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# Transference of passive immunity and growth in dairy calves born to dams with high or low somatic cell counts at dry-off and fed colostrum from cows with high or low somatic cell counts at dry-off

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## ABSTRACT

The purpose of this experiment was to evaluate the transference of passive immunity (TPI) and growth achieved by calves born to dams with low or high SCC at dry-off and fed with colostrum from cows with low or high SCC at dry-off. Forty multiparous (3.2 lactations; SD = 1.1), dry, and pregnant Holstein cows were used. The cows were separated into 2 groups based on the SCC in the last 3 monthly records before dry-off. An SCC of 200,000 cells/mL was used as the cut-off point to categorize cows with or without mastitis at dry-off, and 2 groups of 20 cows each were formed: L-cow cows (last 3 SCC before dry-off less than 200,000 cells/mL) and H-cow cows (last 3 SCC before dry-off greater than 200,000 cells/mL). At calving, 40 calves were obtained (20 calves born to L-cow cows [L-calf], and 20 calves born to H-cow cows [H-calf]; females = 21 and males = 19), and 40 colostrum units (20 from L-cow cows [L-col]; and 20 from H-cow cows [H-col]). The experimental design was a  $2 \times 2$  factorial, with 2 factors and 2 levels within each factor (type of calf: L-calf and H-calf, and type of colostrum: L-col and H-col), determining 4 treatments (n = 10 per treatment): L-calfxL-col (L-calf fed with L-col); L-calfxH-col (L-calf fed with H-col); H-calfxL-col (H-calf fed with L-col); and H-calfxH-col (H-calf fed with H-col). Male and female calves were homogeneously distributed within each treatment. All calves received 4 L of colostrum, L-col or H-col depending on the assigned treatment, with an oro-esophageal tube within 3 h after birth. Yield, chemical composition and IgG were determined. The TPI and the apparent efficiency of IgG absorption (AEA) were also determined. Nutrient intake and body growth and development traits

of the calves (BW, heart girth, and withers height) were determined in the first 30 d of life. The colostrum produced by L-cow presented a lower SCC compared with H-cow. Colostrum protein yield of L-cow was 0.21 kg higher than H-cow, and colostrum of L-cow had 24% higher IgG concentration. The TPI was not affected by the interaction calf type  $\times$  colostrum type, and there was no effect of the colostrum type on the level TPI and AEA achieved by calves. However, an effect of calf type on TPI and AEA achieved was observed, as L-calf achieved greater TPI than H-calf (28.8 and 22.8 g IgG/L, respectively; SEM = 1.5), and L-calf presented a higher AEA than H-calf (30.0% and 24.5%, respectively; SEM = 1.4). The BW, heart girth, and withers height were not affected by calf type, colostrum type, or by the interaction calf type  $\times$  colostrum type. We concluded that cows with high SCC at dry-off produced colostrum with higher SCC and lower IgG concentrations, but when ingested by calves it did not affect TPI, feed intake, growth, or development. Calves born to cows with high SCC at dry-off presented a lower AEA of IgG, which translated into a lower serum concentration of IgG, irrespective of type of colostrum that was fed.

**Key words:** mastitis, colostrum quality, apparent efficiency of IgG absorption, calf rearing

## **INTRODUCTION**

The syndesmochorial placenta of the cow does not allow for maternal-fetal blood exchange, which prevents the transfer of protective immunoglobulins to the fetus during gestation (Arthur et al., 1996). Calves need to absorb immunoglobulin from colostrum to acquire protection against diseases in the first weeks of life (Butler, 1983; McGuirk and Collins, 2004). The absorption of IgG from colostrum in the small intestine of the calf (transference of passive immunity, **TPI**; Godden, 2008) is considered proper when the blood concentration of

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-25. Nonstandard abbreviations are available in the Notes.

IgG is higher than 10 g/dL, equivalent to 5.1 g/L of total serum proteins or 8.1% Brix (Lombard et al., 2020).

One of the factors that most affect the TPI is the quality of the colostrum, which is closely linked to its IgG concentration. Colostrum is considered of good quality if it has an IgG concentration of at least 50 g/L (Godden, 2008). In turn, different factors can affect the concentration of IgG in colostrum, for example breed, age, parity, and dry period length (Muller and Ellinger, 1981; Rastani et al., 2005; Phipps et al., 2017). Another factor that affects colostrum quality is an inflammation of the mammary gland, which several authors have reported to affect its concentration of IgG and have described a negative relationship between SCC and colostrum quality (Maunsell et al., 1998; Gulliksen et al., 2008; Puppel et al., 2020).

The SCC can be used as a reliable indicator of mastitis (Ruegg and Reinemann, 2002) and it is widely accepted that cows with an SCC greater than 200,000 cells/mL can be considered to have subclinical mastitis (Dohoo and Leslie, 1991; Djabri et al., 2002; Pantoja et al., 2009b). In this sense, Niemi et al. (2021) reported that recurrently infected cows, which maintained persistently high SCC during the previous lactation, are more predisposed to present mastitis at the beginning of the following lactation. Likewise, cows with mastitis at the next calving and even in early lactation, which may affect colostrogenesis (Newman et al., 2010).

Although there is no consensus among the different authors on the cut-off point for SCC in colostrum to consider it of good or poor quality, it is clearly reported that the SCC in the first milking of colostrum can be an indicator of colostrum quality (Puppel et al., 2020). For example, an increase in SCC has been related to a deterioration of the protein quality of milk and colostrum in dairy cows, mainly because of proteolysis (Forsbäck et al., 2010), which may explain why some authors reported that colostrums at the first milking with SCC greater than 400,000 cells/mL presented a lower IgG concentration (Morrill et al., 2012; Puppel et al., 2020). Another study that used the SCC of colostrum as an indicator of mastitis observed that SCC greater than 50,000 cells/mL in first-milking colostrum was the only factor that was significantly associated with the production of low-quality colostrum (Gulliksen et al., 2008). Also, Kehoe et al. (2007) reported that cows from dairy farms with an average SCC in bulk tank milk greater than 200,000 cells/ mL had a lower concentration of IgG in the colostrum compared with dairy farms with SCC lower than 200,000 cells/mL. Theoretically, the ingestion of colostrum with a high SCC may affect the level of TPI achieved by the newborn calf through a decrease in the IgG concentration of the colostrum, due to proteolysis that reduces the 
 Table 1. Ingredients and nutrients composition of the diets offered to dry and prepartum cows throughout the experimental period

Item	Dry cow	Prepartum cow
DM (% as fed)	$55.8 \pm 1.5$	$49.3 \pm 1.2$
Composition (% of DM)		
CP	$15.1 \pm 0.8$	$15.6 \pm 0.4$
NDF	$53.1 \pm 2.1$	$46.3 \pm 1.8$
ADF	$34.8\pm0.9$	$31.4 \pm 0.7$
NE <sub>L</sub> (Mcal/kg of DM)	$1.53\pm0.1$	$1.54 \pm 0.1$
DCAD (mEq/100 g of DM)		$-9.7 \pm 07$
Ingredients of diet (% of DM)		
Corn silage	36	47
Solvent-extracted soybean meal	10	20
Corn dry distiller grain + solvent	23	
Wheat straw	31	23
Corn grain, ground, dry		8
Vitamin and mineral premix <sup>1</sup>	0.1	0.1
Magnesium oxide		0.3
Anionic salt premix <sup>2</sup>		2.3
Total DM offered (kg/d/cow)	12	15

<sup>1</sup>Provided (per kg of DM): 0.85 g of Cu, 2.6 g of Zn, 0.9 g of Se, 1.0 g of Mn, 23 mg of I, 3 mg of Co, 63,700 IU of vitamin A, 14,700 IU of vitamin D, and 1,250 IU of vitamin E.

<sup>2</sup>Anionic salt premix (magnesium oxide, calcium chloride, magnesium chloride, magnesium sulfate).

concentration of IgG (Forsbäck et al., 2010), an interference in the intestinal absorption of IgG due to high levels of bacteria in the colostrum, that may bind free IgG in the gut lumen or directly block uptake and transport of Ig molecules across intestinal epithelial cells (James et al., 1981), or both mechanisms. However, results in this area are scarce and unclear. In this sense, Ferdowsi Nia et al. (2010) observed a trend that the higher the SCC in colostrum, the lower the serum concentration of IgG in calves 3 h after colostrum ingestion. However, Leite et al. (2017) did not observe differences in the TPI achieved by calves that consumed colostrum from cows with or without a subclinical mammary infection at calving.

There is evidence in dairy cattle that in utero exposure to several stresses (e.g., nutritional, metabolic, infectious) during the last stages of gestation could lead to changes in the structure and function of tissues that may have consequences on physiology and susceptibility to disease in the progeny, which has been described as evidence of developmental or fetal programming (Abuelo, 2020). Recently, Antanaitis et al. (2022) observed that cows with subclinical mastitis during late gestation had an increased risk of stillbirth in the first 24 h than healthy cows. Even if it could be expected that calves born to cows that suffered mammary infections during pregnancy could have a lower efficiency of IgG absorption at the intestinal level and a compromised TPI, no studies have been found. Santos et al. (2013) observed that calves born and fed with colostrum from healthy cows had a higher level of TPI and serum transferrin concentrations, and lower serum haptoglobin concentrations than calves

born and fed with colostrum of cows with subclinical or clinical mastitis, although this study did not determine the efficiency of IgG absorption. This suggests that those calves were undergoing an infectious and inflammatory process already at 24 h of life (Eckersall and Conner, 1988; Gruys et al., 2005).

Although SCC may have negative effects on TPI, as far as we know, no attempts have been made to isolate the effects of high SCC at dry-off and high SCC in the colostrum on the efficiency of IgG absorption, TPI, and development of dairy calves. We hypothesize that dairy cows with high SCC at dry-off will affect TPI in neonatal calves either through producing colostrum with a lower concentration of IgG, through a deleterious effect on innate IgG absorption capacity of the calf during late gestation, or both. The aim of our work was to evaluate the TPI and growth achieved by calves born to dams with low or high SCC at dry-off, and fed with colostrum from cows with low or high SCC at dry-off.

### **MATERIALS AND METHODS**

The experiment was conducted in accordance with the recommendations on the use of animals in experimentation, education and research established by the Honorary Commission for Animal Experimentation of the University of the Republic (Uruguay; protocol: CEUA-FVET No. 845/19 Exp 111900–000326–19) and was carried out at the Experimental Station No. 2 of the Veterinary School, San José, Uruguay.

## Animals and Experimental Design

From a group of 60 preselected cows, 40 multiparous (3.2 lactations; SD = 1.1), dry and pregnant Holstein cows, with a BW of 586 kg (SD = 82) were used. The cows were separated into 2 groups based on the SCC in the last 3 monthly records before dry-off. An SCC of 200,000 cells/mL was used as the cut-off point, which has been previously used to categorize cows with or without mastitis at the day of milk control (Rajala-Schultz et al., 2011; Vásquez et al. al., 2018; Rowe et al., 2020) and even to study the infection dynamics during the dry period (Pantoja et al., 2009; Madouasse et al., 2010; Lipkens et al., 2019a,b). Only cows and calves that did not present any health problems or complications at birth were used.

In this way, 2 groups of 20 cows each were formed. A first group with cows that presented in each of the last 3 SCC before dry-off less than 200,000 cells/mL (mean = 117,000 cells/mL; range: 45,000 to 179,000 cells/mL; L-cow) and a second group with cows that presented in each of the last 3 SCC before dry-off greater than 200,000 cells/mL (mean 509,000 cells/mL; range: 308,000 to

833,000 cells/mL; **H-cow**). The cows in the L-cow group did not present any case of clinical mastitis in the previous lactation, and the cows in the H-cow group presented at least 1 event of clinical mastitis and at least 1 positive bacteriological culture for some of the most prevalent causal pathogens of mastitis (*Staphylococcus aureus, Streptococcus dysgalactiae, Streptococcus uberis,* CNS, and *Escherichia coli*) in the previous lactation.

At calving of the cows, 40 calves were obtained (20 calves born to L-cow cows [L-calf]; and 20 calves born to H-cow cows [H-calf]; females = 21 and males = 19), and 40 colostrum units (20 colostrum from L-cow cows [L-col]; and 20 colostrum from H-cow cows [H-col]). The experimental design was a  $2 \times 2$  factorial, with 2 factors and 2 levels each (type of calf: L-calf and H-calf, and type of colostrum: L-col and H-col, determining 4 treatments (n = 10 per treatment): L-calfxL-col (L-calf fed with L-col); L-calfxH-col (L-calf fed with H-col); H-calfxL-col (H-calf fed with L-col); and H-calfxH-col (H-calf fed with H-col). Male and female calves were homogeneously distributed within each treatment, and the ratio male: female in each treatment was 5:5, 5:5, 5:5, and 4:6 for L-calfxL-col, L-calfxH-col, H-calfxL-col, and HcalfxH-col, respectively. There were no losses of calves in any treatment during the experiment. Each cow was considered as the experimental unit for type of colostrum, and each calf was considered as the experimental unit for the combination of type of colostrum  $\times$  type of calf. The sample size was calculated based on a previous study (Ferrando, 2023), requiring 8 calves per treatment to detect a difference of 5 g of IgG/L with  $\alpha = 0.05$  and  $1 - \beta = 0.80$ , and with a SD of 4.0 g IgG/L. An additional 2 calves were included per group to account for possible losses unrelated to the experiment.

The selected cows were dried-off 60 d before the expected calving date and all cows were managed in the same way until calving (actual duration of the dry period was 62.5 d; SD = 15.0). From 60 to 20 d before calving, the cows were fed with a diet formulated to fulfill the requirements of dry cows with 210 d of gestation, and 20 d before expected calving, the cows were admitted to the precalving pen, where the diet was changed to fulfill their requirements. Both diets were formulated as suggested by the NRC (2001; Table 1). Throughout the period, the cows had adequate environmental conditions, with access to shade, water, and without mud.

All cows were immunized against rotavirus, coronavirus, and *E. coli* K99, applying a single dose of vaccine (Rotavec Corona, MSD-Animal Health, Burgwedel Biotech GmbH, Burgwedel, Germany) at dry-off. Likewise, the cows were immunized against the pneumoenteric complex (bovine herpesvirus type 1 and type 5, bovine viral diarrhea virus type 1 and type 2, bovine parainfluenza virus type 3, bovine respiratory syncytial virus,

 Table 2. Nutrients composition of feeds used to feed calves of all treatments throughout the first 30 d of life

Item	Whole milk	Starter concentrate		
DM, (%)	14.5	87.5		
Fat, % of DM	31.4	2.0		
Protein, % of DM	25.6	18.0		
Lactose, % of DM	32.7			
CF, % of DM		6.0		
ME, (Mcal/kg of DM)	5.3	3.1		

Pasteurella haemolytica, Pasteurella multocida, E. coli K99, E. coli J5, and Salmonella dublin) with 2 doses of the Neumosan V4J5 vaccine (Virbac–Santa Elena, Montevideo, Uruguay), one dose at the dry-off and the second dose upon admission to prepartum. Also, at the dry-off after the last milking, antibiotic therapy was given to cure and prevent mastitis during the dry period with NafpenzalDC (MSD Animal Health–Intervet International B.V. De Bilt, the Netherlands) and internal teat sealing was performed with Mastblock (Hipra-Animal Health, Porto Alegre, Brazil).

Prepartum cows were monitored for 24 h to provide care during calving if necessary. Immediately after calving, cows were taken to the milking parlor, where all the colostrum produced in the first milking was collected within 3 h by mechanical milking. The colostrum was classified according to the type of cow in L-col or H-col and stored individually at  $-20^{\circ}$ C until use.

Immediately after birth, calves were removed from their dams to prevent them suckling colostrum directly from the udders of their dams. They were taken to the nursery where received neonatal care and within each calf type were fed with the corresponding colostrum according to the assigned treatment. No calf was fed with its own dam's colostrum. The only characteristic used to typify the colostrums was the SCC of the producing cow in the last 3 controls before drying off. Subsequently, the calves were housed in individual pens of 2  $\times$  1 m, with a cement floor with a wooden grid raised 10 cm above the ground, with individual feeders and drinkers available. The pens were located inside a barn, equipped with curtains to control exposure to sunlight and air currents. Each calf was randomly allocated to a pen. The calves remained in these installations during the entire measurement period, which lasted 30 d (from birth to 30 d of age).

Within 3 h of life (the time between birth and colostrum dosing was 132.5, 137.5, 133.6, and 134.1 min; SEM = 5.19 for H-calfxH-col, H-calfxL-col, L-calfxHcol, and L-calfxL-col, respectively) all calves received 4 L of colostrum in a single dose with an oral-esophageal tube, because it ensured that all calves received the same amount of colostrum, at a similar temperature and time, to isolate the effect of colostrum composition as the only variable that influenced the TPI process.

The colostrum was thawed in a water bath ( $38^{\circ}$ C for ~1 h). From 12 h after calving until d 30 of life, they were fed with whole milk (Table 2) produced by the cows of the herd of the experimental farm, and the amount was equivalent to 15% of the BW at calving (4.92 L; SD = 0.07), divided into 2 equal daily feedings (0700 and 1800 h), which were offered in individual buckets provided with teats. This quantity was maintained from 1 to 30 d. From 2 d of life a starter concentrate was offered ad libitum in individual feeders (Table 2), and calves had water ad libitum throughout the day. Only the first author was aware of the allocation of the treatments to the experimental units at the different stages of the study.

## Sampling and Feed Analysis

Once a week, samples of the feeds used during the dry period and the prepartum were taken. The starter concentrate used during the experimental period was sampled once a week throughout the experimental period. The samples were dried in a forced-air oven at 60°C for 48 h and ground to pass through a 1-mm Wiley mill screen (Arthur H. Thomas Co., Philadelphia, PA). Feed samples were analyzed for DM, ash, total N, and ether extract (AOAC, 1990; methods 934.01, 942.05, 955.04, and 920.39, respectively); NDF using heat-stable  $\alpha$ -amylase and sodium sulfite, and ADF (Van Soest et al., 1991). To calculate the concentration of ME of the feeds, the methodology proposed by the NRC (2001) was used, using information of the chemical composition of each feed.

To determine the chemical composition of the milk ingested by the calves, 2 milk samples were taken per day, 1 sample from each feeding session (0700 and 1800 h). The milk samples were collected in containers using bronopol (Lactopol, Montevideo, Uruguay) as preservative agent; the samples were used to determine fat, protein, lactose, and TS by mid-infrared spectrometry (Bentley 2000; Bentley Instruments), using method ISO 9622:2013 (ISO, 2013). The ME of milk was calculated as suggested by the NRC (2001) using the following equation:

ME (Mcal/kg of DM of milk) =  $[(0.057 \times \% \text{ protein} + 0.092 \times \% \text{ fat} + 0.0395 \times \% \text{ lactose}) \times 0.97] \times 0.96.$ 

## **Colostrum Sampling and Analysis**

Within 3 h postpartum, each cow was milked completely, and the colostrum was collected in a stainless steel container. Colostrum production from the first milking was recorded using DemaTron70 electronic lactometers (Gea Farms Technologies. Düsseldorf, Germany).

Each colostrum sample obtained was gently shaken for 1 min to homogenize its components. From each colostrum produced by each cow, 2 samples were taken. One sample was taken in 50-mL vials, using bronopol (Lactopol, Montevideo, Uruguay) as preservative agent, and within 48 h, it was sent to the laboratory to determine the chemical composition (fat, protein, lactose, TS) and SCC. The second sample was taken in sterile 10-mL vials and stored at  $-20^{\circ}$ C until analyzed for IgG concentration.

The determination of the IgG concentration in the colostrum samples was carried out through the radial immunodiffusion (RID) technique, using commercial kits (Triple J Farms, Bellingham, WA). Frozen colostrum samples were thawed at room temperature on the day of the determination. Once thawed, the samples were shaken for 1 min to homogenize the components and then diluted with deionized water at a 1:3 dilution ratio for subsequent analysis (Barry et al., 2019). In each RID plate, 3 of the 24 wells were filled with reference sera (1.80, 14.02, and 28.03 g/L of IgG for low, medium, and high controls respectively). The interassay CV was 9.2%, 7.6%, and 10.4% for the low, medium, and high controls, respectively. The plates were incubated at room temperature (20-24°C) for 24 h. The diameters of the precipitate rings were measured using a digital caliper to a precision of 0.01 mm. Samples that produced a value outside the range of the reference curve were further diluted and retested.

The determination of the chemical composition of the colostrum was carried out by the method described for the analysis of the chemical composition of milk. The colostrum SCC was determined by flow cytometry, using a Somacount 300-unit (Bentley Instruments), according to the ISO 13366-2:2006 (ISO, 2006) method.

## Serum Sampling and Analysis

Transfer of passive immunity was determined by IgG concentration and serum total protein (**STP**). From each calf, a 6-mL blood sample was taken from the jugular vein 48 h after colostrum intake ( $50.2 \pm 0.22$  h of age) in a blood tube without anticoagulant (Vacutainer, BD, Plymouth, UK). The blood samples were immediately centrifuged ( $3,000 \times g$  for 15 min at 4°C) and the serum was collected in 2-mL Eppendorf tubes and stored at  $-20^{\circ}$ C until analysis.

The determination of the IgG concentration in serum samples was carried out with the same technique, the same commercial kits and with the same procedures as those described for the determination of the IgG concentration in the colostrum. The serum samples were thawed at room temperature on the day of the determination and were immediately placed on the plates without prior dilution. In each RID plate, 3 of 24 wells were filled with reference sera at a concentration of 1.80, 14.02, and 28.03 g/L of IgG and the interassay CV was 9.8%, 6.8%, and 10.9% in low, medium, and high controls respectively. Samples that produced a value outside the range of the reference curve were diluted 1:2 with deionized water and retested.

Serum total proteins were determined by optical refractometry, using a clinical refractometer (Atago, Master-SUR/NM model, Tokyo, Japan). The %Brix of the serum samples was determined by digital refractometry (Atago, PAL-Grape Must model; scale 0 to 33% Brix) as an indirect measure of the IgG concentration. Both refractometers were calibrated with distilled water before the analysis of each batch of samples.

The apparent efficiency of IgG absorption (AEA) of IgG at the intestinal level was determined using the equation suggested by Quigley et al. (2002):

$$AEA = [serum IgG (g/L) \times plasma volume$$
  
(L)/IgG intake (g)] × 100,

where plasma volume was calculated as follows (Quigley et al., 1998):

Plasma volume =  $0.089 \times [BW \text{ at birth (kg)}].$ 

## DM and Nutrient Intake

Daily intake of feed in calves (milk and concentrate) was measured as the difference of weight between the feed offered and refused. The intake of DM, CP, and ME was determined from the chemical composition of the feeds, which was analyzed using the techniques previously described. For the statistical analysis of feed and nutrient intake, a single data point was used for each animal, corresponding to the average intake of each feed over the 30 d of measurement.

## **Body Growth and Development**

In all calves, BW, heart girth, and withers height were determined at 2 moments, immediately after birth and before colostrum ingestion (d 0), and on the last day of the experiment (d 30). The BW was determined with an electronic balance (Terko-TK3506, New Zealand). The heart girth was measured with a cloth tape measure, with a scale in centimeters, and the withers height was determined with a ruler equipped with a digital distance-meter (Bosh-Professional GLM 20, Stuttgart, Germany). Both the daily gain of BW (ADG), heart girth, and withers height Pastorini et al.: MASTITIS, COLOSTRUM, AND TPI IN DAIRY CALVES

	Treat	ment <sup>1</sup>		
Item	H-cow	L-cow	SEM	P-value <sup>2</sup>
Colostrum yield (kg)	5.11	5.32	0.53	0.77
Fat				
%	5.14	5.70	0.39	0.33
kg	0.27	0.31	0.04	0.55
Protein				
%	14.17	17.87	0.54	< 0.01
kg	0.72	0.93	0.09	0.08
Lactose				
%	2.82	2.91	0.10	0.56
kg	0.14	0.16	0.02	0.58
тs				
%	22.13	26.48	0.59	< 0.01
kg	1.13	1.41	0.14	0.19
$Ig\tilde{G}^{3}(g/L)$	74.7	92.8	4.42	0.01
Brix (%)	22.7	25.9	0.79	0.01
SCC (Logio)	6.2	5.8	0.12	0.02

 Table 3. Production and composition of colostrum produced by Holstein cows with high (H-cow) and low (L-cow) SCC at the dry-off

<sup>1</sup>Cows with high SCC at dry-off (mean 509,000 cells/mL) and cows with low SCC at dry-off (mean 117,000 cells/mL).

<sup>2</sup>Significant differences were if  $P \le 0.05$ , and trends were if  $0.05 < P \le 0.10$ .

<sup>3</sup>Immunoglobulin G concentration determined by RID.

were obtained as the difference between the measurement obtained on d 30 and the measurement obtained on d 0, divided by the length of the measurement period (30 d).

#### Statistical Analysis

All data were analyzed using SAS software version 9.0 (SAS Institute Inc., Cary, NC). Data were initially submitted for analysis to detect outliers and to check the normality of the residuals through univariate procedures (PROC UNIVARIATE). The SCC did not present a normal distribution, so the data were log-transformed. No data from any of the variables were excluded from the statistical analysis.

Data of yield and composition, IgG concentration and Brix percentage of colostrum were analyzed using the PROC MIXED procedure with the following model:

$$Y_{ij} = \mu + C_i + A_j + e_{ij},$$

where  $Y_{ij}$  is the dependent variable,  $\mu$  is the overall mean,  $C_i$  is the fixed effect of colostrum type (L-col and H-col) produced by cows,  $A_j$  is the random effect of the cow and  $e_{ij}$  is the residual error. The BW of the cow, gestation length, calving date, and parity of the cow were included in the model as covariates, but because they were not significant, they were removed from the final model.

The chi-squared test was used to analyze the proportion of colostrum samples from L-cow and H-cow according to different ranges of IgG concentration and % Brix, and



**Figure 1.** Distribution of IgG (g/L; top) and %Brix (bottom) in colostrum samples from Holstein cows with low SCC (L-cow; black bars) or high SCC (H-cow; gray bars) at dry-off. *P*: refers to the significance of the chi-squared analysis for the proportions of L-cow and H-cow within each IgG concentration or %Brix range. An asterisk (\*) indicates differences between the proportions of L-cow and H-cow within the %Brix range.

to analyze the proportion of colostrum samples with a concentration of IgG and % Brix greater than 71 g/L of IgG and 25% Brix, respectively. These cut-off points were used because it is estimated that calves should receive 4 L of colostrum with a minimum IgG concentration of 70 g/L (~25% Brix) to achieve an excellent TPI (Lombard et al., 2020), assuming that the AEA between 2 to 3 h after birth is 25% (Roche et al., 2015).

Data of intake (colostrum and nutrients), body growth, and development (BW, heart girth, and withers height), AEA, and TPI were analyzed using the PROC MIXED procedure with the following model:

$$Y_{ijkl} = \mu + C_i + B_j + D_k + B_j \times D_k + e_{ij},$$

where  $Y_{ijkl}$  is the dependent variable,  $\mu$  is the overall mean,  $C_i$  is the random effect of the calf,  $B_j$  is the fixed effect of colostrum category (L-col and H-col),  $D_k$  is the fixed effect of the calf category (L-calf and H-calf), and  $B_j \times D_k$  is the fixed effect of the interaction between colostrum category and calf category, and  $e_{ij}$  is the residual error. The sex of the calves and the time between birth and colostrum dosing were included in the statistical model as covariates but they were not significant they were removed from the final model.

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**Table 4.** Intake, transference of passive immunity (TPI) and apparent efficiency of absorption of IgG (AEA), at 48 h after colostrum intake, by neonatal calves born to and fed with colostrum<sup>1</sup> from Holstein cows with high (H-cow) or low (L-cow) SCC<sup>2</sup> at dry-off

	H-o	H-calf		L-calf		P-value		
Item	H-col	L-col	H-col	L-col	SEM	T-calf <sup>3</sup>	T-col <sup>4</sup>	$T$ -calf $\times$ $T$ -col <sup>5</sup>
Intake of IgG								
Total, g	281.0	332.2	262.5	301.7	22.51	0.31	0.05	0.80
g/kg of BW	8.6	9.9	7.9	9.4	0.63	0.39	0.03	0.90
TPI								
IgG, g/L	22.7	22.9	27.4	30.0	2.10	0.01	0.55	0.62
Brix. %	8.9	8.9	9.8	9.6	0.35	0.03	0.85	0.68
STP. <sup>6</sup> g/dL	6.2	6.0	6.6	6.9	0.30	0.03	0.81	0.35
AEA, %	25.1	23.8	29.4	30.7	1.95	0.01	0.99	0.53

<sup>1</sup>H-col = colostrum from H-cow; L-col = colostrum from L-cow.

 $^{2}$ H-calf = calves born to H-cow; L-calf = calves born to L-cow.

<sup>3</sup>Effect of calf type.

<sup>4</sup>Effect of colostrum type.

<sup>5</sup>Effect of the interaction between the calf type and colostrum type.

<sup>6</sup>Serum total proteins by clinical refractometer.

Means were compared with the Tukey test. Significant differences were declared at  $P \le 0.05$ , and trends were discussed at  $0.05 < P \le 0.10$ .

#### RESULTS

No differences were observed in colostrum yield at first milking between H-cow and L-cow (5.11 and 5.32 kg, respectively; SEM = 0.53; P = 0.77). Likewise, no differences were observed in fat and lactose (% and kg) between H-cow and L-cow (P > 0.33), and in TS yield (P = 0.186). However, protein concentration was 3.7 percentage points higher in L-cow than in H-cow (P < 0.01), and this was reflected in a greater TS concentration (4.4 percentage points) in L-cow (Table 3). Colostrum of L-cow had 24% higher IgG concentration and 3.2 percentage points higher % Brix than H-cow (P = 0.01). The colostrum produced by L-cow cows presented a lower SCC than colostrum produced by H-cow (5.8 vs. 6.2 Log10 SCC; SEM = 0.12; P = 0.02; Table 3).

The distribution of IgG and % Brix concentrations in the colostrum of L-cow and H-cow are presented in Figure 1. Overall, all colostrum samples presented IgG concentrations of more than 50 g/L, except one sample from a H-cow (43.5 g/L). The proportion of colostrum samples with concentrations greater than 71 g/L of IgG (85%) from L-cow was greater (P = 0.02) than colostrum from H-cow cows, and the proportion of colostrum samples with % Brix greater than 25% Brix from L-cow (70%) was greater (P < 0.01) than colostrum from H-cow (20%; Figure 1). The median of IgG concentration and %Brix was higher in the colostrum produced by L-cow compared with colostrum produced by H-cow (90.9 vs. 72.6 g/L and 25.8 vs. 22.4% Brix; L-cow and H-cow, respectively). No effect of interaction between colostrum type and calf type was observed, nor of calf type on total IgG intake expressed as grams or grams/kilogram of BW (Table 4). However, an effect of colostrum type was observed (Table 4). Calves fed L-col had a higher total IgG intake than calves fed H-col, either expressed in grams (317.0 and 271.8 g respectively; SEM = 15.3; P = 0.31) or grams/kilogram of BW (9.7 vs. 8.3 g/kg of BW; SEM = 0.4; P = 0.03).

Transference of passive immunity as determined by different methods was not affected by the interaction calf type × colostrum type, and there was also no effect of colostrum type used on the level TPI achieved by calves (Table 4). However, an effect of calf type on TPI achieved was observed, as L-calf calves achieved greater TPI than H-calf calves, irrespective of the method used (Table 4). The IgG concentration was 28.8 and 22.8 g/L for L-calf and H-calf, respectively (SEM = 1.5; P = 0.01), and the TPI determined by %Brix was 9.7% and 8.9% Brix for L-calf and H-calf, respectively (SEM = 0.3; P = 0.03). Additionally, when the TPI was evaluated by STP it was 6.8 versus 6.1 g/dL for L-calf and H-calf calves, respectively (SEM = 0.2; P = 0.03).

Neither an interaction between colostrum type and calf type, nor a colostrum type effect were observed for AEA (Table 4). However, an effect of calf type was observed, where L-calf calves achieved a higher AEA than H-calf calves (30.0% and 24.5%, respectively; SEM = 1.4; P = 0.01).

Neither an effect of an interaction between colostrum type and calf type, nor an effect of calf type was observed on milk, starter concentrate, or nutrient intake; however, an effect of colostrum type on DM, CP, and NEL intake from starter concentrate was observed (Table 5). Calves

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Table 5. Feed and nutrient intake of calves born and fed with colostrum<sup>1</sup> from Holstein cows with high (H-cow) or low (L-cow) SCC<sup>2</sup> at dry-off

	H-o	H-calf		L-calf		<i>P</i> -value		
Item	H-col	L-col	H-col	L-col	SEM	T-calf <sup>3</sup>	T-col <sup>4</sup>	$T$ -calf $\times$ $T$ -col <sup>5</sup>
Milk intake								
DM, g/d	737.46	721.00	690.50	720.36	36.94	0.52	0.86	0.54
CP, g/d	189.42	184.03	176.12	183.41	9.49	0.47	0.92	0.51
ME, Mcal/d	3.87	3.79	3.63	3.79	0.19	0.54	0.83	0.54
Starter concentrate intake								
DM, g/d	45.72	61.61	52.14	78.37	10.08	0.26	0.04	0.62
CP, g/d	8.23	11.09	9.38	14.11	1.81	0.26	0.04	0.61
ME, Mcal/d	0.14	0.19	0.16	0.24	0.03	0.26	0.05	0.61
Total intake								
DM, g/d	783.18	782.61	742.64	798.74	42.15	0.77	0.52	0.51
CP. g/d	197.65	195.12	185.50	197.52	10.30	0.64	0.65	0.49
ME, Mcal/d	4.01	3.98	3.79	4.03	0.21	0.69	0.62	0.52
CP/EM (g/Mcal)	49.31	49.08	48.97	49.05	0.18	0.33	0.68	0.41

<sup>1</sup>H-col = colostrum from H-cow; L-col = colostrum from L-cow.

 $^{2}$ H-calf = calves born to H-cow; L-calf = calves born to L-cow.

<sup>3</sup>Effect of calf type.

<sup>4</sup>Effect of colostrum type.

<sup>5</sup>Effect of the interaction between the calf type and colostrum type.

that received L-col had a higher DM (70.0 vs. 48.9 g/d; SEM = 7.13; P = 0.04), CP (12.6 vs. 8.8 g/d; SEM = 1.28; P = 0.04), and ME (0.21 and 0.15 Mcal; SEM = 0.02; P = 0.05) intake from the starter concentrate than calves that received H-col.

Calf growth and development as evaluated by BW, withers height, and heart girth is shown in Table 6. No effect of colostrum type (P > 0.34), calf type (P > 0.19), or their interaction (P > 0.32) was observed on any of the growth and development parameters studied.

## DISCUSSION

Clinical and subclinical mastitis are factors that affect the quantity and quality of colostrum (Maunsell et al., 1998; Gulliksen et al., 2008; Puppel et al., 2020), and if a neonatal calf is fed with low-quality colostrum, it could have an increased risk of having a failure of TPI (FTPI; Ferdowsi Nia et al., 2010; Forsbäck et al., 2010), with the consequent delay in growth and development, and a greater risk of morbidity and mortality (Lombard et al., 2020). It was also hypothesized that calves born to cows with high SCC at dry-off might have a reduced TPI. It was observed that colostral protein yield and IgG mass were lower in colostrum produced by H-cow cows, but this did not translate into a higher incidence of FTPI or growth retardation when calves ingested this type of colostrum, albeit a trend for a lower intake of starter was detected. In contrast, H-calf calves achieved a lower TPI and AEA of IgG, and this occurred irrespective of type of colostrum.

As expected, SCC in colostrum produced by Hcow cows was higher than colostrum by L-cow cows (1,584,000 vs. 631,000 cells/mL). It has been previously reported that the higher the SCC, the lower the colostrum yield (Maunsell et al., 1998; Ferdowsi Nia et al., 2010). Nonetheless, in our study, although colostrum yield was within the reported range (Borchardt et al., 2022, Rossi et al., 2023, Westhoff et al., 2023), no differences were observed between treatments. The type of colostrum did not affect fat concentration, in disagreement with Puppel et al. (2020), who showed an inverse relationship between SCC and fat concentration. Similarly, lactose concentration in colostrum was similar between groups, which is consistent with the lack of effect of SCC on colostrum yield, as lactose is one of its most osmotically active constituents and determines its volume (Oldham and Sutton, 1983). Protein content was lower in colostrum produced by H-cow cows as has been previously reported (Maunsell et al., 1998; Morrill et al., 2012; Puppel et al., 2020). This lower protein concentration could be due to the action of native proteases and proteases-especially those of the plasmin-plasminogen system-produced by psychrotrophic bacteria (Fajardo-Lira et al., 2000; Albenzio et al., 2004), whose activity may increase as soon as SCC increases above 100,000 cells/mL (Le Roux et al., 2003), and results in a higher proteolysis of colostrum proteins (Barbano et al., 1991; Forsbäck et al., 2010; Barbosa et al., 2013).

In our study, all except one colostrum sample produced by one H-cow cow were considered of good quality (>50 g/L, Weaver et al., 2000; McGuirk and Collins, 2004). The concentration of IgG was higher in the colostrum produced by L-cow cows compared with the colostrum by H-cow cows, which is consistent with previous studies (Maunsell et al., 1998; Kehoe et al., 2007; Morrill

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Table 6. Body growth and performance of calves born and fed with colostrum<sup>1</sup> from Holstein cows with high (H-cow) or low (L-cow) SCC<sup>2</sup> at dry-off

	H-o	H-calf		L-calf		<i>P</i> -value		
Item	H-col	L-col	H-col	L-col	SEM	T-calf <sup>3</sup>	T-col <sup>4</sup>	$T$ -calf $\times$ $T$ -col <sup>5</sup>
Initial								
BW, kg	33.9	34.3	32.1	34.0	1.82	0.58	0.54	0.69
Withers height, cm	71.1	69.6	69.1	69.7	1.02	0.36	0.67	0.32
Heart girth, cm	75.4	75.3	74.0	75.5	1.41	0.69	0.63	0.57
Final								
BW, kg	50.0	50.2	48.4	51.6	2.20	0.95	0.46	0.49
Withers height, cm	78.0	77.8	76.0	76.8	1.14	0.19	0.75	0.67
Heart girth, cm	85.6	85.3	84.2	84.2	1.29	0.56	0.78	0.64
Total BW gain								
Total kg	16.1	15.9	16.2	17.6	0.96	0.36	0.59	0.40
% of initial BW	48.3	47.8	51.1	53.0	3.52	0.26	0.83	0.73
Daily gains								
BW, g/d	538.2	528.5	541.0	585.4	32.02	0.36	0.59	0.40
Withers height, cm/d	0.23	0.27	0.23	0.24	0.03	0.54	0.34	0.56
Heart girth, cm/d	0.34	0.34	0.34	0.32	0.03	0.83	0.75	0.82

<sup>1</sup>H-col = colostrum from H-cow; L-col = colostrum from L-cow.

 $^{2}$ H-calf = calves born to H-cow; L-calf = calves born to L-cow.

<sup>3</sup>Effect of calf type.

<sup>4</sup>Effect of colostrum type.

<sup>5</sup>Effect of the interaction between the calf type and colostrum type.

et al., 2012). Brix refractometry results were consistent with IgG findings, as the measurement of total colostrum solids is highly correlated with the concentration of total proteins (Bielmann et al., 2010; Morrill et al., 2012). During colostrogenesis IgG is concentrated in colostrum mainly by active mechanisms such as endocytosis or transcytosis through mammary epithelial cells, and a lower proportion can also be transferred to colostrum through leaky tight joints (Baumrucker and Bruckmaier, 2014; Hernández-Castellano et al., 2018). Therefore, damaged epithelial cells or altered cellular function caused by an IMI could reduce IgG transport and result in a decrease in the concentration and total mass of IgG in colostrum (Larson et al., 1980). However, some authors reported that in cows with mastitis, the tight junctions between the epithelial cells of the mammary gland are damaged, thus reducing the integrity of the blood-milk barrier and allowing a greater passage of plasma proteins and antibodies from the blood to the milk (Nguyen and Neville, 1998; Bruckmaier and Wellnitz, 2017). For example, González-Cabrera et al. (2024) recently reported that an intramammary administration of LPS at parturition increased immunoglobulin concentrations in colostrum of dairy goats. Also, during mastitis cases caused by bacteria the IgG concentration in the colostrum of dairy cows can decrease because of proteolysis by endogenous and exogenous proteases (Fajardo-Lira et al., 2000; Albenzio et al., 2004).

Several authors have reported that the SCC of colostrum is much higher than that of milk (Hallberg et al., 1995; Andrew, 2001). In most cases these high SCC do not imply that there is mastitis, but it is result of a physiological feature described as the passage of cells through gaps in the junctions of mammary epithelial cells (Nguyen and Neville 1998). Although SCC has been used as an indicator of colostrum quality (Puppel et al., 2020), the cut-off points of SCC in colostrum are not clearly defined. Indeed, although some authors reported that an SCC in first-milking colostrum between 40,000 and 50,000 cells/mL or greater than 50,000 cells/mL was associated with a lower concentration of IgG in colostrum (Whist and Østerås, 2007; Gulliksen et al., 2008), others reported that the different components of colostrum and particularly the IgG concentration would be lower than 50 g/L when the SCC in colostrum is greater than 400,000 cells/mL (Morrill et al., 2012; Puppel et al., 2020). In our study, the SCC of colostrum was greater than 500,000 cells/mL, and the results are consistent with those reported by the previously cited authors where the increase in SCC caused a reduction in IgG concentration.

Irrespective of treatment, calves achieved an average intake of 295 g of IgG, which is greater than the minimum recommended to achieve an adequate TPI (200 g of IgG intake; Conneely et al., 2014). The higher IgG intake in calves that were fed with colostrum L-col is consistent with the higher concentration of IgG in that colostrum, as the volume assigned to each calf was the same. When IgG intake was measured as g/kg of BW, an effect of colostrum type was also observed, where calves fed with L-col received a higher dose of IgG. This is explained by the higher concentration of IgG in L-col , as no differences were observed in BW at birth between

H-calf and L-calf calves. However, although calves fed colostrum produced by L-cow received a greater mass of IgG, this was not reflected in the level of TPI achieved. This could be because even though calves fed L-col had a higher IgG intake, no differences were observed in AEA between calves fed L-col or H-col (27.3% and 27.3%; SEM = 1.95), suggesting that both groups of calves were at near-maximum intestinal absorption levels of IgG. This assumption is in line with Roche et al. (2015), who mentioned that maximum IgG absorption occurs between 2 to 3 h after birth and is  $\sim$ 25%. Our results are opposite to those reported by Ferdowsi Nia et al. (2010), who observed a linear reduction in the serum concentration of IgG at 3 h after the first ingestion of colostrum as the SCC in colostrum increased. The authors suggested that colostral SCC might have interfered with normal intestinal Ig absorption. However, a higher colostral SCC was observed by Ferdowsi Nia et al. (2010), which might have exerted a more detrimental effect on intestinal IgG absorption than in our experiment.

In our study, calves born to L-cow cows achieved a greater TPI than those born to H-cow cows, and we have not found similar reports to compare with our study. Lombard et al. (2020) proposed 4 categories of TPI in neonatal calves based on the concentration of serum IgG (excellent:  $\geq 25.0$  g/L, good: 18.0–24.9 g/L, fair: 10.0–17.9 g/L, and poor: <10 g/L). Thus, calves born to L-cow cows achieved an excellent TPI (28.8 g/L), whereas those born to H-cow cows achieved a good TPI (22.8 g/L). In addition, calves born to L-cow cows achieved an AEA of IgG that was 5.6 percentage points higher that calves born to H-cow cows, regardless of the type of colostrum ingested. The reason for these findings remains speculative but could be explained by changes in the physiology and structure of the intestinal tissues, due to a fetal programming process experienced in utero by the progeny of cows that suffered stress or diseases in the last stages of pregnancy (Abuelo, 2020). In this sense, maternal factors (prenatal factors), such as health or stress, can alter the development of fetal programming in exposed fetuses (Jeong, 2022). These factors are involved in a series of disorders linked to the intestinal and immunological microbiomes, producing disorders or diseases such as intestinal inflammation (de Vos et al., 2022). In addition, factors such as thermal stress or metabolic stress suffered by cows can cause an increase in the oxidative status of the newborn, an increase in inflammatory processes, an increase in the apoptosis of epithelial cells in the jejunum, and a decrease in the serum concentration of IgG (Diddeniya et al., 2024). Indeed, Santos et al. (2013) observed that calves born and fed colostrum from healthy cows had higher TPI and serum transferrin levels and lower serum haptoglobin concentrations than calves born and fed colostrum from cows with

subclinical or clinical mastitis. Haptoglobin is an acute phase protein of inflammation that is used as a predictor of acute inflammatory processes, and its increase is a function of the severity of the injury or inflammatory process (Eckersall and Conner, 1988), whereas transferrin is a negative protein of acute phase that decreases during infectious processes (Gruys et al., 2005). Nguyen et al. (2018) reported that prenatal inflammation induced by intra-amniotic exposure to LPS resulted in several responses in fetal gut of preterm pigs, including reduced villus heights. Therefore, we suggest that calves born to H-cow cows may have experienced some inflammatory process that could have direct consequences on AEA of IgG. However, it is necessary to carry out complementary studies to deepen the knowledge on whether these results are compatible with a fetal programming process due to the stress that occurs when a calf is gestated by a cow with mastitis, and how the impairment of intestinal absorption of IgG occurs.

It has been reported that calves that achieve an adequate TPI obtain long-term benefits, such as improved feed efficiency, reduced age at first calving (Robison et al., 1988; DeNise et al., 1989), reduced morbidity and mortality rates (Lombard et al., 2020) and improved preand postweaning ADG (Robison et al., 1988; Abuelo et al., 2021). Several experiments have studied the association between TPI and ADG with conflicting results; a positive association between IgG concentration and ADG has been reported in some studies (Elsohaby et al., 2019), but no differences were found in others (Nocek et al., 1984; Crannell and Abuelo, 2023). In the present study, ADG was not affected by the treatments, this could be associated with all calves achieving good or excellent TPI levels. Similarly, Sutter et al. (2023) reported no differences in ADG in calves with fair and excellent TPI; only the calves that achieved a poor TPI presented a worse performance. Similarly, the lack of differences between treatments regarding withers height and heart girth gains are consistent with the results observed for ADG. This absence of differences between treatments in development and growth parameters could be explained by the fact that, in addition to reaching in all cases more than acceptable levels of TPI that allowed maintaining an optimal health status during the experiment, all calves received the same nutritional management which resulted in no differences being observed in feed and nutrient intake throughout the experimental period.

### CONCLUSIONS

Cows with high SCC at dry-off produced colostrum with higher SCC and lower IgG concentrations, but when ingested by calves it did not affect TPI, feed intake, or growth and development. Calves born to cows with high SCC at dry-off presented a lower AEA of IgG in the small intestine, which translated into a lower serum concentration of IgG, irrespective of type of colostrum that was fed.

#### NOTES

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**Nonstandard abbreviations used:** AEA = apparent efficiency of IgG absorption; FTPI = failure of TPI; H-calf = calves born to H-cow cows; H-calfxH-col = H-calf fed with H-col; H-calfxL-col = H-calf fed with L-col; H-col = colostrum from H-cow cows; H-cow = cows that presented in each of the last 3 SCC before dry-off greater than 200,000 cells/mL; L-calf = calves born to L-cow cows; L-calfxH-col = L-calf fed with H-col; L-calfxL-col = L-calf fed with L-col; L-calfxL-col = L-calf fed with H-col; L-calfxL-col = L-calf fed with L-col; L-col = colostrum from L-cow cows; L-cow = cows that presented in each of the last 3 SCC before dry-off less than 200,000 cells/mL; RID = radial immunodiffusion; STP = serum total protein; TPI = transference of passive immunity.

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