases determined by clinical examinations combined with various diagnostic tests. Serum biochemical reference values are used to establish normality and to diagnose diseases and physiological alterations [3, 4, 7, 9, 16, 17]. Although many reference values have been established for dairy cows [3, 4, 17], there have been few published references for beef cows [9, 16]. In particular, the reports for Japanese Black cows have been very scarce [20]. It is well known that variables, such as reproductive, as well as lactation status have an influence on many blood parameters [3, 4, 9, 16, 17]. To the best of the authors' knowledge, reference values have not been established for particular stages of Japanese Black breeding cows. Therefore, the current study was conducted to establish reference values of serum biochemical parameters in those particular stages of Japanese Black breeding cows kept on ordinary farms in Kagoshima Prefecture, Japan.

Privately owned Japanese Black breeding herds (71 farms) in Kagoshima Prefecture, Japan, were enrolled in this study. The number of cows in each herd ranged from 30 to less than 300. Eight hundred fifty-seven multiparous Japanese Black breeding cows (3 to 9 years old) on these farms were used, and blood samples were collected once per head by the authors between April 2011 and May 2014. All cows were clinically healthy, housed indoors and stayed with their calves generally for three months. All cows were fed grasses (grown at the farm or purchased), such as rice straws, Italian ryegrass, oats or Rhodes grass, in addition to supplemental concentrate purchased from several feed companies. Although the contents and amounts of grasses and supplemental concentrate purchased were different for each farm, the feed fundamentally met the requirements of the Japanese Feeding Standard for Beef Cattle [2], and cows in the lactation and prepartum stages were fed more than cows in the maintenance stage. About 12 cows on each farm were sampled at random during the prepartum stage (between 20 and 80 days prior to parturition without lactation, n=290), lactation stage (between 20 and 80 days after parturition, without pregnancy, with lactation, n=283) and maintenance stage (non-lactation, between 120 and 200 days after parturition with pregnancy, n=284). These periods were chosen based on the study by Watanabe *et al.* [20]. Animals were cared for according to the Guide for the Care and Use of Laboratory Animals of the Joint Factory of the Veterinary Medicine, Kagoshima University.

Blood samples were collected from the jugular vein into plain vacuum tubes between 10 a.m. and noon. Serum was separated within 60 min after blood collection and stored at −20°C until analysis. The following biochemical parameters were determined using a Labospect 7180 autoanalyzer (Hitachi High-Technologies Corporation, Tokyo, Japan): total protein (TP), albumin (Alb), globulin (Glb), Alb/Glb (A/G) ratio, urea nitrogen (UN), glucose (Glu), total cholesterol (T-Cho), aspartate aminotransferase (AST), γ-glutamyltransferase (GGT), nonesterified fatty acids (NEFA) and beta-hydroxybutyrate (βHB). Serum retinol

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Table1. Serum biochemical values of Japanese Black cows in different breeding stages

	Prepartum stage (n=290)			Lactation stage (n=283)			Maintenance stage (n=284)		
	Mean	SD	Normal range[a)]	Mean	SD	Normal range	Mean	SD	Normal range
TP (g/d*l*)	7.04	0.47	6.11–7.98	7.20	0.49	6.21–8.19	7.12	0.48	6.17–8.08
Albumin (g/d*l*)	3.22	0.26	2.70–3.74	3.27	0.30	2.67–3.88	3.22	0.30	2.62–3.81
Globulin (g/d*l*)	3.82	0.47	2.87–4.77	3.91	0.51	2.89–4.94	3.89	0.50	2.90–4.89
A/G	0.86	0.15	0.56–1.15	0.85	0.16	0.53–1.17	0.84	0.15	0.53–1.14
UN (mg/d*l*)	9.33	3.51	2.32–16.34	7.92	3.30	1.32–14.51	8.57	3.01	2.56–14.59
T-Cho (mg/d*l*)	112.8	23.1	66.6–159.1	125.5	30.1	65.3–185.6	102.9	22.8	57.4–148.5
Glucose (mg/d*l*)	51.9	5.0	41.9–62.0	54.2	6.7	40.9–67.5	54.3	6.6	41.1–67.5
AST (IU/*l*)	54.6	9.4	35.9–73.4	67.8	13.5	40.8–94.8	58.4	12.3	33.9–83.0
GGT (IU/*l*)	14.9	4.2	6.4–23.4	17.6	4.7	8.3–26.9	17.1	5.4	6.3–27.8
NEFA (μEq/*l*)[b)]	187.0		60.1–581.8	123.9		36.2–424.0	96.2		36.5–253.6
βHB (μmol/*l*)[b)]	419.4		215.4–816.7	342.3		196.0–597.6	285.4		152.3–534.9
Retinol (μg/d*l*)	31.8	6.2	19.3–44.3	29.9	6.3	17.3–42.5	29.6	6.8	16.1–43.1

a) Normal range=mean ± 2SD. b) A log10 transformation was applied to NEFA and βHB. TP=total protein, A/G=albumin/globulin ratio, UN=urea nitrogen, T-Cho=total cholesterol, AST=aspartate aminotransferase, GGT=γ-glutamyltransferase, NEFA=nonesterified fatty acids, βHB=beta-hydroxybutyrate.

level was also analyzed using high performance liquid chromatography (JASCO, Tokyo, Japan) as previously reported [1]. The results obtained for each stage were expressed as the mean ± 2SD. As distributions of NEFA and βHB were skewed to the left, statistical analysis was conducted using logarithmic transformation. The values less than the mean −3SD and values more than the mean + 3SD were regarded as outliers. This method was determined based on a study by Kida [14].

The results of serum biochemical analysis for the three stages are shown in Table 1. The mean concentration of TP and T-Cho and the mean activities of AST and GGT in the lactation stage tended to be higher than in the prepartum and maintenance stages. The mean concentration of Glu in the prepartum stage tended to be lower than in the lactation and maintenance stages. The mean concentrations of NEFA and βHB in the prepartum stage tended to be higher than those in the lactation and maintenance stages. The mean concentration of retinol was approximately 30 μg/d*l* in all stages.

The main objective of this research was to investigate and clarify the blood parameters that might differ with breeding stages in Japanese Black cows in Kagoshima region.

The serum TP value is usually used as an indicator of the nutritive status of animals, as it reflects feed intake and metabolism [10]. In the present study, the TP value in the lactation stage tended to be higher than those of the other stages. Therefore, increased protein in blood might have reflected efficient and increased protein utilization with lactation. This agrees with reports from other investigations [4, 16].

The serum T-Cho value is influenced by energy intake. Thus, it is a useful indicator of feed intake [11]. In previous studies, increased cholesterol concentrations were found in cows during lactation [3, 4, 11]. In the present study, the T-Cho value in the lactation stage tended to be higher than those of the other stages. Therefore, the T-Cho concentration in the lactation stage might be associated with improved feed intake.

Serum UN has been reported to be a sensitive indicator of the available digestible crude protein, which can facilitate the efficiency of protein utilization [10, 12]. A large portion of most dietary protein is hydrolyzed and deaminated by rumen microflora, producing peptides and free ammonium in the rumen [10]. The amount of ammonium converted to urea in the body reflects the total amount of degraded protein and the rate of ammonium incorporated into microbial protein [10]. Microbial growth rate, in turn, is substantially affected by the availability of energy for the rumen microbes [10]. The serum Alb value has been reported to be a long-term indicator of available digestible crude protein status, because of its long half-life [10, 15]. In the present study, the UN value in the lactation stage tended to be lower than other stages. Alb value in the lactation stage tended to be higher than other stages. Additionally, the T-Cho value, which is influenced by energy intake [10], in the lactation stage was the highest among all stages. Therefore, the efficiency of protein utilization by rumen microbes in the lactation stage might have improved, and this might have caused the UN concentration to decrease.

Serum AST and GGT represent liver-associated enzymes that leak into the bloodstream following liver damage [17, 18]. In the present study, the AST and GGT activities in the lactation stage tended to be higher than those of the other stages. These results might be due to higher hepatic function associated with milk production. These results agree with reports from other investigations [3, 4, 17].

Serum NEFA is the metabolite directly associated with energy balance [10]. NEFA is elevated during short-term insufficiency of energy, which is likely the result of increased lipolysis [10]. The βHB is the ketone body of choice for routine measurement, because of its stability in serum [10]. βHB concentration is less constrained physiologically and more likely to reflect nutritional status compared with blood glucose [10]. Although, milk production by dairy cattle is much higher compared with Japanese Black cattle [19], the NEFA and βHB values of dairy cows in the prepartum stage were lower than those in the lactation stage [4, 14]. In the present study, the NEFA and βHB values in the prepartum stage tended to be higher than those in the lactation stage. The higher NEFA and βHB in the prepartum stage might reflect that cows in this stage had lower feed intake capacity and higher energy requirements for their fetuses.

Retinol is known to have many functions, including maintenance of epithelial cells, vision, gene regulation, immune cell function and breeding [5, 6]. The adequate serum retinol concentration range was reported to be from 25 to 60 μg/d*l* [6]. A previous study demonstrated that the mean serum concentration of retinol in Japanese Black breeding cows was 21.3 μg/d*l*, which was below the adequate range [13]. On the other hand, in the present study, the retinol value was approximately 30 μg/d*l*, which was within the adequate range [6]. Therefore, cows in all stages seemed to be fed sufficient vitamin A.

Although the data in the present study were obtained only from ordinary farm in one prefecture of Japan and all herds in the lactation and prepartum stages were fed more than cows in the maintenance stage, with the amount and diet components varying in each herd and stage, these results suggest that blood biochemical values of multiparous Japanese Black cows vary with the breeding stage, and it is considered that the blood parameters obtained in this study are useful as indices for health management of Japanese Black breeding cows.

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