doi: 10.17221/40/2017-VETMED

Effect of colostrum composition on passive calf immunity in primiparous and multiparous dairy cows

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ABSTRACT: The aim of this study was to determine the effect of colostrum quality and composition on passive calf immunity in primiparous and multiparous dairy cows. Twenty-four primiparous and 24 multiparous dairy cows were used in this study. Calves born from primiparous dairy cows comprised the first group and calves born from multiparous dairy cows constituted the second group. After birth, colostrum samples were immediately taken from dairy cows. Venous blood samples were collected from the calves before the first colostrum intake and on the 2nd, 7th, 14th and 28th days after the first colostrum intake. Blood and colostrum samples were analysed for biochemical parameters, immunoglobulin and mineral levels. Fat and crude protein levels in colostrum were determined using the Gerber and Kjeldahl methods, respectively. Immunoglobulin levels in the colostrum of multiparous cows were significantly ($P < 0.05$) higher compared to primiparous cows while fat ratio, LDH activity, Ca, Mg, P and K levels were lower. There was a positive correlation among colostrum immunoglobulin, gamma-glutamyltransferase, crude protein and total protein. Serum immunoglobulin, total protein, globulin and gamma-glutamyltransferase activity in all calves were increased following the colostrum feeding. However, the serum immunoglobulin, total protein, globulin and gamma-glutamyltransferase levels in the second group of calves were higher than those of the first group of calves. There was a positive correlation among serum immunoglobulin, gamma-glutamyltransferase, globulin and total protein. Fe concentrations in all calves decreased over the course of 14 days and were lower in the second group of calves compared to the first group. In conclusion, the results of this study show that the colostrum quality of multiparous cows was better than that of primiparous cows. Colostrum crude protein, total protein, gamma-glutamyltransferase along with colostrum immunoglobulin might be used to determine colostrum quality. Serum immunoglobulin, total protein, globulin and gamma-glutamyltransferase activities could be used to determine the passive transfer status of calves.

Keywords: calves; immunoglobulin G; mineral concentrations

The first secretion from the mammary glands after birth is called the colostrum, and it incorporates many important factors and components (Cortese 2009). The colostrum contains not only nutrients such as protein, carbohydrate, fat, vitamins and minerals but also biologically active molecules required for specific functions. The most important bioactive components in the colostrum are antimicrobial and growth factors (Pakkanen and Aalto 1997). Other important components are cytokines and hormones. Immunoglobulins are the most important antimicrobial factors in the colostrum (Godden 2008). Colostral immunoglobulin G (IgG) has dual protective role in newborns. IgGs ab-

Supported by the Scientific Research Projects Office of Selcuk University, Turkey (Project No. 12102014; Doctoral thesis).

doi: 10.17221/40/2017-VETMED

sorbed from the small intestines into the circulation provide protection against septicaemia and other immunological alterations, whereas unabsorbed IgGs have a local protective effect on the intestines (Logan and Pearson 1978; Fecteau et al. 2009).

The quality of the colostrum is related to the level of immunoglobulin it contains (Godden 2008). A high-quality colostrum should have an IgG concentration of 50 g/l. Colostrum quality can vary depending on factors such as cattle age and race and colostrum volume, as well as on the application of certain agents by producers (Godden 2008; Gulliksen et al. 2008).

Passive immune transfer can be determined by measuring the serum IgG concentration at 24 and 48 hours after colostrum administration in newborn calves (Smith and Foster 2007; Godden 2008). Calves with a blood IgG level of less than 10 g/l at 24 and 48 hours after birth are considered to have failed in acquiring passive transfer (Filteau et al. 2003; Godden 2008; Cakiroglu et al. 2010). In some studies (Wittum and Perino 1995; Gungor et al. 2004) serum IgG concentration has been classified as adequate passive transfer (APT) (> 16 g/l), partial failure of passive transfer (PFPT) (8 to 16 g/l) or failure of passive transfer (FPT) (< 8 g/l). Failure of passive transfer is not a disease, but it is a condition that predisposes to disease development (Weaver et al. 2000). Different methods are used in determining FPT in calves. These include radial immunodiffusion (RID) and enzyme-linked immunosorbent assay (ELISA) tests that directly determine the serum-plasma IgG concentration and the sodium sulfite turbidity test, zinc sulfate turbidity test, glutaraldehyde coagulation test, serum total protein concentration and serum gamma-glutamyltransferase (GGT) activity, which indirectly estimate the IgG concentration (Godden 2008). The hypothesis of this study was that the colostrum composition and quality of primiparous and multiparous dairy cows would be different and would manifest as adverse effects on passive immunity, biochemical indicators and trace mineral levels in the calves of primiparous cows. The first aim of this study was to determine the effects of colostrum quality and composition on passive calf immunity in primiparous and multiparous dairy cows. The second aim of this study was to determine trace mineral and total protein (TP) content in colostrum and the relationship between colostrum crude protein and TP.

MATERIAL AND METHODS

The study protocol was approved by the institutional ethics committee. Twenty-four primiparous, 24 multiparous dairy cows (Holstein) and 48 healthy calves born to these dams were used as animal material. Calves born normally, standing up within 60 minutes after birth and showing the suction reflex were used. Calves from primiparous dairy cows constituted the first group (Group 1, n = 24) while calves from multiparous dairy cows comprised the second group (Group 2, n = 24). Colostrum that made up 10% of their body weight (min: 35 kg, max: 42 kg) was given to the calves included in this study using a feeding bottle in the 24 hours following their birth. Half of the colostrum to be given (5% of body weight) was given in the first six hours, while the other half was given within the 6–24-hour period following birth. Calves were taken into individual huts after birth and were fed two meals of milk that made up approximately 10–12% of their body weights. Calves were provided with water *ad libitum* and were given calf starter feed and alfalfa hay after the seventh day. Health and the general condition of calves, the presence of disease and the administration of medical treatment were recorded by following the animals over a period of one month.

Collection of blood samples. Venous blood samples (8 ml) were collected in clot activator vacuum tubes (BD serum tube, United States) five times from the *vena jugularis*, before the delivery of colostrum (day 0) and after the colostrum was fed on the 2nd, 7th, 14th and 28th days. Blood samples were stored at room temperature for 30 min and allowed to clot, then centrifuged at 5000 *g* for 10 min at + 4 °C using a refrigerated centrifuge. Serum was then removed and stored at –20 °C pending analysis.

Collection of colostrum samples. After birth, the udder was washed and disinfected and then 100 ml of colostrum were transferred into sterile tubes (BD falcon tube, United States). Colostrum samples were stored at –20 °C pending analysis.

Colostrum analysis. The amount of fat in the colostrum was determined using the Gerber method (Vandeputte-Van Messom and Burvenich 1989). The amount of crude protein in the colostrum samples was determined using the Kjeldahl method (Quigley et al. 1994), and the total protein concentration was determined using an autoana-

doi: 10.17221/40/2017-VETMED

lyser (BT 3000 plus, Biotecnical Inc, SPA, Rome, Italy) from three-fold distilled water-diluted colostrum samples. The activities of gamma glutamyltransferase (GGT), lactate dehydrogenase (LDH) and alkaline phosphatase (ALP) in colostrum samples were determined using an autoanalyser (BT 3000 plus, Biotecnical Inc, SPA, Rome, Italy). For this purpose, colostrum samples were centrifuged and fat was removed. Colostrum was diluted three times for LDH and ALP analysis and 150 times for GGT measurement. Colostrum IgG levels were measured with a commercially available test (Bovine Immunoglobulin ELISA kit/Bio-X Diagnostics S.A., Rochefort, Belgium) using a spectrophotometer (Thermo Multiskan GO, Microplate Spectrophotometer, USA). Concentrations of colostrum calcium (Ca), boron (B), cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), sodium (Na), potassium (K), magnesium (Mg), manganese (Mn), phosphorus (P), sulfur (S) and zinc (Zn) were determined using inductively coupled plasma atomic emission spectrometry (ICP-AES, Vista model, Varian, Australia).

Blood analysis. Serum IgG levels were measured with a commercially available test (Bovine Immunoglobulin ELISA kit/Bio-X Diagnostics S.A., Rochefort, Belgium) using a spectrophotometer (Thermo Multiskan GO, Microplate Spectrophotometer, USA). Concentrations of serum total protein (TP), albumin (ALB), globulin (GLB), blood urea nitrogen (BUN), GGT, LDH, aspartate aminotransferase (AST), ALP, alanine aminotransferase (ALT), calcium, phosphorus, magnesium, glucose, triglyceride and cholesterol were determined using an autoanalyser (BT 3000 plus, Biotecnical Inc, SPA, Rome, Italy). Serum samples were diluted with 20-fold distilled water for serum GGT analysis on the second day of calf life (Rocha et al. 2012). Concentrations of serum B, Zn, Fe, Cu, Mn, Cd, Cr, Na and S were determined using inductively coupled plasma atomic emission spectrometry (ICP-AES, Vista model, Varian, Australia).

Statistical analysis. The SPSS software program (Version 15.0, SPSS Inc., Chicago, USA) was used for statistical analysis. The data were expressed as mean ± standard error of the mean (SEM). The Kolmogorov-Smirnov test was used for normality. The difference between the two groups was determined using the independent *t*-test or Welch's *t*-test with unequal variances. Two-way repeated measures ANOVA was used to evaluate differences within each treatment group during the experiment and to determine the significance levels of variation. Sphericity was tested with Mauchly's test. If sphericity could not be assumed, we used the Greenhouse-Geisser correction for the evaluation of data. Pearson's correlation was used to investigate the relationship between IgG and biochemical parameters in colostrum and serum samples. Statistical significance was accepted at $P < 0.05$.

RESULTS

Composition and quality of colostrum

Colostrum analysis results are given in Table 1. The levels of IgG and boron in the colostrum of multiparous dairy cows were significantly ($P < 0.05$) higher than in the colostrum of primiparous dairy cows, while the concentrations of Ca, Mg, P, K, fat and LDH were found to be significantly lower ($P < 0.05$). A positive correlation was determined between colostrum crude protein and colostrum total protein. In addition, a significant correlation was found between colostrum IgG levels, crude protein, total protein levels and GGT activity ($P < 0.05$; Table 2).

Passive immune status in calves

Group 1 consisted of 13 males and 11 females while group 2 consisted of 12 males and 12 females. The passive transfer status of calves is shown in Table 3. No calf deaths were observed in either group over the course of 28 days. In the first group, no disease was found in 18 of the calves but diarrhoea was observed in six calves. One of the six calves that had diarrhoea had FPT; two of them had PFPT while three of them had APT. Cryptosporidium was detected in the faeces of these calves and the required treatment protocol was carried out; all of the calves recovered. Diarrhoea was noted in one calf, pneumonia in two calves and omphalitis in one calf, whereas no disease symptoms were seen in 20 of the calves in the second group. A failure of passive transfer was detected in one calf with diarrhoea, FPT was detected in one of the two calves with pneumonia and PFPT was detected in the other one. APT was detected in the calf with omphalitis.

doi: 10.17221/40/2017-VETMED

Table 1. Immunoglobulin G, fat, protein, biochemical parameters and mineral concentrations in the colostrums of primiparous (n = 24) and multiparous (n = 24) cows (mean ± SEM)

Parameters	Primiparous cows	Multiparous cows	P-values
IgG (g/l)	73.81 ± 10.65	117.45 ± 16.56	0.033
Alkaline phosphatase (U/l)	823.50 ± 143.05	1461.38 ± 306.76	0.068
Lactate dehydrogenase (U/l)	2221.75 ± 232.62	1540.88 ± 113.26	0.013
Glutamyl-transferase (U/l)	45 793.75 ± 3312.46	41 918.75 ± 2363.12	0.346
Total protein (g/l)	136.63 ± 9.79	154.81 ± 14.91	0.314
Fat (%)	7.46 ± 0.56	5.44 ± 0.65	0.022
Crude protein (%)	16.51 ± 0.70	18.12 ± 0.92	0.170
Ca (mmol/l)	84.07 ± 3.85	64.09 ± 2.41	< 0.001
B (µmol/l)	5.45 ± 1.63	10.47 ± 1.58	0.032
Cd (nmol/l)	163.70 ± 13.20	140.60 ± 13.30	0.214
Cr (nmol/l)	1749.93 ± 216.91	1821.08 ± 176.72	0.802
Cu (µmol/l)	12.68 ± 0.79	12.38 ± 0.29	0.075
Fe (µmol/l)	53.16 ± 2.33	57.28 ± 3.40	0.310
K(mmol/l)	55.06 ± 1.30	46.55 ± 1.98	0.001
Mg (mmol/l)	21.37 ± 0.80	18.73 ± 1.08	0.028
Mn (µmol/l)	6.38 ± 1.28	4.51 ± 0.13	0.162
Na (mmol/l)	32.05 ± 1.99	33.45 ± 1.97	0.618
P (mmol/l)	94.04 ± 3.83	73.56 ± 2.50	< 0.001
S (mmol/l)	57.53 ± 1.78	59.94 ± 4.42	0.617
Zn (µmol/l)	522.34 ± 31.82	450.28 ± 29.07	0.101

The levels of serum IgG and biochemical parameters of calves

The serum IgG, TP, GLB and GGT levels in calves in the first and second groups before receiving colostrum (day 0) and after receiving colostrum, on days 2, 7, 14 and 28, are shown in Table 4. Serum IgG concentrations in both groups showed a significant increase (P < 0.05) on days 2, 7, 14 and 28 as compared to day 0. When the IgG levels of groups were compared according to days, IgG levels of the 2nd group of calves on the 2nd, 7th, 14th and 28th days were determined to be significantly higher (P < 0.05) as compared to the 1st group of calves. TP and globulin concentrations of the 1st and 2nd group calves showed a significant increase (P < 0.05) on the 2nd, 7th, 14th and 28th days following colostrum consumption (P < 0.05) as compared to day 0. Moreover, the concentrations of TP and globulin on days 2 and 7 were found to be significantly higher (P < 0.05) in the second group as compared to the first group. A significant increase (P < 0.05) was seen in the serum GGT activities of calves of both groups after the intake of colostrum on the second day as compared to day 0. Second day serum GGT activity was observed to be significantly higher (P < 0.05) in calves of the second group as compared to the first group.

Table 2. Degree of correlation of colostrum immunoglobulin G (IgG), total and crude protein, fat, gamma-glutamyltransferase (GGT), alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) concentrations

Parameters	Total protein	IgG	Fat	GGT	ALP	LDH
Crude protein (%)	0.473**	0.657**	–0.188	0.274	0.127	–0.002
Total protein (g/l)		0.366*	–0.121	0.142	0.185	–0.133
IgG (g/l)			–0.103	0.319*	0.091	–0.109
Fat (%)				0.078	–0.146	0.267
GGT (U/l)					–0.141	0.082
ALP (U/l)						0.121

*P < 0.05; **P < 0.01

Changes in the levels of serum albumin, ALP, AST, LDH, ALT, total cholesterol, triglyceride, BUN and glucose are shown in Table 5. Serum albumin concentrations on days 14 and 28 showed a significant increase (P < 0.05) in both groups as compared to day 0. A significant increase (P <

Table 3. Passive immunity status of calves (n (%)) on the second day after colostrum consumption

	Immunoglobulin G levels (g/l)		
	< 8	8–16	> 16
Group 1 (n = 24)	4 (16.66)	13 (54.16)	7 (29.16)
Group 2 (n = 24)	3 (12.50)	7 (29.16)	14 (58.33)

doi: 10.17221/40/2017-VETMED

Table 4. Changes in serum immunoglobulin G (IgG), total protein (TP), globulin (GLB) concentrations and gamma-glutamyltransferase (GGT) enzyme activities of group 1 (n = 24) and group 2 (n = 24) calves (mean ± SEM)

Parameters	Group	Days				
		0	2	7	14	28
IgG (g/l)	1	0.11 ± 0.02^{A}	14.29 ± 1.51^{a,B}	12.47 ± 1.29^{a,B}	10.99 ± 1.05^{a,B}	12.02 ± 0.76^{a,B}
	2	0.11 ± 0.02^{A}	19.29 ± 1.86^{b,B}	17.34 ± 1.70^{b,B}	15.70 ± 1.33^{b,B}	15.22 ± 1.17^{b,B}
TP (g/l)	1	42.10 ± 0.64^{A}	57.01 ± 1.76^{a,B}	58.04 ± 1.12^{a,B}	57.35 ± 1.10^{B}	58.48 ± 0.92^{B}
	2	42.83 ± 1.26^{A}	62.87 ± 1.83^{b,B}	62.90 ± 1.79^{b,B}	58.96 ± 1.45^{B}	58.72 ± 1.29^{B}
GLB (g/l)	1	8.87 ± 0.30^{A}	25.36 ± 1.55^{a,C}	23.53 ± 1.20^{a,BC}	20.50 ± 0.94^{B}	20.17 ± 1.02^{B}
	2	9.45 ± 1.18^{A}	31.22 ± 1.99^{b,D}	28.85 ± 1.82^{b,CD}	23.28 ± 1.33^{BC}	22.26 ± 1.25^{B}
GGT (U/l)	1	12.29 ± 0.64^{A}	452.86 ± 53.98^{a,E}	210.67 ± 39.30^{D}	84.25 ± 11.70^{C}	33.50 ± 3.51^{B}
	2	12.95 ± 0.78^{A}	712.50 ± 102.28^{b,E}	195.42 ± 32.82^{D}	103.50 ± 14.78^{C}	37.54 ± 3.48^{B}

Different letters in the same rows (A, B, C, D, E) and columns (a, b) are statistically significant (P < 0.05)

0.05) was seen in serum ALP, AST, LDH and ALT enzyme activities in calves of both groups following the intake of colostrum on the 2nd day as compared to day 0. Significant changes (P < 0.05) were detected in the concentrations of cholesterol, triglyceride, glucose and BUN in both groups af-

Table 5. Changes in serum biochemical parameters in group 1 (n = 24) and group 2 (n = 24) calves (mean ± SEM)

Parameters	Group	Days				
		0	2	7	14	28
Albumin (g/l)	1	33.23 ± 0.44^{B}	31.94 ± 0.46^{A}	34.30 ± 0.40^{B}	36.60 ± 0.57^{C}	38.26 ± 0.53^{a,D}
	2	33.19 ± 0.44^{B}	31.65 ± 0.46^{A}	34.14 ± 0.38^{C}	35.60 ± 0.57^{D}	36.47 ± 0.57^{b,DE}
Aspartate aminotransferase (U/l)	1	24.83 ± 1.90^{A}	55.29 ± 3.47^{B}	26.54 ± 0.96^{A}	34.71 ± 3.71^{A}	31.87 ± 2.01^{A}
	2	32.46 ± 4.50^{A}	56.29 ± 3.69^{B}	28.00 ± 1.51^{A}	28.17 ± 2.11^{A}	36.21 ± 3.18^{A}
Alkaline phosphatase (U/l)	1	504.71 ± 50.26^{B}	705.58 ± 49.76^{C}	561.63 ± 43.64^{BC}	417.29 ± 31.86^{AB}	341.29 ± 24.62^{A}
	2	463.46 ± 33.38^{AB}	642.29 ± 37.48^{C}	564.04 ± 38.25^{BC}	441.33 ± 31.38^{AB}	402.13 ± 34.98^{A}
Lactate dehydrogenase (U/l)	1	946.04 ± 32.32^{A}	1623.13 ± 55.04^{D}	1137.58 ± 34.90^{C}	1028.86 ± 49.87^{AB}	1184.83 ± 53.71^{B}
	2	986.71 ± 51.84^{A}	1555.33 ± 66.39^{C}	1124.21 ± 32.75^{AB}	1011.38 ± 41.09^{A}	1220.92 ± 57.00^{B}
Alanine aminotransferase (U/l)	1	4.71 ± 0.35^{A}	16.21 ± 1.55^{B}	7.79 ± 0.51^{A}	8.17 ± 1.17^{A}	6.83 ± 1.07^{A}
	2	5.54 ± 0.84^{A}	17.00 ± 1.96^{B}	8.08 ± 0.74^{A}	7.71 ± 2.19^{A}	5.46 ± 0.72^{A}
Blood urea nitrogen (mmol/l)	1	4.42 ± 0.19^{B}	3.62 ± 0.20^{AB}	3.36 ± 0.15^{A}	3.94 ± 0.36^{AB}	3.26 ± 0.14^{A}
	2	4.73 ± 0.21^{C}	3.41 ± 0.18^{AB}	3.27 ± 0.11^{AB}	3.87 ± 0.29^{ABC}	3.11 ± 0.11^{A}
Cholesterol (mmol/l)	1	0.68 ± 0.04^{A}	1.24 ± 0.06^{B}	1.41 ± 0.05^{B}	1.46 ± 0.09^{BC}	1.92 ± 0.13^{C}
	2	0.76 ± 0.03^{A}	1.07 ± 0.06^{B}	1.62 ± 0.11^{C}	1.48 ± 0.11^{C}	1.87 ± 0.13^{C}
Triglyceride (mmol/l)	1	0.31 ± 0.02^{A}	0.63 ± 0.08^{B}	0.32 ± 0.03^{A}	0.25 ± 0.03^{A}	0.30 ± 0.03^{A}
	2	0.32 ± 0.02^{B}	0.48 ± 0.04^{C}	0.36 ± 0.04^{BC}	0.20 ± 0.02^{A}	0.26 ± 0.02^{AB}
Glucose (mmol/l)	1	4.90 ± 0.49^{A}	6.05 ± 0.21^{B}	5.76 ± 0.20^{AB}	5.16 ± 0.19^{AB}	5.31 ± 0.16^{AB}
	2	3.85 ± 0.39^{A}	5.89 ± 0.18^{C}	5.95 ± 0.24^{C}	5.03 ± 0.13^{AB}	4.86 ± 0.21^{AB}
P (mmol/l)	1	2.53 ± 0.05^{A}	2.97 ± 0.08^{B}	3.12 ± 0.07^{B}	3.05 ± 0.08^{B}	3.03 ± 0.07^{B}
	2	2.45 ± 0.08^{A}	2.77 ± 0.08^{B}	2.95 ± 0.14^{B}	3.02 ± 0.08^{B}	3.00 ± 0.08^{B}
Ca (mmol/l)	1	2.82 ± 0.04^{B}	2.70 ± 0.04^{B}	2.46 ± 0.05^{A}	2.31 ± 0.03^{A}	2.33 ± 0.04^{A}
	2	2.92 ± 0.04^{D}	2.66 ± 0.04^{C}	2.55 ± 0.04^{BC}	2.36 ± 0.03^{A}	2.42 ± 0.05^{AB}
Mg (mmol/l)	1	0.66 ± 0.02^{C}	0.57 ± 0.02^{AB}	0.60 ± 0.01^{B}	0.57 ± 0.01^{AB}	0.53 ± 0.02^{A}
	2	0.67 ± 0.01^{C}	0.55 ± 0.02^{A}	0.62 ± 0.01^{BC}	0.56 ± 0.01^{AB}	0.56 ± 0.02^{AB}

Different letters in the same rows (A, B, C, D, E) and columns (a, b) are statistically significant (P < 0.05)

doi: 10.17221/40/2017-VETMED

Table 6. Correlation between immunoglobulun G levels and selected biochemical parameters in group 1 (n = 24) and group 2 (n = 24) calves

	Day	TP	GLB	GGT	AST	ALT	ALP	LDH
Group 1	0	0.018	0.150	–0.002	0.198	–0.112	–0.114	0.162
	2	0.735**	0.808**	0.761**	0.320	0.025	–0.333	0.320
	7	0.692**	0.831**	0.457*	0.429*	–0.020	0.317	0.147
	14	0.451*	0.698**	0.609**	0.240	0.085	0.092	–0.125
	28	0.510*	0.814**	0.122	–0.085	–0.376	0.045	0.014
Group 2	0	0.332	0.446**	0.122	–0.057	–0.062	0.012	–0.088
	2	0.771**	0.816**	0.379	–0.058	–0.161	0.349	0.035
	7	0.889**	0.919**	0.426*	0.054	0.294	0.141	0.019
	14	0.848**	0.918**	0.729**	–0.110	–0.288	–0.022	–0.373
	28	0.809**	0.881**	0.489*	–0.203	–0.118	0.296	0.193
Total	0	0.183	0.207	0.068	0.011	–0.072	–0.057	0.002
	2	0.758**	0.818**	0.554**	0.107	–0.075	–0.038	0.113
	7	0.829**	0.891**	0.398**	0.200	0.192	0.210	0.060
	14	0.678**	0.825**	0.678**	0.002	–0.177	0.052	–0.246
	28	0.680**	0.851**	0.354*	–0.144	–0.255	0.262	0.137

ALP = alkaline phosphatase, ALT = alanine aminotransferase, AST = aspartate aminotransferase, GGT = gamma-glutamyltransferase, GLB = globulin, LDH = lactate dehydrogenase, TP = total protein

*P < 0.05; **P < 0.01

ter the consumption of colostrum up until the 28th day (Table 5).

A positive correlation was determined between serum IgG levels and serum GLB and TP levels in the first group of calves on days 2, 7, 14 and 28; between serum IgG and GGT activity on days 2, 7 and 14; between serum IgG and serum AST activity on the 7th day only. As for the 2nd group, a positive correlation was determined between serum IgG and GLB levels on days 0, 2, 7, 14, 28; between serum IgG and TP levels on days 2, 7, 14 and 28; between serum IgG and GGT activities on days 7, 14 and 28 (Table 6).

Changes in the concentrations of Ca, P, Mg, B, Cd, Cu, Cr, Fe, Mn, S, Na and Zn in calves over the period of 28 days are shown in Table 5 and Table 7. In both groups, a significant fall (P < 0.05) was determined in serum levels of Fe, Mn and B after the consumption of colostrum, while a significant increase (P < 0.05) was determined in serum levels of Cu, P and S. When the initial (day 0) trace element levels of calves of the first and second groups are examined, serum B, Fe and S levels of calves delivered by primiparous dairy cows (Group 1) were observed to be significantly lower (P < 0.05) as compared to those values in calves delivered by multiparous dairy cows (Group 2).

DISCUSSION

The quality of colostrum is related to its level of IgG and the mother's age/parity is also indicated to exert an influence (Godden 2008; Rocha et al. 2012; Gokce and Erdogan 2013). Tyler et al. (1999a) indicated that the amount of IgG in the colostrum of cows lactating three or more calves is 19.5 grams more than the amount of IgG in the colostrum of primiparous cows. In the present study, the IgG concentration in the colostrum of multiparous cows was found to be significantly higher (P < 0.05) than that in the colostrum of primiparous cows (Table 1). Godden (2008) indicated that colostrum containing > 50 g/l of IgG is of high quality. In this study, mean colostral IgG levels were 117.45 g/l in multiparous cows and 73.81 g/l in primiparous cows, showing that both primiparous and multiparous cows have high quality colostrums.

Colostrum has high protein content (Godden 2008). The protein level in colostrum is conventionally determined using the Kjeldahl method (Quigley et al. 1994). Here, despite the colostrum level of total protein (g/l) being lower than the crude level of protein (%), we determined that using an autoanalyser, which is easier than the Kjeldahl method, may be employed to measure the protein

doi: 10.17221/40/2017-VETMED

Table 7. Changes in serum trace element levels in group 1 (n = 24) and group 2 (n = 24) calves (mean ± SEM)

Parameters	Group	Days			
		0	2	7	14
B (µmol/l)	1	24.12 ± 4.85^{a,C}	10.49 ± 2.25^{a,B}	8.82 ± 1.57^{a,B}	2.04 ± 0.79^{a,A}
	2	74.54 ± 9.87^{b,B}	50.05 ± 8.33^{b,B}	48.31 ± 8.16^{b,B}	0.22 ± 0.14^{b,A}
Cd (nmol/l)	1	152.14 ± 15.12	145.02 ± 12.63	147.69 ± 13.88	135.23 ± 12.10
	2	142.35 ± 12.81	122.78 ± 13.17	156.59 ± 8.90	162.82 ± 11.83
Cr (nmol/l)	1	909.58 ± 135.96	740.36 ± 92.88	790.35 ± 116.92	659.59 ± 79.80
	2	990.35 ± 139.42^{B}	769.78 ± 81.34^{AB}	770.55 ± 54.81^{AB}	603.82 ± 35.00^{A}
Cu (µmol/l)	1	12.03 ± 0.38^{A}	16.16 ± 0.37^{B}	20.04 ± 1.08^{a,C}	18.77 ± 0.65^{C}
	2	13.78 ± 0.81^{A}	18.04 ± 0.98^{B}	24.46 ± 1.26^{b,C}	20.53 ± 0.66^{B}
Fe (µmol/l)	1	63.01 ± 4.12^{a,B}	38.13 ± 2.51^{a,A}	38.66 ± 3.76^{a,A}	33.12 ± 2.86^{A}
	2	104.36 ± 10.02^{b,B}	90.93 ± 14.32^{b,B}	80.01 ± 9.67^{b,B}	34.37 ± 4.12^{A}
Mn (µmol/l)	1	4.81 ± 0.23^{B}	4.10 ± 0.04^{AB}	4.35 ± 0.14^{AB}	4.00 ± 0.01^{A}
	2	4.52 ± 0.11^{AB}	4.42 ± 0.16^{AB}	4.64 ± 0.12^{B}	4.09 ± 0.05^{A}
Na (mmol/l)	1	125.30 ± 1.39^{a,BC}	126.91 ± 1.44^{a,BC}	121.25 ± 1.18^{AB}	115.27 ± 1.89^{A}
	2	116.31 ± 2.15^{b}	116.49 ± 2.69^{b}	115.86 ± 2.46	115.23 ± 2.14
S (mmol/l)	1	17.94 ± 0.32^{a,A}	22.38 ± 0.72^{B}	22.39 ± 0.45^{a,B}	22.43 ± 0.50^{B}
	2	19.42 ± 0.50^{b,A}	24.25 ± 0.80^{B}	24.49 ± 0.77^{b,B}	23.29 ± 0.71^{B}
Zn (µmol/l)	1	19.93 ± 1.26	18.48 ± 1.50	16.23 ± 1.18	15.56 ± 0.73
	2	19.82 ± 1.71	18.19 ± 1.18	17.27 ± 0.97	15.62 ± 0.76

Different letters in the same rows (A, B, C) and columns (a, b) are statistically significant ($P < 0.05$)

concentration of colostrum due to the significant ($P < 0.05$) positive correlation between the two techniques.

The fat content of colostrum is higher than that of normal milk. High fat content is indicated to have an important role in providing calves with energy (Quigley and Drewry 1998; Blum and Hammon 2000). Zarcula et al. (2010) determined that the fat content of colostrum in Holstein cows decreases as the number of lactations increase, by 7.87%, 5.38% and 5.6%, respectively, in the 2nd, 3rd and 4th lactations. In the work presented here, mean colostrum fat content in multiparous cows was determined to be 5.44%, while in primiparous cows the value was 7.46%. The possible reason for the low colostral fat content in multiparous cows could be related to the production of colostrum in high amounts (dilution effect).

It has been stated that colostrum enzyme activities can also be used in the evaluation of colostrum quality (Lombardi et al. 2001; Zarrilli et al. 2003a; Zarrilli et al. 2003b). Colostrum has a high GGT activity (Blum and Hammon 2000; Zanker et al. 2001). Rocha et al. (2012) reported that colostrum GGT activity in primiparous cows is higher than in multiparous cows and this situation can be due to the dilution effect associated with multiparous cows producing a higher amount of colostrum as compared to primiparous cows. In our research, mean colostrum GGT activity was found to be higher in primiparous cow colostrum than in multiparous cow colostrum, supporting the results of Rocha et al. (2012). Moreover, colostrum ALP and LDH enzyme activities are indicated to be high in the first colostrum that is milked, followed by a decrease in the following milking (Zanker et al. 2001; Maden et al. 2004; Rocha et al. 2012). According to our findings, the absence of a positive correlation between the levels of colostral IgG and ALP and LDH, on the one hand, and the positive correlation between the levels of colostral IgG and GGT, on the other hand, show that GGT activity can be used in the determination of colostrum quality (Table 2).

The principal mineral source of newborn calves following birth is colostrum. The concentration of Ca, Mg, P and Cl is indicated to be higher in cow colostrum as compared to milk; K levels are lower while those of Fe are indicated to be 10–17 times higher than in milk (Bastan 2013). Kume and Tanabe (1993) reported that colostrum Ca, P, Mg, Na, Fe, Zn, Cu and Mn levels are high in the first milking and decrease in the following milking. In

doi: 10.17221/40/2017-VETMED

the same research, Ca, P and Mg levels were described to be higher in primiparous cow colostrum as compared to multiparous cow colostrum. In our research, meanwhile, it was determined that Ca, P, Mg and K concentrations on primiparous cow colostrum were significantly higher ($P < 0.05$) as compared to those in multiparous cow colostrum while the B concentration was significantly lower ($P < 0.05$) (Table 1). Possible reasons for colostrum mineral levels being higher in primiparous cows compared to multiparous cows in our research could be related to mineral levels in the ration or to fodder utilisation and daily milk production.

An intake of good-quality colostrum at the right time and in sufficient amounts is required for sufficient passive immunity in new-born calves (Weaver et al. 2000; Godden 2008; Gokce and Erdogan 2013). Morbidity and mortality rates in the neonatal period decrease in calves taking in sufficient immunoglobulin in colostrum in the first hours of life (Morin et al. 2001). It is possible to determine passive immune transfer by measuring serum IgG concentrations in new-born calves 24 and 48 hours following the feeding of colostrum (Filteau et al. 2003; Gungor et al. 2004; Smith and Foster 2007). In our study, FPT and PFPT were determined to be higher in the first group of calves as compared to the second group of calves, while the adequate passive transfer rate was determined to be lower (Table 3). In other research, FPT levels in calves delivered by primiparous cows were found to be higher as compared to those from multiparous cows (Furman-Fratczak et al. 2011; Gokce and Erdogan 2013). According to the public database of the National Animal Health Monitoring System of the United States of America, the FPT rate in dairy cattle in 1991 was 41%, and it fell to 19.2% in 2007 (NAHMS 2010). Tyler et al. (1998) reported an FPT rate of approximately 34.5% in research conducted on 3479 calves. In this work, the possible reasons for the low rate (14.5%) of failure of passive transfer in all of the calves included in this study can be related to the low number of animals, taking samples from a single farm and the high standard of caring, nutrition and colostrum management.

Strong correlations have been determined between serum IgG levels and serum TP concentrations in neonatal calves (Vandeputte et al. 2011; Rocha et al. 2012). A serum IgG level of 10 g/l in calves is regarded to be equal to a serum TP concentration of 52 g/l (Tyler et al. 1999b). In this study, serum IgG concentrations of calves whose serum TP concentrations were high were also high (Table 8). It was observed that the serum TP concentrations of the second group of calves on the second day following the consumption of colostrum (62.87 g/l) was significantly higher ($P < 0.05$) than those of the first group of calves (57.01 g/l); as well as that, the serum TP concentrations were increased significantly ($P < 0.05$) in both groups on days 2, 7, 14 and 28 as compared to day 0 (Table 4). While some researchers (Feitosa et al. 2010; Rocha et al. 2012) have observed a significant difference ($P < 0.05$) following the consumption of colostrum between serum TP concentrations in calves delivered by cattle that had a single delivery and cattle that were multiparous, Costa et al. (2008) reported no differences between serum TP concentrations following consumption of colostrum in calves delivered by cattle that had a single delivery and cattle that were multiparous. In our research, calves from the second group had higher TP concentrations compared to the first group, which is in agreement with the research of Feitosa et al. (2010) and Rocha et al. (2012).

Table 8. Changes in calf immunoglobulin (IgG), total prpotein (TP) and gamma-glutamyltransferase (GGT) activities according to immune status on the 2nd day after colostrum consumption (mean ± SEM)

Parameters	FPT ($n = 7$)	PFPT ($n = 20$)	APT ($n = 21$)
IgG (g/l)	4.64 ± 0.76	11.86 ± 0.52	24.84 ± 1.04
TP (g/l)	46.20 ± 2.46	56.27 ± 1.26	66.81 ± 1.29
GGT (U/l)	275.00 ± 87.55	459.00 ± 66.15	830.53 ± 115.73

APT = adequate passive transfer, FPT = failure of passive transfer, PFPT = partial failure of passive transfer

In the diagnosis of FPT, the threshold value for TP concentration is indicated to be 42 g/l on the first day following consumption of colostrum; this value has a sensitivity of 80% and a specificity of 100% (Perino et al. 1993). Tyler et al. (1999a) reported that serum TP concentrations must be above 55 g/l for adequate passive transfer. A serum TP concentration that is > 60 g/l in newborn calves with normal hydration (not dehydrated) is regarded as representing adequate passive transfer while a concentration of < 50 g/l is regarded as FPT (Turgut 2000). In our research, the mean serum TP level in calves with FPT was 46.20 g/l; in calves with PFPT it was 56.27 g/l and in calves with APT the value was 66.81 g/l (Table 8). This shows that TP can be useful in the evaluation of passive transfer.

doi: 10.17221/40/2017-VETMED

In our study, serum GGT activity increased (37-fold and 55-fold in the 1st and 2nd groups, respectively) on the second day. Parish et al. (1997) reported that daily serum GGT activities in calves on days 1, 4 and 7 were 200 U/l, > 100 U/l and > 75 U/l respectively, and that calves with a GGT activity that is < 50 U/l in the first two weeks have FPT. While the GGT activity on the 2nd day was > 200 U/l in 42 calves included in our research, it was 80–200 U/l in six calves. At the same time, individual IgG levels and GGT activities of calves showed a change in the same direction. These findings show that 2nd day GGT activity is a useful parameter in the evaluation of passive transfer. When the individual serum GGT activities on the 14th day were evaluated, serum GGT activity was > 50 U/l in 36 calves, while lower in the remaining calves. This shows that GGT activity is not a useful parameter in the evaluation of passive transfer from the 14th day onwards. Gungor et al. (2004) reported that there is a positive correlation between the serum GGT activity and serum IgG levels of calves consuming colostrum. Perino et al. (1993) indicated that serum GGT activity increased 26-fold on the 1st day following colostrum consumption as compared to the beginning, and that there was a correlation between serum GGT activity and protein concentration and the rise in serum IgG. In this study, we observed a significant ($P < 0.05$) positive correlation between mean serum IgG levels and GGT activity, TP and globulin concentrations in calves among days 2, 7, 14 and 28. Despite this, due to individual differences, these parameters, alongside IgG, are more beneficial in the evaluation of passive transfer in calves on day 2 as well.

The increase in ALP activity in newborn calves is stated to be related to endogenous enzyme production together with colostrum consumption (Zanker et al. 2001; Rocha et al. 2012). In our study, after colostrum consumption, an increase in ALP activity was observed in both groups on the 2nd day but a decrease was determined on the 7th, 14th and 28th days (Table 5). However, the absence of a positive correlation between serum ALP activity and serum IgG levels on the 2nd day shows that it is not a useful parameter in the evaluation of passive transfer in calves (Table 6). Pekcan et al. (2013) indicated that AST, LDH and ALT enzymes are not suitable parameters in the evaluation of passive transfer state. In this study as well, despite the significant increase ($P < 0.05$) in ALT, AST and LDH activities on the 2nd day, a correlation with IgG levels could not be determined (Table 6). These results show that 2nd day serum AST, ALT and LDH activities are not informative parameters in the determination of passive transfer state in calves.

The triglyceride concentration of newborn calves is markedly influenced by fat absorption and colostrum consumption time and amount (Kuhne et al. 2000). In this work, an increase in serum TG concentrations was determined in both groups on the 2nd day following colostrum consumption; as for later, a gradual decrease was determined on the 7th, 14th and 28th days (Table 5). The possible cause of the increase in higher serum TG in the 1st group compared to the 2nd group following colostrum consumption could be based on significantly higher ($P < 0.05$) colostrum fat content of primiparous cows compared to that of multiparous cows.

Colostrum is one of the most important sources of minerals for newborn calves. While colostrum mineral concentrations, such as those of Ca, P, Mg, Fe, Zn, Cu and Mn, are high in the first milking, a decrease in mineral content occurs over time after birth (Foley and Otterby 1978; Kincaid and Cronrath 1992). It has been reported that the serum mineral state of newborn calves is not only related to colostrum intake, but can be related at the same time to placental mineral transfer (Kume and Tanabe 1993). Blum and Hammon (2000) reported that colostrum consumption time/amount of consumed colostrum does not have a significant influence on concentrations of Ca, P, Mg and Fe. In our research, a gradual decrease was seen in serum Fe concentrations of both groups over a period of 14 days. Moreover, serum Fe levels in the first group of calves on days 0, 2 and 7 were determined to be significantly lower ($P < 0.05$) compared to those of calves in the 2nd group (Table 7). Iron deficiency and the resulting anaemia can be seen later on in calves consuming milk (Kincaid 1999). Egli and Blum (1998) reported that plasma Fe concentrations of calves decrease in the first 28 days before showing a subsequent increase. Kume and Tanabe (1993) reported that the placental Fe transfer level of calves delivered by cattle that had their first delivery is low due to the high Fe demand of growing calves, and that these calves have a low haemoglobin level. In our study, low serum Fe levels of calves in the 1st group could possibly be related to low placental and colostral Fe transfer (Table 7). Therefore, iron supplementation after birth can be beneficial,

doi: 10.17221/40/2017-VETMED

especially to the calves of primiparous cow. In our work, calf serum mineral levels on day 0 were within the normal range, showing that placental transfer of minerals to calves was effective and also that, changes in mineral levels in the days after birth can take place depending on colostrum and milk intake.

In conclusion, we have here determined that the colostrum of multiparous cows is of better quality than that of primiparous cows, that protein and GGT activity can be used alongside IgG in the determination of colostrum quality and that an autoanalyser can be useful in determining colostrum protein levels. Moreover, it was determined that adequate passive transfer rates were more common in the calves of multiparous cows than in those of primiparous cows, that 2nd day TP, globulin and GGT levels in particular can be used in the determination of FPT in calves since individual differences can be observed on the 7th and 14th days, that placental transfer can be important for mineral and trace element levels in newborn calves alongside colostrum, that calves delivered by primiparous cows have a lower iron level compared to those delivered by multiparous cows and that iron deficiency can develop in calves in the first 14 days following birth. Thus, iron supplementation could be beneficial for calves after birth, in particular for those born to primiparous cows.

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Received: March 13, 2017
Accepted after corrections: December 15, 2017