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Serum immunoglobulin concentrations after feeding maternal colostrum or maternal colostrum plus colostral supplement to dairy calves

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Summary

Maternal colostrum or maternal colostrum plus colostral supplement, composed of a blend of lyophilized colostrum and dried whey, was fed to 32 Holstein calves as soon as possible after birth (mean ± SEM = 2.0 ± 0.2 hours) and, again, 12 hours later. Mean immunoglobulin concentration in colostrum was 59.2 mg/ml; mean immunoglobulin fraction in supplement was 11.4%. Serum immunoglobulin concentrations were measured at 0, 12, 24, 48, and 72 hours, and at 28 and 56 days. Hour/treatment interactions were significant for total immunoglobulin, IgG_1, and IgM concentrations. Immunoglobulin concentrations were highest at 12 hours (total immunoglobulin, IgG_1, IgM) or 24 hours (IgG_2) in calves fed colostrum plus supplement, whereas all immunoglobulin concentrations were highest at 24 hours in calves fed maternal colostrum only. Peak mean immunoglobulin concentrations did not differ between treatments. Supplementation of colostrum did not increase peak mean serum immunoglobulin concentration, but did alter the serum concentration-time profile from 12 to 72 hours after birth.

Calves are born hypo- or agammaglobulinemic[1-4]; therefore, absorption of colostral immunoglobulins is essential for acquisition of passive immunity.[1,2,5] Among the most important factors that influence passive transfer of colostral immunoglobulins are age at first feeding and mass of immunoglobulins ingested.[5-10] In some circumstances, sufficient maternal colostrum may not be available within the first 24 hours after birth, or colostrum with low immunoglobulin concentration may be produced by the dam. In these instances, products containing immunoglobulins extracted from dried cheese whey or natural colostrum have been used to replace colostrum or to increase immunoglobulin content of colostrum.[11] However, feeding excess immunoglobulin may impair development of active immunity[4,12] and reduce efficiency of immunoglobulin absorption.[5,13-15] Effects of these products on acquisition of passive immunity have not been determined, particularly when they are fed to supplement colostrum containing various concentrations of immunoglobulins. The purpose of the study reported here was to determine whether adding a colostral supplement to maternal colostrum had an effect on serum immunoglobulin concentration in calves.

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Materials and Methods

Experimental design—Thirty-two Holstein calves (16 bulls and 16 heifers) were assigned to a 2× 2 factorial arrangement of sex and colostral supplementation in a randomized complete-block experimental design. Calves were blocked according to birth date, and were fed maternal colostrum (MC) or maternal colostrum supplemented (MCS) with 125 g of a colostral supplement consisting of a blend of lyophilized bovine colostrum and dried whey.[a]

Animal management—Pregnant cows were housed in a drylot until 7 days before their expected calving dates; they were then moved to 5- × 3-m stalls bedded with sawdust. Eight cows calved in the drylot more than 7 days before their expected calving date. Newborn calves were not allowed to suckle, but were left with their dams for 30 to 40 minutes to allow each dam to clean and stimulate its calf. Calves were then moved to the calf barn and weighed. Their navels were dipped in 7% iodine solution, and calves were placed in individual pens (2.5 × 1.5 m) bedded with shavings. Pens were cleaned daily, and shavings were replaced every other week.

[a] Immuno Bac, Alltech Inc, Nicholasville, Ky.

Maternal colostrum was hand-milked from the dam shortly after parturition, transported to the calf barn, weighed, and placed into two 2-L bottles. When < 4 L of colostrum was obtained from any cow, previously obtained colostrum was added to make 4 L. Surplus colostrum was stored at 4 C or −20 C, and frozen colostrum was thawed with warm water before use.

Two liters of MC or MCS was fed to each calf as soon as possible after birth, and again, 12 hours later. After 24 hours, all calves were fed commercial milk replacer twice daily. Commercial calf starter was offered after 3 days, and water was available at all times. Intake of MC, MCS, and milk replacer were recorded at each feeding. Prevalence of scours and signs of respiratory tract infections and other illnesses were reported daily.

Sample collection and analysis of immunoglobulin concentration—Samples (10 ml) of colostrum were stored at −20 C prior to analysis of IgG_1, IgG_2, IgM, and IgA concentrations by radial immunodiffusion.[b] Colostrum was diluted (10:1) in saline (0.9% NaCl) solution for analysis of IgG_1 concentration. Colostral supplement was reconstituted 10:1 with tap water and analyzed as for colostrum. After 22 hours of incubation at 22 C, diameter of precipitant rings was measured by use of an ocular micrometer on a stereomicroscope.[c] An effort was not made to determine whether immunoglobulins in colostral supplement were intact proteins or whether processing affected immunoglobulin structure.

Blood samples (10 ml) were collected by jugular venipuncture into evacuated containers without anticoagulant as soon as possible after birth (0 hours) and 12, 24, 48, and 72 hours thereafter, and when calves were 28 and 56 days old. Blood samples were allowed to coagulate, and serum was obtained by centrifugation (3,000 × g). Samples were stored at −20 C prior to analysis of IgG_1, IgG_2, and IgM concentrations as for colostrum, but without dilution.

Statistical analysis—Serum immunoglobulin concentrations at 12, 24, 48, and 72 hours were analyzed as a split-plot experimental design. The model used was $Y_{ijkl} = \mu + B_i + S_j + T_k + ST_{jk} + BST_{ijk} + H_l + TH_{kl} + SH_{jl} + BW + Ig + e_{ijkl}$; where Y_{ijkl} = serum immunoglobulin concentration; μ = overall mean concentration; B_i = effect of i^{th} block; S_j = effect of the j^{th} sex; T_k = effect of k^{th} treatment; ST_{jk} = effect of sex/treatment interaction; BST_{ijk} = effect of block/sex/treatment interaction; H_l effect of l^{th} hour; TH_{kl} = effect of treatment/hour interaction; SH_{jl} = effect of sex/hour interaction; BW = body weight at birth (included as a covariable); Ig = cumulative intake of immunoglobulin from maternal colostrum (included as a covariable); and e_{ijkl} = residual. Block/sex/treatment interaction was used as an error term to test treatment and sex main effects and sex/treatment interaction. Analyses were conducted by use of a mixed model algorithm.[16] Serum immunoglobulin concentrations at 28 and 56 days of age were analyzed as a randomized complete block design, using analysis of covariance, with BW and cumulative immunoglobulin intake as covariables. Values of $P < 0.05$ were considered significant. Data for one heifer were removed prior to statistical analysis, because it refused to consume the second feeding; therefore, least squares means are presented.

[b] VMRD Inc, Pullman, Wash.

[c] American Optical, Buffalo, NY.

Results

Body weight at birth ranged from 26.3 to 50.8 kg (Table 1). Mean age at first feeding was 2.0 hours (SEM = 0.2). Ten calves refused some MC (n = 4) or MCS (n = 6) during the first or second feeding. Any MC or MCS that was not initially consumed was consumed in an additional feeding 2 to 4 hours later. Signs of respiratory tract disease were not observed during the study. Five calves (2 that were fed MC, 3 that were fed MCS) were treated for scours for > 1 day during the study; 8 others were treated for 1 day only.

Mean total immunoglobulin concentration in colostrum did not differ between treatments and, for all calves, ranged from 6.3 to 94.9 mg/ml. For all calves, IgG_1, IgG_2, IgM, and IgA were 85.3, 6.2, 3.4, and 5.1%, respectively, of mean total immunoglobulin concentration. Colostral supplement was obtained and fed in 2 lots, which contained 14.3 and 8.5% total immunoglobulin.

Mean cumulative intake of total immunoglobulin and mean cumulative intake of each isotype did not differ by treatment (Table 2). For those calves that

Table 1—Statistics of body weight, immunoglobulin concentration of maternal colostrum, colostral supplement, and age at first blood sample and feeding

Variable	n	Min	Max	Mean	SEM
Body weight (kg)	31	26.3	50.8	38.9	1.0
Age at first feeding (h)	31	0.8	4.0	2.0	0.2
Age at first sampling (h)	31	0.5	2.5	1.0	0.1
Maternal colostrum (mg/ml)					
Total immunoglobulin	31	6.3	94.9	59.2	4.1
IgG_1	31	4.6	80.8	50.6	3.6
IgG_2	31	0.2	7.4	3.7	0.3
IgM	31	0.6	5.7	2.0	0.2
IgA	31	0.1	6.6	3.0	0.4
Colostral supplement (%)					
Total immunoglobulin	2	8.5	14.3	ND	ND
IgG_1	2	6.7	11.7	ND	ND
IgG_2	2	0.7	1.2	ND	ND
IgM	2	0.4	0.5	ND	ND
IgA	2	0.5	0.8	ND	ND

ND = not determined.

Table 2—Least-squares means of cumulative immunoglobulin intake by calves fed maternal colostrum (MC) or maternal colostrum supplemented (MCS) with 125 g of colostral supplement

Immuno-globulin (g)	Colostrum			Supplement	
	MC	MCS	SEM	MCS	SEM
First feeding					
IgG_1	100.4	99.7	8.5	12.4	0.4
IgG_2	7.5	7.7	1.0	1.4	0.1
IgM	4.3	3.4	0.4	0.7	0.1
Total	112.2	110.8	9.0	14.5	0.4
Second feeding					
IgG_1	198.8	198.7	15.0	24.5	0.6
IgG_2	14.8	15.2	1.8	2.8	0.1
IgM	8.6	6.7	0.8	1.3	0.1
Total	222.2	220.5	17.0	28.7	0.6

received colostral supplement, the supplement provided a mean additional 14.5 and 14.2 g of immunoglobulins at the first and second feedings, respectively.

Serum immunoglobulin concentrations in all calves prior to feeding were less than the limits of detection by radial immunodiffusion (0.11, 0.12, and 0.08 mg/ml for IgG_1, IgG_2, and IgM, respectively), although small precipitant rings were observed. One MC calf and 1 MCS calf were hypogammaglobulinemic (serum IgG + IgM concentration < 5 mg/ml) at 24 hours. Peak mean serum immunoglobulin concentration was not significantly different between calves fed MC and calves fed MCS.

When analyzed by analysis of covariance, using BW and cumulative immunoglobulin intake from maternal colostrum as covariables, hour/treatment interactions were significant for total immunoglobulin, IgG_1, and IgM concentrations (Table 3). Orthogonal polynomials were used to discriminate linear, quad-ratic, or cubic effects of hour within each treatment. In all cases, orthogonal polynomials indicated different serum concentration-time profiles for calves fed MC vs calves fed MCS. Mean serum IgG_1 concentration in calves fed MC followed a cubic pattern, and peaked at 18.70 mg/ml at 24 hours. In calves fed MCS, no effect was significant, indicating that mean serum IgG_1 concentration did not change after peaking at 14.02 mg/ml at 12 hours.

Mean serum IgG_2 concentration in calves fed MC increased in a quadratic manner for 24 hours, then remained unchanged for the next 48 hours. In calves fed MCS, mean serum IgG_2 concentration increased to 1.83 mg/ml at 24 hours, then decreased to 1.45 mg/ml by 72 hours. Quadratic and cubic effects were significant for IgM in calves fed MC, whereas no coefficients were significant in calves fed MCS.

A sex/hour interaction was significant for serum IgM concentration (Table 4). Mean serum IgM concentration in heifers increased to 2.24 mg/ml by 24 hours, whereas mean serum IgM concentration in bull calves did not vary after 12 hours. For each sampling time, however, mean serum IgM concentration was higher in heifers than in bull calves.

Table 3—Least-squares means* of serum immunoglobulin concentration in calves fed MC or MMCS

Isotype and sampling time (h)	Concentration (mg/ml) MC	MCS	SEM
IgG_1†			
12	14.38	14.02	2.15
24	18.70	13.00	1.92
48	14.19	13.50	1.98
72	13.13	13.87	1.95
IgG_2			
12	1.30	1.41	0.20
24	1.68	1.83	0.19
48	1.66	1.56	0.19
72	1.56	1.45	0.19
IgM†			
12	1.55	1.47	0.21
24	2.06	1.45	0.20
48	1.62	1.40	0.20
72	1.49	1.27	0.20
Total†			
12	17.02	16.60	2.36
24	22.60	16.22	2.16
48	17.54	16.32	2.20
72	16.28	15.80	2.19

* Covariately adjusted for body weight at birth and cumulative immunoglobulin intake from maternal colostrum. † Significant hour/treatment interaction ($P < 0.05$).

Table 4—Least-squares means* of serum IgM concentration in heifers and bull calves

Sample time (h)	Concentration (mg/ml) Heifers	Bull calves	SEM
12	1.73	1.30	0.22
24	2.24	1.26	0.20
48	1.72	1.30	0.20
72	1.65	1.11	0.20

* Covariately adjusted for body weight at birth and cumulative IgM intake from maternal colostrum.

Table 5—Least-squares means* of serum immunoglobulin concentration at 28 and 56 days of age in calves fed MC or MCS

Sample time and isotype	Concentration (mg/ml) MC	MCS	SEM
28 d			
IgG_1	10.82	9.22	0.96
IgG_2	1.04[b]	0.90[a]	0.05
IgM	0.56[a]	0.65[b]	0.03
Total	12.40	10.81	1.01
56 d			
IgG_1	8.60	7.52	0.58
IgG_2	1.41	1.37	0.10
IgM	0.75	0.64	0.05
Total	10.77	9.51	0.58

[a,b] Means in the same row with different superscripts differ ($P < 0.06$).
See Table 3 for key.

Table 6—Least-squares means* of serum immunoglobulin concentration at 28 and 56 days of age in heifers and bull calves

Sample time and isotype	Concentration (mg/ml) Heifers	Bull calves	SEM
28 d			
IgG_1	11.33	8.72	0.98
IgG_2	1.08[a]	0.87[b]	0.05
IgM	0.63	0.58	0.03
Total	12.99	10.23	1.00
56 d			
IgG_1	10.85	9.43	0.60
IgG_2	1.41	1.37	0.10
IgM	0.75	0.64	0.05
Total	8.67	7.46	0.59

[a,b] Means in the same row with different superscripts differ ($P < 0.01$).
See Table 3 for key.

Mean serum IgG_2 concentration at 28 days of age tended to be lower and serum IgM concentration tended to be higher in calves fed MCS than in calves fed MC (Table 5); however, the differences were not significant, and there were no differences between groups at 56 days of age. Effects of sex on serum IgG_2 concentration at 28 days of age were significant, and sex tended to influence total serum immunoglobulin concentration and IgG_1 concentration at 28 days of age, but not significantly (Table 6).

Discussion

Serum immunoglobulin concentration-time profiles in calves fed MCS may have been altered because of changes in uptake of immunoglobulin by intestinal epithelium, altered outflow of immunoglobulin from serum into extravascular tissues, or a change in vascular pool size. Michanek et al[17,18] suggested that large amounts of immunoglobulin consumed at a first meal may initiate cessation of macromolecular absorption, thereby reducing efficiency of absorption of subsequent meals. Loss of immunoglobulins from serum once a threshold immunoglobulin concentration is reached, thereby maintaining or lowering serum immunoglobulin concentration, has been reported in mice[19] and human beings,[20] but not in cattle.[13] If such a threshold did exist, the relationship between serum immunoglobulin concentration and immunoglobulin intake should be curvilinear. In this study, regression of the natural logarithm of serum IgG_1 concentration at 24 hours on IgG_1 intake indicated significant linear and quadratic coefficients (log of serum IgG_1 concentration at 24 hours [mg/ml] = $0.777 + 0.174 \times IgG_1$ intake (g) $- 3.38E^{-5} \times IgG_1$ intake^2; $r^2 = 0.49$). On the other hand, serum IgG_2 concentration at 24 hours was not related to IgG_2 intake, and serum IgM concentration was linearly related to IgM intake (log of serum IgM concentration at 24 hours [mg/ml] = $-0.312 + 0.087 \times$ IgM intake (g); $r^2 = 0.38$). Other investigators[5,10] have reported that the relationship between serum IgG concentration and IgG intake is linear within the ranges of immunoglobulin they fed, which suggests that a serum threshold does not exist and that intestinal uptake is not altered when immunoglobulin consumption is increased. The relationship between immunoglobulin intake and absorption is not well understood.

The significant sex/hour interaction on serum IgM concentration (Table 4) was independent of BW, as BW and intake of IgM were included in the model as covariables. Additional research is indicated to corroborate or refute this observation.

At 28 days of age, mean serum IgG_2 concentration tended to be lower in calves fed MCS than in calves fed MC, although differences were not significant. That only IgG_2 concentration at 28 days of age was affected by feeding MCS suggests that immunoglobulin supplementation to maternal colostrum did not markedly influence development of active immunity in calves.[4,12,21]

Provision of additional immunoglobulin by colostral supplementation did not significantly increase mean serum immunoglobulin concentration. However, calves in this study were fed 4 L of colostrum with high immunoglobulin concentration at optimal times (2 and 14 hours after birth). It is not known whether colostral supplements will be of benefit when colostrum is unavailable, when colostrum is of particularly poor quality, or when management practices are not optimal.

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