Effects of Lyophilized Colostrum and Different Colostrum Feeding Regimens on Passive Transfer of Immunoglobulin G in Majorera Goat Kids

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ABSTRACT

Three experiments were conducted including 180 Majorera kids. In the first experiment, the effect of use of lyophilized colostrum vs. frozen colostrum on immunoglobulin G (IgG) blood serum concentration was evaluated. Kids ($n = 40$) received the same management and IgG mass [3368 mg/kg of body weight (BW)] during the colostrum feeding period. The IgG in blood serum of kids from the lyophilized colostrum group was greater than that for kids that received frozen colostrum. The second experiment evaluated the effect of total IgG ingested by kids ($n = 60$) on IgG in blood serum during the colostrum feeding period. Three groups of animals received 3368, 1684, and 842 mg of IgG/kg of BW in 4 feedings for 2 d [high IgG concentration (H-IgG), medium IgG concentration (M-IgG), and low-IgG concentration (L-IgG), respectively]. The IgG blood serum in the kids that received H-IgG was greater than in the other 2 treatment groups, and no statistical differences were found for IgG in blood serum of kids that received either M-IgG or L-IgG. The third experiment evaluated the effect of timing of lyophilized colostrum meals on IgG blood serum concentration. Four groups of kids ($n = 80$) were used. Two groups received 1684 mg of IgG/kg of BW (higher level-1 d and higher level-2 d) and the other 2 groups received 842 mg of IgG/kg of BW (lower level-1 d and lower level-2 d). Two groups received 2 feedings in 1 d, and the other 2 groups received 4 feedings over a 2-d period, as denoted. Higher level-1 d kids had greater IgG blood serum concentration than the higher level-2 d kids, and no statistical differences were found between lower level-1 d and lower level-2 d kids.

(**Key words:** goat kid, colostrum, lyophilized, immunoglobulin G)

Abbreviation key: CAEV = caprine arthritis encephalitis, H-IgG = high concentration (3368 mg/kg of BW) of

IgG, HL-1d = high concentration of IgG in lyophilized paste in 1 d, HL-2d = high concentration of IgG in lyophilized paste in 2 d, L-IgG = low concentration of IgG (842 mg/kg of BW), LL-1d = low concentration of IgG in lyophilized paste in 1 d, LL-2d = low concentration of IgG in lyophilized paste in 2 d, M-IgG = medium concentration of IgG (1684 mg/kg of BW).

INTRODUCTION

To minimize or even annul the mother-kid link, which is established in the first hours after birth (Ramírez et al., 1996) in artificial rearing management, the kids must be removed after parturition. This practice facilitates the acceptance of being bottle-fed, thus improving the adoption of artificial rearing. The latter system is of particular interest in areas where diseases such as caprine arthritis encephalitis (CAEV) are present, as colostrum is one of the direct means of transmission (Guerrault, 1990). To control CAEV, the colostrum has to be pasteurized. This procedure, however, reduces the colostrum IgG concentration (Argüello et al., 2003); therefore, it can be argued that lyophilized colostrum could be more effective, as it maintains a higher IgG concentration. In ruminants, the placenta impedes the transfer of Ig from the dam to the fetus; consequently, the consumption of colostrum by the progeny of these species holds a fundamental role in the acquisition of immunity. Some studies have found lyophilized colostrum to be stable, easy to handle, and suitable for the passive immunization of calves (Klobasa et al., 1998; Husu et al., 1993). Dos Santos et al. (1994) found no relationship between colostral IgG concentration and serum IgG concentration in 1-d-old kids. Chen et al. (1999) observed significant differences in serum IgG concentrations between kids fed colostrum with high or low protein concentration (20 and 10 g/dL). In calves, the IgG colostrum concentration is more important than the quantity of colostrum fed if the total IgG colostrum fed was the same. Stott and Fellah (1983) observed that IgG serum was higher in calves fed 1 L of colostrum with 60 mg/mL of IgG than in those fed 2 L of colostrum with 30 mg/mL of IgG. The relationship between the IgG fed and

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IgG in the calf's blood was linear according to Stott and Fellah (1983). In kids of the Majorera breed, the quantity of IgG consumed presented a high positive correlation with IgG blood concentrations, particularly in the first 72 h of the animal's life, but this correlation diminished significantly at 84 h (Argüello et al., 2004b).

No literature has been found related to the use of lyophilized goat colostrum in kids. The present experiments will be the first to approach the use of this technology in goat kids and will try to explain how the principal effects (IgG colostrum concentration, timing of lyophilized colostrum meals, and amount of IgG fed) will affect the IgG serum blood concentration in kids.

The objectives of this experiment were to determine the effect of lyophilized colostrum fed, the effect of total IgG fed to the kids, and the effect of the timing of lyophilized colostrum meals on IgG blood serum during the colostrum feeding period.

MATERIALS AND METHODS

Three different experiments were designed. A colostrum pool was made from fresh goat colostrum (16.84 mg of IgG/mL of colostrum), 4 L of which were frozen (-20°C) and 30 L were lyophilized, producing a colostrum powder with an IgG concentration of 61.041 mg of IgG/g. This pool was used to feed the kids in the 3 experiments. Lyophilized colostrum pastes were prepared by mixing lyophilized colostrum powder plus sterilized hot water (40°C) in accordance with the method used by Argüello et al. (2003). Majorera kids were used for these experiments. The Majorera breed is a dairy goat with an average milk production of 546 L/210 d. The Majorera goat kids have an average birth weight of 3.50 kg for males and 3.15 kg for females (Capote et al., 1992). The management of the kids before starting the experiments was the same. Immediately after birth, they were separated from their mothers and dried. Their umbilical cords were disinfected, and they were weighed, identified, and randomly distributed into groups with the same number of males and females. Frozen colostrum or lyophilized colostrum paste was administered by bottle-feeding at 2, 14, 26, and 38 h postpartum depending on the experiment.

In the first experiment, the effect of feeding lyophilized colostrum vs. frozen colostrum on the IgG kid serum was evaluated. The 40 Majorera kids (20 males and 20 females) were randomly assigned to 2 groups. The frozen colostrum group and the lyophilized group were fed with their respective form of colostrum 2 $\times$ /d for 2 d. Each feeding in both groups contained 842 mg of IgG/kg of BW according to the management system proposed by Argüello et al. (2004c). The total IgG received by both groups was 3368 mg of IgG/kg of BW. Each kid from

the frozen colostrum group received 50 mL of thawed colostrum/kg of BW per feeding. The frozen colostrum was thawed at room temperature and after that was warmed to 40°C (internal temperature) in microwave ovens (rotary microwave) in accordance with the system used by Argüello et al. (2003). The IgG concentration of the lyophilized colostrum paste was 22.88 mg of IgG/g. Each kid in the lyophilized colostrum group received 36.80 g of lyophilized colostrum paste/kg of BW per feeding (Table 1).

In the second experiment, the effect of the total IgG received/kg of BW on IgG kid serum was evaluated. Sixty Majorera kids (30 males and 30 females) were randomly assigned to 3 groups. All groups were fed with lyophilized colostrum paste 2 $\times$ /d for 2 d. The first group (high IgG concentration; **H-IgG**) was fed using a lyophilized colostrum paste with an IgG concentration of 22.88 mg of IgG/g; each kid received 36.80 g of lyophilized colostrum paste/kg of BW per feeding, representing 842 mg of IgG per feeding per kg of BW. The total IgG received by the H-IgG kids was 3368 mg/kg of BW. The medium IgG concentration group (**M-IgG**) was fed using lyophilized colostrum paste with an IgG concentration of 11.44 mg of IgG/g, and each kid from this group received 36.80 g of lyophilized colostrum paste/kg of BW per feeding, representing 421 mg of IgG per feeding/kg of BW. The total IgG received by the kids in the M-IgG group was 1684 mg/kg of BW. The final group (low concentration of IgG; **L-IgG**) was fed twice using lyophilized colostrum paste with an IgG concentration of 5.72 mg of IgG/g. Each kid in this group received 36.80 g of lyophilized colostrum paste/kg of BW per feeding, representing 210.5 mg of IgG per feeding/kg of BW. The total IgG received by kids in the L-IgG concentration group was 842 mg/kg of BW.

In the third experiment, the effect of the timing of lyophilized colostrum meals per day on IgG kid serum was evaluated at 2 IgG concentrations/kg of BW. Eighty Majorera kids (40 males and 40 females) were randomly assigned into 4 groups. The first group received lyophilized colostrum paste 2 $\times$ /d during 1 d; this group was assigned as **HL-1d** (high concentration, 1 d). The IgG concentration of the lyophilized colostrum paste was 22.88 mg of IgG/g. Each HL-1d kid received 36.80 g of lyophilized colostrum paste/kg of BW per feeding, representing 842 mg of IgG per feeding/kg of BW. The total IgG received by HL-1d kids was 1684 mg/kg of BW. The second high IgG concentration group received lyophilized colostrum paste 2 $\times$ /d during 2 d; this group was assigned as **HL-2d** (high IgG concentration, 2 d). The IgG concentration of the lyophilized colostrum paste was 11.44 mg of IgG/g. Each HL-2d kid received 36.80 g of lyophilized colostrum paste/kg of BW per feeding, representing 421 mg of IgG per feeding/kg of BW. The total

Table 1. Management and IgG amount received by kids receiving frozen and lyophilized colostrum.¹

	Experiment 1		Experiment 2			Experiment 3			
	FG	LG	H-IgG	M-IgG	L-IgG	HL-1d	HL-2d	LL-1d	LL-2d
Colostrum fed, no. of days	2	2	2	2	2	1	2	1	2
Feedings per d	2	2	2	2	2	2	2	2	2
Total feedings	4	4	4	4	4	2	4	2	4
IgG, mg/kg of BW per feeding	842	842	842	421	210.5	842	421	421	210.5
Total IgG fed, mg/kg of BW	3368	3368	3368	1684	842	1684	1684	842	842

¹FG = Frozen colostrum group, LG = lyophilized colostrum group, HL-1d = high concentration of IgG in lyophilized paste in 1 d, HL-2d = high concentration of IgG in lyophilized paste in 2 d, LL-1d = low concentration of IgG in lyophilized paste in 1 d, LL-2d = low concentration of IgG in lyophilized paste in 2 d, H-IgG = high concentration of IgG in lyophilized paste, M-IgG = medium concentration of IgG in lyophilized paste, and L-IgG = low concentration of IgG in lyophilized paste.

IgG received by the HL-2d kids was 1684 mg/kg of BW. The third group (low IgG concentration) received lyophilized colostrum paste 2×/d in 1 d; this group was assigned as **LL-1d** (low IgG concentration during 1 d). The IgG concentration on lyophilized colostrum paste was 11.44 mg of IgG/g. Each LL-1d kid received 36.80 g of lyophilized colostrum paste/kg of BW per feeding, representing 421 mg of IgG per feeding per kg of BW. The total IgG received by the LL-1d kids was 842 mg/kg of BW. The fourth group (also low IgG concentration) received lyophilized colostrum paste 2×/d during 2 d; this group was assigned as **LL-2d** (low IgG concentration, 2 d). The IgG concentration of the lyophilized colostrum paste was 5.72 mg of IgG/g. Each LL-2d kid received 36.80 g of lyophilized colostrum paste/kg of BW per feeding, representing 210.5 mg of IgG per feeding/kg of BW. Total IgG received by LL-2d kids was 842 mg/kg of BW.

The kids had no access to fresh water until 15 d of life.

To lyophilize goat colostrum, 500 mL of goat colostrum was frozen at −80°C for 2 d. Subsequently, bottles containing frozen colostrum were connected to the lyophilizer (Telstar Cryodos-80) for 5 d. The vacuum level was 7.9×10^{-2} mBa, and the temperature was −55°C.

Samples from the jugular vein were taken every 12 h from birth to 108 h postpartum to determine the IgG concentration in the kids' blood. The blood was introduced into a clotting tube and was centrifuged; the serum obtained was frozen at −20°C until analysis. The IgG quantification was made using radial immunodiffusion in accordance with the method used by Mancini et al. (1965). The agar was prepared by adding 1 L of glycine buffer [pH 7; 7.5 g of glycine, 100 mL of EDTA (0.4 M) in distilled water and adjusting the volume to 1 L] to 20 g of agar. This suspension was placed in a boiling water-bath and stirred until the agar had dissolved, after which it was cooled to 60°C. The antiserum (previously made by the inoculation of rabbits with goat IgG) or a suitable dilution of it made in a glycine buffer was heated to 55°C, after which both solutions were mixed thoroughly, avoiding bubbling, with the aid of a pipette preheated

to 60°C. The antiserum-agar mixture was immediately poured into the petri dish with a final agar thickness of 0.3 cm. The petri dish was sealed and stored at 4°C until use. Circular wells were punched in the gel, using a 3-mm-bore needle. The small cylinders of gel cut out by the needle were removed by suction. Each well received 10 µL of antigen solution (standard curve) or blood serum; the samples were not diluted before the assay. The standard curve was prepared in accordance with Catty and Raykundalia (1988) using a goat IgG solution (50 mg/mL).

Hematocrit values were measured in accordance with the procedure of Kume and Tanabe (1993) to evaluate fluid intake effects (50.23, 49.65, 51.43, 51.67, 48.21, 49.89, 50.11, 49.03, and 48.98% for frozen colostrum, lyophilized colostrum, H-IgG, M-IgG, L-IgG, HL-1d, HL-2d, LL-1d, and LL-2d kids, respectively). No statistical differences were observed between groups.

From d 3 until 5, the kids were fed by milk replacer machine (Mini Robot, Divasa Farmavic, Sociedad Anónima) in accordance with Argüello et al., (2004a). Table 1 summarizes the group management systems.

All variables were normally distributed. The statistical analyses used to evaluate the effects of lyophilized colostrum feeding on IgG blood serum in kids was a GLM procedure with repeated measures between the frozen colostrum group and the lyophilized colostrum group. The effect of the IgG fed to kids during the colostrum feeding period on the IgG blood serum was evaluated using a GLM procedure with repeated measures using H-IgG, M-IgG, and L-IgG groups, and a posthoc Tukey analysis was performed. Finally, the effect of the timing of lyophilized colostrum meals was evaluated using a GLM procedure with repeated measures between HL-1d and HL-2d groups at high IgG concentration and between LL-1d and LL-2d at low IgG concentration (SPSS, 1988).

RESULTS AND DISCUSSION

None of the kids studied in any experiment displayed any detectable concentrations of IgG (the limit of detec-

Table 2. Immunoglobulin G (least squares means) in blood serum of control group and lyophilized group kids.

Time, h	FG kids ¹	LG kids ¹	SEM	<i>P</i>
0	0	0	0	0.009
12	5.52	8.82	0.83	
24	5.11	9.72	1.25	
36	5.44	10.07	1.09	
48	4.87	7.87	0.65	
60	6.09	8.85	0.95	
72	5.93	8.24	0.63	
84	4.32	7.30	0.66	
96	5.38	7.06	0.59	
108	5.46	5.79	0.59	

¹FG = Frozen colostrum group; LG = lyophilized colostrum group.

tion by radial immunodiffusion was <0.11 to 0.12 mg/mL in the blood at birth (Tables 2, 3, and 4) in accordance with Constant et al. (1994) and Argüello et al. (2004c). Table 2 shows the results of the effect of feeding lyophilized colostrum vs. frozen colostrum (experiment 1). According to Table 1, the management and IgG amount received by frozen colostrum group kids and lyophilized colostrum group kids were the same, but the blood serum IgG concentration in both groups was statistically different ($P < 0.009$). At 12 and 108 h after birth, the IgG blood serum concentration was closer between groups, but at 36 h postpartum, lyophilized group kids had 85% more IgG blood serum concentration than did frozen colostrum group kids (5.44 and 10.07 mg/mL in the frozen and lyophilized colostrum group kids, respectively). Contrary to the effect observed in Table 2, Klobasa et al. (1998) observed that immunoglobulins were absorbed with equal efficiency from frozen and lyophilized colostrum in newborn calves. The main difference between the 2 results was that, according to Klobasa et al. (1998), the lyophilized colostrum had the same IgG concentration as the frozen colostrum, but in the

present study, the lyophilized colostrum paste had an IgG concentration higher than frozen colostrum (16.84 and 22.88 mg/mL of IgG for frozen and lyophilized paste colostrum, respectively). This may be the reason for the differences observed between experiments, and according to Muller and Ellinger (1981) or Stott and Fellah (1983), who reported that when compared on equal IgG mass, the amount of colostrum fed had less influence on IgG absorption than the IgG concentration. Thus colostrum, when correctly preserved by lyophilizing, can provide protection for kids.

Table 3 shows the effect of total IgG consumed by kids on IgG blood serum during the colostrum feeding period. The IgG blood serum in H-IgG kids was statistically higher than that in M-IgG and L-IgG kids. No statistical differences were found for IgG blood serum between M-IgG and L-IgG kids, although L-IgG kids showed numerically lower IgG blood serum at all sample times. These results are in accordance with those of Abel Francisco and Quigley (1993) in calves or Dos Santos et al. (1994) working with Brazilian kids. On the contrary to that observed in the present experiment, Al-Jawad and Lees (1985) did not find statistical differences between lambs that received artificial colostrum with high (139 mg/mL) or low (38 mg/mL) IgG concentration.

Experiment 3 evaluated the effect of timing of lyophilized colostrum meals on IgG blood serum in new born kids at 2 different IgG concentrations. The HL-1d treatment presented statistically higher IgG blood serum concentrations than the HL-2d treatment at all sample times tested (Table 4). The amount of lyophilized colostrum paste was the same in both groups; consequently, the IgG concentration was higher in the HL-1d group. No statistical differences were found between LL-1d and LL-2d treatments, but LL-2d kids presented numerically lower IgG blood serum concentration at all sample times. Similar results are reported by Chen et al. (1999) in Nubian kids. The SEM values were very high and might have impeded the detection of statistical differences. High variability in IgG blood serum values have been previously reported in calves (Klobasa et al., 1988). Some artificial colostrum did not permit the immunoglobulin passive transfer (Argüello et al., 2003), which could explain the results related by Al-Jawad and Lees (1985). The feeding of colostrum in 1 d as opposed to 2 d gave better results when an equal IgG mass was used, and handling time was also reduced. In Spain, the standard practice is to have a 2-d colostrum feeding period, and the results shown in experiment 3 are useful for improving the management in dairy goat farms.

CONCLUSIONS

The IgG from lyophilized colostrum paste can be absorbed more efficiently than that from frozen colostrum.

Table 3. Immunoglobulin G (least squares means) blood serum in kids fed high, medium, and low concentrations¹ of IgG from lyophilized paste.

Time	H-IgG	M-IgG	L-IgG	SEM	<i>P</i>
0	0	0	0	0	0.001
12	9.02 ^a	4.03 ^b	1.55 ^b	0.36	
24	9.53 ^b	4.69 ^b	3.26 ^b	0.36	
36	10.28 ^a	5.63 ^b	4.54 ^b	0.51	
48	8.85 ^a	4.42 ^b	3.65 ^b	0.34	
60	8.43 ^a	3.99 ^b	2.81 ^b	0.27	
72	8.14 ^a	3.63 ^b	2.39 ^b	0.23	
84	7.50 ^a	3.24 ^b	2.02 ^b	0.27	
96	7.23 ^a	3.26 ^b	2.34 ^b	0.27	
108	6.79 ^a	3.42 ^b	2.26 ^b	0.24	

^{a,b}Values with different superscripts differ at $P \leq 0.05$.

¹H-IgG = High concentration of IgG in lyophilized paste, M-IgG = medium concentration of IgG in lyophilized paste, and L-IgG = low concentration of IgG in lyophilized paste.

Table 4. Immunoglobulin G blood serum (least squares means) in kids fed high and low IgG concentrations.¹

Time	High IgG concentration				Low IgG concentration			
	HL-1d	HL-2d	SEM	P	LL-1d	LL-2d	SEM	P
0	0	0	0	0.011	0	0	0	0.252
12	7.73	3.55	0.93		3.28	1.22	0.24	
24	6.96	4.49	0.85		3.86	3.49	0.38	
36	9.01	4.83	1.01		3.68	3.29	0.62	
48	9.43	4.43	1.21		3.37	3.17	0.36	
60	8.32	4.27	1.06		3.43	2.70	0.22	
72	5.93	3.84	0.41		3.30	2.49	0.20	
84	5.92	3.30	0.52		3.39	2.13	0.33	
96	5.82	3.45	0.49		3.62	2.74	0.34	
108	6.02	3.52	0.53		3.53	2.06	0.28	

¹HL-1d = High concentration of IgG in lyophilized paste in 1 d, HL-2d = high concentration of IgG in lyophilized paste in 2 d, LL-1d = low concentration of IgG in lyophilized paste in 1 d, and LL-2d = low concentration of IgG in lyophilized paste in 2 d.

The amount of IgG intake by goat kids was clearly shown in the IgG blood serum concentration. If the same IgG mass is used, it is more expedient to feed the colostrum in 1 d rather than 2. Lyophilizing colostrum is a suitable means of creating a colostrum bank with intact immunological qualities. Lyophilized colostrum powder is easy to transport, requires no special conditions for prolonged storage, and could be used to provide newborn kids with immunoglobulins. When using lyophilized vs. fresh/frozen colostrum the desired immunoglobulin mass should be fed on d 1 rather than over a 2-d period.

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