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Short communication: The effect of storage conditions over time on bovine colostral immunoglobulin G concentration, bacteria, and pH

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ABSTRACT

The objective of the present study was to measure the effect of storing colostrum in different conditions for varying amounts of time on IgG concentration, bacteria, and pH. In experiment 1, colostrum from 12 Holstein-Friesian cows (6 primiparous and 6 multiparous) was collected within 3 h of calving, and colostrum from another 12 multiparous cows was collected within 3 h of calving (6 cows) and >9 h postpartum (6 cows). Aliquots were refrigerated or stored at room temperature for up to 72 h, depending on treatment. In experiment 2, colostrum was collected from 6 multiparous cows within 9 h of calving, and aliquots were stored for up to 72 h in temperature-controlled units set at 4, 13, and 20°C. All colostrum samples were analyzed for IgG concentration, total bacteria count, and pH after 0, 6, 12, 24, 36, 48, 60, and 72 h of storage. Storage conditions did not affect the IgG concentration of colostrum. Bacterial growth was most rapid in the first 6 h of storage, reducing thereafter, but bacteria multiplied at a significantly greater rate when stored in warmer conditions (i.e., >4°C). The pH of colostrum was not significantly altered when stored at temperatures <13°C, but when stored at 20°C the pH significantly decreased after 24 h of storage. Storing colostrum in warmer conditions significantly alters both total bacteria count and pH; consequently, colostrum should be stored at ≤4°C.
Key words: colostrum storage, immunoglobulin G, bacteria, pH

Short Communication

Colostrum is the first milk secreted by the cow after parturition (Park, 1993) and contains immunoglobulins that are essential for protecting the neonatal calf against disease (Weaver et al., 2000). Concentration of IgG in colostrum can be influenced by numerous factors, including parity (Pritchett et al., 1991; Conneely et al., 2013), timing of collection postpartum (Moore et al., 2005; Conneely et al., 2013), and storage management (Csapó et al., 1994; McGuirk and Collins, 2004). Exposing colostrum to fluctuations in temperature through storage management may alter the physical properties of colostrum, such as pH and bacterial content, which may affect the absorption of immunoglobulins by calves (James et al., 1981).

In Ireland, >90% of dairy farmers store colostrum, and up to 17% of them store colostrum at room temperature (Cummins et al., 2016). Storing colostrum at >4°C allows growth of bacteria and a reduction in pH (Stewart et al., 2005). The pH of colostrum may differ in colostrum with varying IgG at collection, and thus may influence the bacteria levels differently; however, this has yet to be investigated. The effect of colostrum storage at ≥4°C on IgG concentration, bacteria, and pH has not previously been investigated on a single sample set; hence, it is difficult to ascertain the key changes and interactions that may occur. Therefore, the objective of the present study was to measure the effect of various storage conditions over time on IgG concentration, bacteria, and pH in colostrum of Holstein-Friesian dairy cows in Ireland.

Colostrum samples were collected and stored in various conditions during February and March, 2013, at the Teagasc Moorepark Research farm located in County Cork, in southern Ireland (52°9′N, 8°16′W).

In experiment 1, colostrum was individually collected from 24 Holstein-Friesian cows. Animals in experiment 1 were selected and separated into 2 groups according to (1) parity (group 1) and (2) time of collection postpartum (group 2). In group 1, 6 cows were primiparous and 6 cows were multiparous (in their fourth or greater lactation). In group 2, 6 multiparous cows had calved within 3 before collection and 6 multiparous cows had calved 9 h before collection. The aim was, using evidence from previous research (Conneely et al., 2013), to collect colostrum with varying IgG concentrations to investigate if this caused differences in bacterial growth or pH. In experiment 2, colostrum was individually

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collected from 6 multiparous Holstein-Friesian cows within 3 h postpartum.

All colostrum was treated exactly the same during harvesting. Each cow was individually milked by machine into a clean churn at the next milking time and total volume of colostrum was recorded. In each storage condition, 8 aliquots of 100 mL (1 for each storage time point) were transferred into 120-mL polypropylene storage containers using a clean, dry, 1-L jug. Once filled, storage containers were sealed and placed in their assigned storage conditions. Two storage conditions were investigated in experiment 1: room temperature (**RT**) and cold storage at 4°C. Ambient air temperature of RT storage was measured daily at 0900, 1500, and 2100 h using a thermometer to identify the temperature fluctuations that the samples were exposed to. One aliquot was taken from each storage condition at 0 (time of collection, control), 6, 12, 24, 36, 48, 60, and 72 h postcollection. Samples were immediately frozen at −18°C for later analysis of IgG, total bacterial count (**TBC**), and pH. At the end of the storage period, an overall average RT storage temperature was obtained for each cow's colostrum from the combined readings.

There were 3 allocated storage conditions in experiment 2, controlled using 3 separate temperature-controlled units: 4, 13, and 20°C. To examine samples in duplicate, 2 aliquots were taken from each storage unit and frozen at −18°C for later analysis of IgG, TBC, and pH at 0, 6, 12, 24, 36, 48, 60, and 72 h postcollection.

Aliquots from experiments 1 and 2 were defrosted in a fridge (≤4°C) for approximately 8 h. All laboratory analysis was conducted immediately after defrosting and stored at ≤4°C between analyses to minimize potential growth of bacteria. The IgG concentration was determined by the ELISA method (Bovine IgG ELISA Kit Cat. No. 8010, Alpha Diagnostic International, San Antonio, TX). Samples were assayed in duplicate, with an interassay coefficient of variation of 0.11, and an average of the 2 readings was obtained. The procedure is described by Conneely et al. (2013).

To identify colostral TBC, aliquots were thoroughly mixed and diluted with serial dilution at different rates according to storage condition, using maximum recovery diluents of 9 mL (1 g of peptone/L and 8.5 g of NaCl/L of water). To determine the most appropriate dilution rate for each aliquot, superfluous samples were initially diluted at 10^4. The lowest dilution rate was 10^4 (0-h aliquots), whereas the highest was 10^7 (72-h, 20°C aliquots). Aerobic count plates (3M Petrifilm Aerobic Count Plates; 3M, St. Paul, MN) with 1 mL of diluted aliquot were incubated at 32°C for 48 h. Each aliquot was tested in duplicate, and an average of the 2 results was obtained. A 3M Petrifilm Plate Reader was used to verify TBC (cfu/mL).

Samples were measured for pH status using an OHM Delta 2105.2 pH/mV meter (Delta OHM S.r.L, Caselle di Selvazzano, Italy). Each sample was tested in duplicate to an accuracy of 3 decimal places and an average of the 2 readings was obtained. Calibration was carried out before each test period and the probe was cleaned on a weekly basis according to the product guidelines.

Colostrum IgG concentration and pH were normally distributed. Total bacterial count was right skewed and thus a log-transformation was performed, which then presented a normal distribution. Factors associated with colostrum IgG concentration, TBC, and pH were determined using a fixed effects multiple regression model in PROC MIXED in SAS (ver. 9.3, SAS Institute Inc., Cary, NC). Sample hour (hour at which samples were frozen) was included as a repeated measure for each cow. In experiment 1, temperature was included as a random variable.

Additional variables were tested for an association with IgG, TBC, and pH in a series of univariate analyses. Month of calving, dry period length, protein content of milk postcalving, calving difficulty, sex of the calf, whether the calf was born alive or stillborn, and weight of the calf at birth were considered as confounding variables. Total bacteria count and pH were included as independent variables when the dependent variable was IgG concentration. Immunoglobulin G concentration and pH of colostrum were included as independent variables when the dependent variable was TBC. Total bacteria count and IgG concentration were included as independent variables when the dependent variable was pH. In experiment 1, group (i.e., parity or collection time) was included as an independent variable at all times according to the sample set under investigation.

All variables associated ($P < 0.05$) with the dependent variable in the univariate analyses were included in a multiple regression model. Nonsignificant variables ($P > 0.05$) were sequentially removed using backward elimination. Once all remaining independent variables were associated ($P < 0.05$) with the dependent variable, the removed variables were once again tested for significance with the significant variables forced into the multiple regression model. Interactions between all significant variables were examined. Least squares means were compared.

The mean recorded ambient air temperature for the duration of the experiment was 6°C (range = 0–10°C). The maximum range in temperature on any 1 d was 5°C (Figure 1). The average volume of colostrum produced by cows sampled <3 h postpartum was 6 L (SD = 1.88 L; range = 4–9 L), whereas the average volume of colostrum produced by cows sampled >9 h postpartum was 8 L (SD = 2.67; range = 5–13 L), but this difference was not significant. No confounding variable investi-

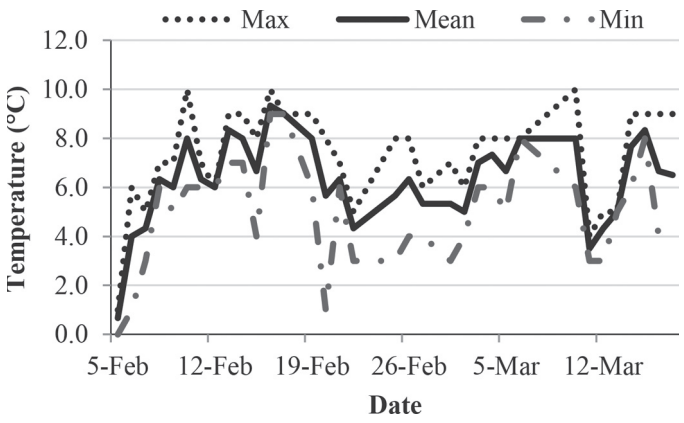

Figure 1. Average daily temperature with maximum (Max) and minimum (Min) temperatures recorded daily at 3 intervals from February 5 to March 17, 2013.

gated (e.g., month of calving and calving difficulty) had a significant effect on IgG concentration, TBC, or pH; thus, they were removed from all models and will not be further discussed.

In experiment 1, the mean IgG concentration of colostrum at 0 h was 116 g/L (Table 1; range = 20–195 g/L). The mean IgG concentration in experiment 2 was 116 g/L (Table 1; range = 67–170 g/L). Colostrum collected from cows <3 h postpartum was greater ($P < 0.01$) in IgG concentration (129.9 ± 8.78 g/L of IgG) than colostrum collected >9 h postpartum (86.6 ± 8.78 g/L of IgG). When colostrum was stratified based on IgG concentration, no difference was noted between colostrum with high IgG concentration (125.4 g/L of IgG) and low IgG concentration (43.7 g/L of IgG) in terms of TBC or pH. No other significant effects of varying colostrum storage on IgG concentration were observed.

In experiment 1, TBC was 455,722 cfu/mL at 0 h (time of collection); an interaction occurred between treatment and length of storage. Regardless of treatment (i.e., storage condition), TBC of colostrum increased at an average rate of 5% (~432,000 cfu/mL) in the first 6 h of storage. From 6 to 24 h of storage, growth rate was 2% (286,800 cfu/mL) in both colostrum storage treatments. From 24 to 48 of storage the growth rate of TBC between each sampling time was 0% in colostrum stored in cold storage, whereas growth rate was 4% (>2,000,000 cfu/mL) in colostrum stored at RT. Although bacterial growth rate was ≥5% in both colostrum treatments from 48 to 60 h of storage, TBC continued to be higher ($P < 0.01$) in colostrum stored at RT (Figure 2). No other significant effects of varying colostrum storage on TBC were observed.

In experiment 2, TBC was 97,858 cfu/mL at h 0 (time of collection). There was an interaction between treatment and length of storage, and colostrum stored at

Table 1. Effect of storing colostrum in cold storage or at room temperature for 72 h on IgG concentration and pH (experiment 1)

	Treatment (Trt)[1]			Hour									*P*-value		
Item	CS	RT	SEM	0	6	12	24	36	48	60	72	SEM	Trt	Hour	Trt × hour
IgG (g/L)	116	116	8.327	116	116	116	116	115	115	116	116	5.915	0.990	0.778	0.634
pH	6.540	6.525	0.0602	6.627^{a}	6.545^{b}	6.529^{bd}	6.507^{ce}	6.544^{b}	6.519^{cd}	6.497^{ef}	6.489^{f}	0.0083	0.806	<0.01	0.092

$^{a-f}$Means with different superscripts differ ($P < 0.05$).
[1]CS = cold storage; RT = room temperature.

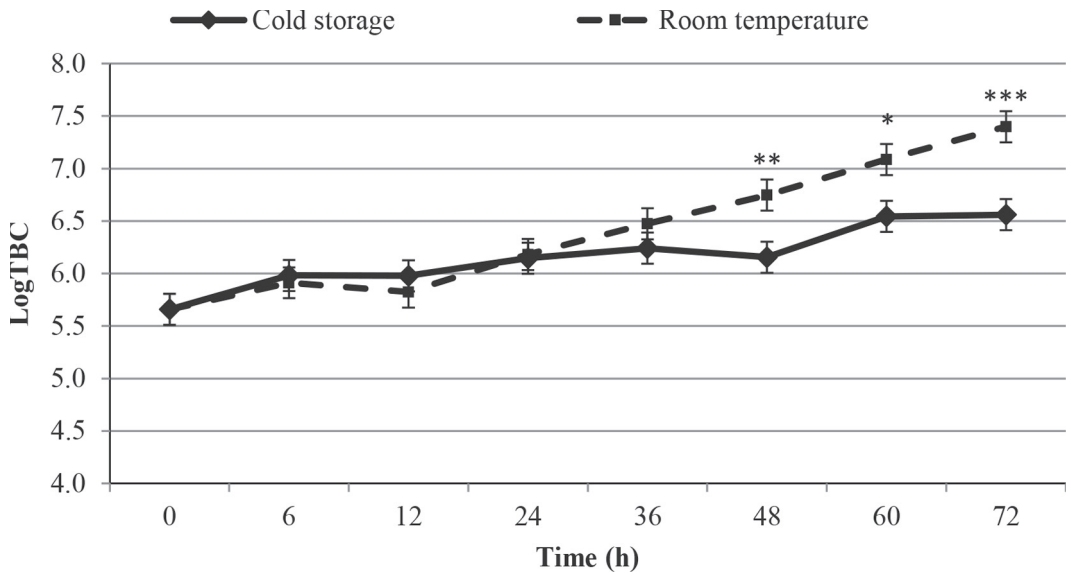

Figure 2. Log total bacterial count (LogTBC) of colostrum stored in cold storage or at room temperature for up to 72 h (experiment 1). Error bars represent 1 SE each side of the LSM. Asterisks represent difference among means at $^{*}P < 0.05$, $^{**}P < 0.01$, or $^{***}P < 0.001$.

20°C had a greater ($P < 0.01$) TBC from 6 h of storage compared with colostrum stored at 13 and 4°C (Figure 3). From 24 h, colostrum stored at 13°C had a greater ($P < 0.01$) TBC than colostrum stored at 4°C (Figure 3), but was lower than colostrum stored at 20°C. Regardless of storage temperature, TBC increased rapidly during the first 6 h after collection, but growth of TBC was more rapid ($P < 0.01$) in colostrum stored at 20°C (Figure 3). Total bacterial growth rate in the first 6 h was 21% (947,000 cfu/mL) in colostrum stored at 20°C, 10% (215,000 cfu/mL) in colostrum stored at 13°C, and 8% (114,000 cfu/mL) in colostrum stored at 4°C. From 6 to 24 h of storage, bacteria continued to grow at a greater rate in colostrum stored at 20°C (12%; 19,000,000 cfu/mL) compared with colostrum stored at 13 (9%; 138,000 cfu/mL) and 4°C (1%; 30,000 cfu/mL). After 24 h of storage, the mean bacterial growth rate in colostrum stored at 20°C was 3.4%, whereas colostrum stored at 13°C grew at a rate of 5.7% and colostrum stored at 4°C grew at a mean rate of 3.6%.

For each unit increase in logTBC in experiment 1 pH decreased by 0.07 units, and for each unit increase in

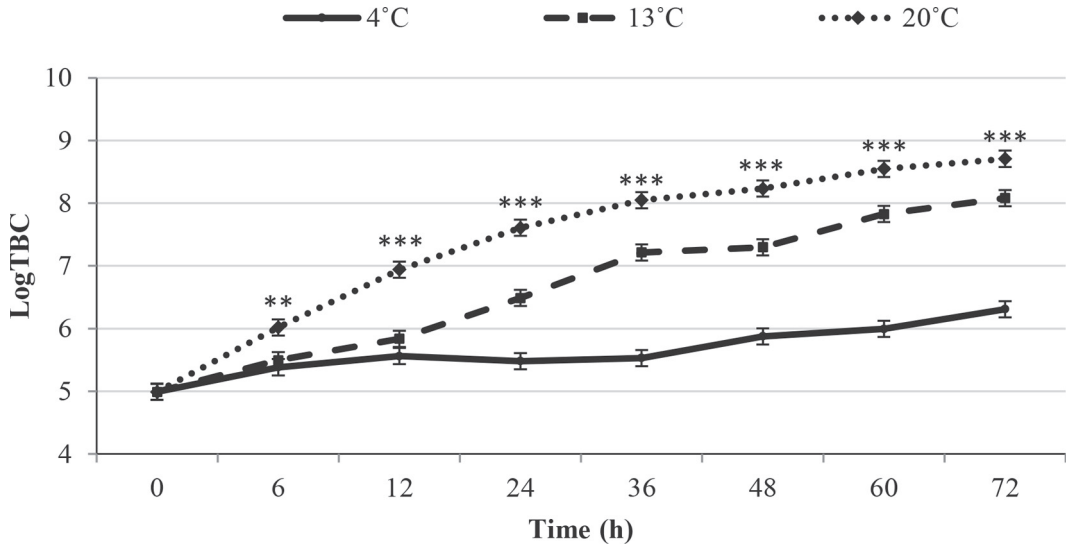

Figure 3. Log total bacterial count (LogTBC) of colostrum stored for 72 h at 4, 13, and 20°C (experiment 2). Error bars represent 1 SE each side of the LSM. Asterisks represent difference among means at $^{**}P < 0.01$ or $^{***}P < 0.001$.

logTBC in experiment 2 pH decreased by 0.23 units. In experiment 1, the greatest colostral pH decrease in all colostrum samples during the initial 6 h of storage (Table 1). When sampled at 36 h, the pH of colostrum had increased ($P < 0.01$), before it continued to decrease by $\geq$0.02 units between samplings until 60 h, where it remained stable until 72 h (Table 1).

In experiment 2, an interaction was noted between pH and length of storage. No difference in pH was observed between any of the 3 treatments for the first 12 h (Figure 4). When pH was measured again after 24 h of storage, we found a large decline in pH for samples stored at 20°C. This decrease continued, with a mean decrease of 0.36 units from 34 to 60 h, after which pH decreased by 0.07 units. In colostrum stored at 13°C, little change occurred before 48 h, after which pH decreased by 0.11 units between each sampling. The pH of colostrum stored at 4°C decreased by a total of 0.05 units during the entire experiment. No other significant effects on pH were observed.

The aim of the present study was to identify the effect of storing colostrum at different temperatures for up to 72 h on IgG concentration, TBC, and pH. To date, no study has identified the effects of storage on IgG concentration, TBC, and pH in one colostrum sample set. Although the effect of parity and time of colostrum collection on IgG concentration has previously been identified, the effect of these on TBC and pH during storage has not. Additionally, a critical storage condition and duration has not been identified. Neither storage method nor duration of storage altered IgG concentration in the present study. Previous research that investigated the effect of freezing on colostral IgG concentration, rather than at temperatures >0°C, as examined in the present study (Schipper et al., 1981). Moreover, our study investigated the effect of storage on colostrum of varying IgG concentrations, whereas previous research had lower mean colostral IgG concentrations (60 to 70 g/L) and did not examine colostrum of varied IgG concentration (Donahue et al., 2012; Morrill et al., 2015).

Agreeing with previous research, the colostrum IgG concentration was significantly lower in samples collected >9 h postpartum (Moore et al., 2005; Conneely et al., 2013). The present study, however, indicated that IgG concentration is not affected when colostrum was stored under different conditions. This suggests that the decrease in IgG absorption in calves that was previously reported (Foley et al., 1978; James et al., 1981) may not be due to changes in IgG concentration in colostrum, but rather interactions of IgG with other components, such as bacteria (James et al., 1981). Moreover, recent research reported that when colostrum with decreased bacterial content was consumed, calves achieved greater rates of passive transfer of IgG (Johnson et al., 2007; Elizondo-Salazar and Heinrichs, 2009; Godden et al., 2012; Gelsinger et al., 2015).

Through subjecting colostrum to various storage conditions, the present study generated colostrum with varying amounts of bacteria. This indicates the important influence various storage conditions can have. Although precautions were taken to collect colostrum using clean equipment, many samples exceeded the previously suggested maximum TBC threshold at time of collection (McGuirk and Collins, 2004). Thus, any bacterial growth during storage is concerning. The

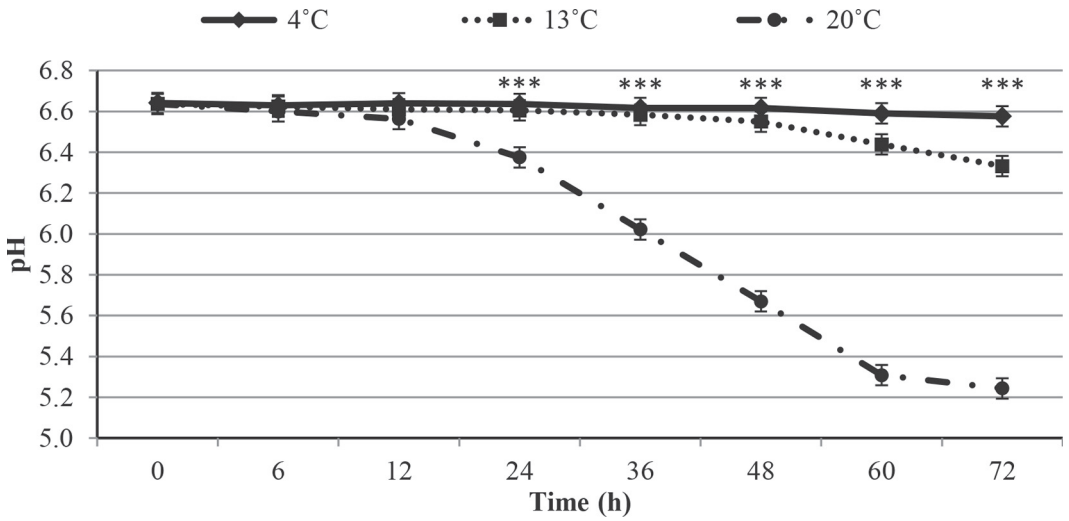

Figure 4. The pH of colostrum stored for up to 72 h at 4, 13, and 20°C (experiment 2). Error bars represent 1 SE each side of the LSM. Asterisks represent difference among means at ***$P < 0.001$.

TBC in experiment 1 (5.66 cfu/mL of LogTBC) and experiment 2 (4.99 cfu/mL of LogTBC) were similar to previous research (4.99 cfu/mL of LogTBC; Stewart et al., 2005). The lower TBC in refrigerated colostrum samples indicates that colostrum should be stored in a fridge to minimize growth of bacteria. Unlike previous research (Stewart et al., 2005), a decline in TBC was not experienced after the initial growth phase. In experiment 1, the continuous bacterial growth of >3% for the duration of the storage period may have been due to the temperature fluctuations encountered (i.e., although temperatures were low on average, colostrum in this treatment was also subject to temperatures up to 10°C), demonstrating the importance of low controlled temperature storage, regardless of average daily temperatures. Colostrum that was stored at a constant temperature of 20°C in the present study had a similar bacterial growth pattern to previous research (Stewart et al., 2005), but it multiplied at a greater rate; this is may be due to the fluctuations of ambient temperatures in the previous study (Stewart et al., 2005). The colostrum stored at 4°C in the present study followed a similar trend to previous research (Stewart et al., 2005).

Interestingly, TBC in experiment 1 initially had the same growth as experiment 2 (0–48 h; 0.49 cfu/mL LogTBC). However, from 48 h of storage, bacterial growth was numerically far less in experiment 1 (0.01 cfu/mL LogTBC) than experiment 2 (0.89 cfu/mL LogTBC). As TBC of colostrum stored at 4°C was far higher at collection in experiment 1 than 2, it was also greater at 48 h. It appears that when the TBC of colostrum stored at 4°C exceeds 1,000,000 cfu/mL (>6.0 cfu/mL logTBC), the growth rate declines. Thus, it may explain why growth became slower in experiment 1, a similar trend to colostrum stored at 13°C or 20°C in experiment 2. As TBC of colostrum stored at 4°C was far higher at collection in experiment 1 than experiment 2, it was also greater at 48 h. It appears that when the TBC of colostrum stored at 4°C exceeds 1,000,000 cfu/mL (>6.0 cfu/mL logTBC) the growth rate declines. Thus, it may explain why growth became slower in experiment 1, a similar trend to colostrum stored at 13°C or 20°C in experiment 2. Moreover, it is possible that the type of bacteria differed between colostrum samples, responding differently to varied storage; however, this was not examined in the present study.

The mean pH of colostrum in the present study was 6.63 units, which corresponds to some previous research (Foley et al., 1978), but is greater than other reports (Stewart et al., 2005). This may be due to difference in terms of colostrum composition, as milk from cows used in the present study had a high protein content (mean = 3.89% ± 0.289), which may influence buffering capacity of the milk and result in greater pH (Park, 1991). Although pH did decline in the present study, no storage condition resulted in a colostrum pH <5.5, which may be the reason that bacteria continued to grow. Previous research reported a ceasing of bacterial growth at a pH <5.5 (Stewart et al., 2005). Despite the rise of TBC to levels similar to previous research (Log_{10} >7.0; Stewart et al., 2005), pH did not decrease <5.0 units, which may be due to the greater initial pH (>6.0 units) compared with previous research (pH = 5.6; Stewart et al., 2005). Although pH decreased by no more than 0.5 units in total, (0.5 units less than previous research; Stewart et al., 2005), it was correlated with TBC growth, indicating an association between the changes in TBC and pH. Furthermore, TBC of colostrum in the present study did not multiply as rapidly as previously reported, thus conditions did not become as acidic. The pH of colostrum is affected by different buffering properties, such as casein, and pH is also quite dependent on the temperature at which the colostrum is stored (Singh et al., 1997). The elevated pH of colostrum at h 36 in experiment 1 may be highlighted due to the small sample set of cows. As the difference was not more than 0.03 units, it is unlikely that this is a significant biological result.

Colostrum is significantly affected by storage conditions and duration. Bacteria and pH are significantly altered and changes are more rapid when colostrum is stored >4°C. The first 6 h postcollection are critical, as a gross increase in TBC occurs. The rate of bacterial growth is greater with higher storage temperatures, and thus colostrum should be refrigerated immediately after collection to minimize bacterial growth. Although IgG is not affected by colostrum storage or duration, the rate of absorption in the calf may be affected by the bacterial and pH, and thus further investigations need to be conducted.

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REFERENCES

Conneely, M., D. Berry, R. Sayers, J. Murphy, I. Lorenz, M. Doherty, and E. Kennedy. 2013. Factors associated with the concentration of immunoglobulin G in the colostrum of dairy cows. Animal 7:1824–1832.

Csapó, J., Z. Csapó-Kiss, T. G. Martin, J. Szentpeteri, and G. Wolf. 1994. Composition of colostrum from goats, ewes and cows producing twins. Int. Dairy J. 4:445–458.

Cummins, C., D. Berry, R. Sayers, I. Lorenz, and E. Kennedy. 2016. Questionnaire identifying management practices surrounding calving on spring-calving dairy farms and their associations with herd size and herd expansion. Animal 10.1017/S1751731116000124.

Donahue, M., S. Godden, R. Bey, S. Wells, J. Oakes, S. Sreevatsan, J. Stabel, and J. Fetrow. 2012. Heat treatment of colostrum on commercial dairy farms decreases colostrum microbial counts while maintaining colostrum immunoglobulin G concentrations. J. Dairy Sci. 95:2697–2702.

Elizondo-Salazar, J. A., and A. Heinrichs. 2009. Feeding heat-treated colostrum to neonatal dairy heifers: Effects on growth characteristics and blood parameters. J. Dairy Sci. 92:3265–3273.

Foley, J. A., A. Hunter, and D. Otterby. 1978. Absorption of colostral proteins by newborn calves fed unfermented, fermented, or buffered colostrum. J. Dairy Sci. 61:1450–1456.

Gelsinger, S. L., C. M. Jones, and A. J. Heinrichs. 2015. Effect of colostrum heat treatment and bacterial population on immunoglobulin G absorption and health of neonatal calves. J. Dairy Sci. 98:4640–4645.

Godden, S. M., D. J. Smolenski, M. Donahue, J. M. Oakes, R. Bey, S. Wells, S. Sreevatsan, J. Stabel, and J. Fetrow. 2012. Heat-treated colostrum and reduced morbidity in preweaned dairy calves: Results of a randomized trial and examination of mechanisms of effectiveness. J. Dairy Sci. 95:4029–4040.

James, R. E., C. Polan, and K. Cummins. 1981. Influence of administered indigenous microorganisms on uptake of [iodine-125] γ-globulin in vivo by intestinal segments of neonatal calves. J. Dairy Sci. 64:52–61.

Johnson, J. L., S. Godden, T. Molitor, T. Ames, and D. Hagman. 2007. Effects of feeding heat-treated colostrum on passive transfer of immune and nutritional parameters in neonatal dairy calves. J. Dairy Sci. 90:5189–5198.

Lomba, F., I. Fumiere, M. Tshibangu, G. Chauvaux, and V. Bienfet. 1978. Immunoglobulin transfer to calves and health problems in large bovine units (1). Ann Rech. Tech. 9:353–360.

McGuirk, S. M., and M. Collins. 2004. Managing the production, storage, and delivery of colostrum. Vet. Clin. North Am. Food Anim. Pract. 20:593–603.

Moore, M., J. W. Tyler, M. Chigerwe, M. E. Dawes, and J. R. Middleton. 2005. Effect of delayed colostrum collection on colostral IgG concentration in dairy cows. J. Am. Vet. Med. Assoc. 226:1375–1377.

Morrill, K. M., K. E. Robertson, M. M. Spring, A. L. Robinson, and H. D. Tyler. 2015. Validating a refractometer to evaluate immunoglobulin G concentration in Jersey colostrum and the effect of multiple freeze–thaw cycles on evaluating colostrum quality. J. Dairy Sci. 98:595–601.

Park, S. C. N. L. J. 1993. The mammary gland and lactation. Pages 711–727 in Dukes' Physiology of Domestic Animals. 11 ed. M. J. Swenson and W. O. Reece, ed. Comstock Publishing Company Inc., New York, NY.

Park, Y. W. 1991. Relative buffering capacity of goat milk, cow milk, soy-based infant formulas, and commercial nonprescription antacid drugs. J. Dairy Sci. 74:3326–3333.

Pritchett, L. C., C. C. Gay, T. E. Besser, and D. D. Hancock. 1991. Management and production factors influencing immunoglobulin g1 concentration in colostrum from Holstein cows. J. Dairy Sci. 74:2336–2341.

Schipper, I., P. Kotta, G. Staples, G. Fisher, and G. Erickson. 1981. Immunoglobulin-G content in bovine colostrum preserved by freezing, fermentation and chemical preservatives. N. D. Farm Res. 39:25–27.

Singh, H., O. McCarthy, and J. Lucey. 1997. Physico-chemical properties of milk. Pages 469–518 in Advanced Dairy Chemistry. Volume 3. Springer, New York, NY.

Stewart, S., S. Godden, R. Bey, P. Rapnicki, J. Fetrow, R. Farnsworth, M. Scanlon, Y. Arnold, L. Clow, and K. Mueller. 2005. Preventing bacterial contamination and proliferation during the harvest, storage, and feeding of fresh bovine colostrum. J. Dairy Sci. 88:2571–2578.

Weaver, D. M., J. W. Tyler, D. C. VanMetre, D. E. Hostetler, and G. M. Barrington. 2000. Passive transfer of colostral immunoglobulins in calves. J. Vet. Intern. Med. 14:569–577.