

2019

# Evaluation of colostrum on an immunological and microbiological level and its relation to calf health: a case study of calves at UVM Miller Research Center

Brooke Ann Pietrafesa  
*University of Vermont*

Follow this and additional works at: <https://scholarworks.uvm.edu/hcoltheses>

---

## Recommended Citation

Pietrafesa, Brooke Ann, "Evaluation of colostrum on an immunological and microbiological level and its relation to calf health: a case study of calves at UVM Miller Research Center" (2019). *UVM Honors College Senior Theses*. 291.  
<https://scholarworks.uvm.edu/hcoltheses/291>

This Honors College Thesis is brought to you for free and open access by the Undergraduate Theses at ScholarWorks @ UVM. It has been accepted for inclusion in UVM Honors College Senior Theses by an authorized administrator of ScholarWorks @ UVM. For more information, please contact [donna.omalley@uvm.edu](mailto:donna.omalley@uvm.edu).

1 **Interpretive Summary**

2 Evaluation of colostrum on an immunological and microbiological level and its relation to calf  
3 health: a case study of calves at UVM Miller Research Center

4 *By Brooke A. Pietrafesa*

5  
6 Colostrum is the first milk that is let down from the mammary gland of a mammal  
7 following birth. Calves are born with immature immune systems and need to consume colostrum  
8 to obtain immunoglobulins. Bacteria present within colostrum can cause adverse health effects in  
9 calves. This study evaluated colostrum fed to calves and its effect on calf health and explored the  
10 relationship between immunoglobulin and bacterial content of the colostrum. It was observed  
11 that the samples had high bacteria counts and the relationship between immunoglobulin and  
12 bacteria counts and their effect on average daily gain should be further explored.

13  
14  
15  
16 **RUNNING HEAD: ANALYSIS OF COLOSTRUM QUALITY**

17 **Evaluation of colostrum on an immunological and microbiological level and its relation to**  
18 **calf health: a case study of calves at UVM Miller Research Center**

19  
20 Brooke A. Pietrafesa\* and John W. Barlow\*<sup>1</sup>

21 \*Department of Animal and Veterinary Sciences, University of Vermont, Burlington 05405

22  
23  
24  
25  
26  
27  
28  
29 <sup>1</sup>John W. Barlow: ([john.barlow@uvm.edu](mailto:john.barlow@uvm.edu))

## ABSTRACT

30  
31 The objective of this study was to evaluate the colostrum quality fed to calves at the  
32 UVM Miller Research Center on an immunological and bacteriological level as well as to  
33 explore the relationship between these factors and the health of the calves. Colostrum samples  
34 fed to twelve calves during their first and second feedings were collected. This colostrum was  
35 evaluated using a colostrometer, Brix refractometer, and was also plated on 3M Petrifilm  
36 coliform count plates, lactic acid bacteria plates, staph express count plates, and aerobic count  
37 plates. During the study the weights of the calves were taken once per week and their rectal  
38 temperatures were taken twice per week. Of the 18 colostrum samples, 7 of the Brix  
39 refractometer readings did not match the colostrometer readings. 9 of the 18 colostrum samples  
40 analyzed had coliform counts and total aerobic counts higher than the recommended levels.  
41 There was a significant relationship ( $p=0.0092$ ) between a calf having a fever within the first 14  
42 days of life and the average daily gain of the calf. The colostrum Brix values were also compared  
43 to the bacterial cfu/mL counts for coliform bacteria ( $p=0.0145$ ), aerobic bacteria ( $p=0.0381$ ),  
44 lactic acid bacteria ( $p=0.0209$ ) and *Staphylococcus* bacteria ( $p=0.0364$ ). Each showed a  
45 moderate linear negative correlation between the variables. The relationship between the sum of  
46 the lactic acid bacteria counts of the colostrum samples fed to the calves and the average daily  
47 gain of the calves was significant ( $p=0.0406$ ). There was also a significant relationship between  
48 the average daily gain and the total sum of the aerobic counts ( $p=0.0191$ ) as well as total sum of  
49 the coliform counts with ( $p=0.0229$ ). Although significant relationships were found, more  
50 studies should be performed with larger sample sizes to further explore the immunological and  
51 bacteriological relationships as well as to compare the different methods of evaluating colostrum  
52 IgG to the gold standard of RID to make an accurate recommendation.

## INTRODUCTION

53  
54  
55  
56  
57  
58  
59  
60  
61  
62  
63  
64  
65  
66  
67  
68  
69  
70  
71  
72  
73  
74  
75

Colostrum is the first milk that is let down from the mammary gland of a female mammal after giving birth. It is rich in nutrients and immunoglobulins (Metges et al, 2013). Colostrum plays an instrumental role in promoting calf health, growth, and survival rate. In utero, ruminants have no exchange of immunoglobulins across the placenta (Halleran et al, 2017). Therefore, at birth, calves do not have a fully developed immune system and must rely on colostrum for adequate passive transfer of immunoglobulins (Godden, 2008).

In a survey of 1261 US dairy farms, the most common way to evaluate colostrum on-farm was by visual assessment (45% of farms), while less subjective tools such as a colostrometer (hydrometer) or a Brix refractometer were only used on 11% and 4% of farms respectively (USDA, 2016). Hydrometers and refractometers measure colostrum properties that are proxies for IgG content (Heinrichs and Jones, 2016). Although refractometers and colostrometers are tools commonly used on-farm, neither is the gold standard to evaluate colostrum quality. Radial immunodiffusion assays (RID) are the gold standard for evaluating colostrum quality and are the most accurate method of evaluating IgG content. However, RID is an assessment that is performed in a laboratory and requires 18-24 hours to determine results. Therefore, it is not conducive to on-farm use (Bielmann et al, 2010).

Failure of passive transfer occurs when calves fail to absorb adequate amounts of immunoglobulins (Beam et al, 2009). Up to 19.2% of all dairy calves are estimated to suffer from failure of passive transfer (Beam et al, 2009) which has been linked to increased calf morbidity and mortality (Donovan et al, 1997) and a reduction in calf growth rate (Robison et al, 1988). Therefore, if calves are not receiving adequate colostrum, they can become more susceptible to disease.

76           Although colostrum is essential to calf health, it can also be one of the earliest potential  
77 exposures to infectious agents (Brady et al, 2015). Bacteria and pathogens can be transmitted in  
78 colostrum either by direct shedding from the mammary gland or through post-harvest  
79 contamination. (Stewart et al, 2005). Coliform bacteria in colostrum have been associated with  
80 reduced passive transfer of IgG from the intestine into circulation which results in reduced serum  
81 IgG concentrations in the calf (Johnson et al, 2007; Godden et al, 2012). It has also been shown  
82 that feeding low-bacteria colostrum may reduce the occurrence of scours in newborn calves  
83 (Gelsinger et al, 2015). On the other hand, lactic acid bacteria have been shown to have  
84 beneficial health effects and have been proposed as types of probiotics (Ljungh and Wadstrom,  
85 2006). Therefore, having increased levels of lactic acid bacteria present in the colostrum may be  
86 beneficial to calves.

87           Since the health of the calf can be affected by the colostrum it receives, and because  
88 reduced health can result in reduced growth and development, it is important to find ways to  
89 monitor calf health. Calves that become sick may be asymptomatic during the early stages of  
90 illness and may not show clinical signs for several days to over one week (Schaefer et al, 2007).  
91 Monitoring rectal temperatures may be a way to spot illness in its early stages and to evaluate  
92 calf health (Vinson et al, 2014).

93           The aim of this observational study was to evaluate the immunological and bacterial  
94 quality of colostrum fed to dairy calves at the UVM Miller Research Center as well as to  
95 evaluate the potential association between calf health measures and bacterial contamination or  
96 immunoglobulin content of colostrum. It was hypothesized that calves fed colostrum with lower  
97 Brix values and higher bacteria counts would be at increased risk for adverse health events  
98 (rectal temperature above 103°F and reduced average daily gain pre-weaning). It was also

99 hypothesized that there would be a negative correlation between colostrum bacteria counts  
100 (cfu/mL) and colostrum Brix values.

## 101 **MATERIALS AND METHODS**

### 102 *Study design, animal use, and farm colostrum management practices*

103 In a prospective observational study, 12 calves from the UVM-CREAM herd born  
104 between July and September 2018 were enrolled. Animal use procedures were approved under  
105 University of Vermont Institutional Animal Care and Use protocol 18-007. Calves were followed  
106 for 8 weeks of life until each animal's approximate weaning date. Farm practices for colostrum  
107 harvest, testing, storage and feeding were not changed during the course of this study. The farm  
108 personnel (undergraduate students enrolled in the UVM-CREAM program) harvested colostrum  
109 from the first and second milking from cows who calved in. At the time of harvest, the date of  
110 colostrum harvest, cow of origin, and colostrometer reading prior to frozen storage were  
111 recorded. The calves in this study each received two feedings of colostrum, one given within the  
112 first 30 minutes after birth and the second feeding given 8 hours following the first. They were  
113 fed colostrum either fresh from their dam, or colostrum from another dam from thawed-frozen 4-  
114 liter aliquots where the colostrum quality had been determined using a colostrometer  
115 immediately prior to storage in the freezer. The calves were fed using a bottle to ensure that they  
116 each received at least 6 pints of colostrum during each feeding. 50mL aliquots were taken from  
117 the colostrum fed to the calves during the first and second feedings and stored in the freezer at  
118 the Miller Research Center at -20°C until they were picked up and brought to the lab for analysis.  
119 In addition to colostrum collection, body weights were taken at birth and then once per week  
120 until 8 weeks of age and rectal temperatures using a rectal thermometer were taken twice per

121 week until 8 weeks of age. Blood was also drawn from the calves and spun down to collect  
122 serum.

### 123 *Sample analysis*

124       Once taken to the lab, the 50mL aliquots of colostrum were stored in a -20°C freezer.  
125 Each colostrum sample was then thawed in a water bath at 40°C, which is the optimal  
126 temperature for thawing colostrum, prior to being analyzed (Balthazar et al, 2015). Once thawed  
127 and at room temperature, the colostrum was tested using a Brix refractometer to determine the  
128 total sucrose which can be equated to the IgG content of the colostrum. The blood samples from  
129 the calves were also tested for serum IgG levels using a Brix refractometer. A microbiological  
130 analysis of each colostrum sample consisted of plating 1mL of thawed colostrum on 3M  
131 Petrifilm plates including lactic acid bacteria (LAB) plates, coliform count plates (CC), staph  
132 express plates which are selective for *Staphylococcus* bacteria (STX), and aerobic bacteria (AC)  
133 plates. These plates were used to determine the colony counts of each type of bacteria within the  
134 colostrum samples (Moore and Sischo, 2015). The CC and STX plates were incubated for 24  
135 hours, and the LAB and AC plates were incubated for 48 hours. The number of colonies were  
136 counted after incubation and the number of cfus/mL were determined for each sample of  
137 colostrum. Where plates demonstrated overgrowth of bacteria (number of colonies were > 300 or  
138 too numerous to count) 10-fold serial dilutions of samples were analyzed to achieve a countable  
139 plate (around 30-300 colonies). In some cases, the number of colonies on some plates were  
140 estimated using the method from the 3M Petrifilm Interpretation Guide, where a plate count was  
141 obtained by counting colonies within a defined number of sectors in the matrix. Statistical  
142 analysis was performed using the JMP software, including correlation analysis. All continuous  
143 data including the cfu/mL counts as well as the log 10 of the cfu/mL counts were checked for

144 normality using the Shapiro-Wilk Test on the JMP software. The raw cfu counts did not follow a  
145 normal distribution, however, taking the log 10 of the cfu/mL counts improved the normality of  
146 the data. Therefore, the log 10 of cfu/mL counts were used in the correlation analysis. All of the  
147 cfu counts also displayed a bimodal distribution. The residual plots were examined for the data,  
148 which showed random dispersal around the horizontal axis, supporting the fact that a linear  
149 model would be appropriate for the data.

150

## 151 **RESULTS**

### 152 *Colostrum management evaluation*

153 The measurements of colostrum quality taken at the UVM Miller Research Center with  
154 the colostrometer did not all match the measurements taken with the Brix refractometer in the  
155 lab. Of the 18 colostrum samples, 6 of the Brix refractometer readings did not match the  
156 colostrometer readings (Table 1). All 6 of these samples were measured to be “green” with the  
157 colostrometer, which equates to “good” quality. When using a refractometer, a Brix score equal  
158 to or above 22% indicates the colostrum is of good quality (Bielmann et al, 2010). The Penn  
159 State Extension Page states that a Brix value of 22% corresponds to 50mg/mL of IgG which  
160 results in a “green” reading on a colostrometer (Heinrichs and Jones, 2016). Therefore, all of the  
161 colostrum samples in the table that had a “green” colostrometer reading should have a Brix value  
162 that is greater than or equal to 22% which was not true for some of the samples.

### 163 *Colostrum culturing and analysis*

164

165 McGuirk and Collins (2004) have recommended that colostrum fed to calves should  
166 contain a total plate count of fewer than 100,000 cfu/mL and total coliform count of fewer than  
167 10,000 cfu/mL. Of the colostrum samples fed to calves in this study, 9 of the 18 samples



168 analyzed had coliform counts higher than the recommended level of 10,000 cfu/mL (Table 2)  
169 and 9 of the 18 colostrum samples had total plate counts (aerobic counts) higher than the  
170 recommended level of 100,000 cfu/mL (Table 3). Currently there is no widely used threshold for  
171 recommended counts of lactic acid bacteria and *Staphylococcus* species in colostrum fed to  
172 calves. Descriptive analysis of cfu data demonstrated a bimodal distribution of bacteria counts  
173 for all types of bacteria tested including coliform, total aerobic bacteria, lactic acid bacteria, and  
174 *Staphylococcus* species, suggesting there were 2 distinct groups of colostrum samples, those over  
175 1,000,000 (extremely high) and those below 1,000,000 (all others). The mean (stdev) log  
176 coliform cfu count, total aerobic cfu count, lactic acid bacteria count, and *Staphylococcus* counts  
177 of the extremely high colostrum samples were 6.76 (0.9), 8.32 (0.4), 8.03 (0.6) and 7.10 (0.2),  
178 respectively. In contrast, the mean (stdev) log coliform cfu count, total aerobic cfu, lactic acid  
179 bacteria count, and *Staphylococcus* counts of the other samples were 3.53 (0.9), 4.92 (0.4), 4.00  
180 (0.7), and 3.47 (0.8), respectively.

181 Correlation analysis was performed comparing the log bacteria cfu counts to colostrum  
182 Brix % values. The correlation between all of the bacteria counts (coliform count, aerobic count,  
183 lactic acid bacteria count, and Staphylococcal count) and the colostrum Brix values were  
184 significant with p-values of 0.0146, 0.0381, 0.0209, and 0.0364, respectively (Figure 1). The  
185 relationship between the bacteria counts (coliform count, aerobic count, lactic acid bacteria  
186 count, and Staphylococcal count) and the colostrum Brix values all demonstrated a moderate  
187 linear negative correlation as evidenced by the following respective correlation coefficient  
188 values: -0.565, -0.492, -0.539 and -0.496.

189 A correlation analysis was also performed on the sum of the cfu/mL counts for coliform  
190 and lactic acid bacteria of the colostrum. This was a significant relationship with a-p value of  
191 0.0060 and a correlation coefficient of 0.918 showing a strong positive correlation (Figure 2).

### 192 *Effect on calf health*

193 During the course of the study, 3 calves (Jinx, Leo, and Linus) had incidences of having a  
194 fever, or rectal temperature of 103°F or greater for two consecutive days in a row, however, all  
195 calves in the study had at least one day febrile during the 8-week window. Using correlation  
196 analysis, it was determined that a calf having a rectal temperature greater than 103°F for at least  
197 one day within the first 14 days of life affected the average daily gain of the calf and this  
198 relationship was significant ( $p=0.0092$ ). The calves who had a fever during this time period  
199 ended up having a lower average daily gain (ADG) within the first 8 weeks of life (Figure 3).  
200 There was also a significant relationship between the sum of lactic acid bacteria counts of the  
201 colostrum samples fed to the calves and the ADG of the calves over the 8 weeks of the study  
202 ( $p=0.0406$ ) and these variables displayed a moderate positive linear correlation as indicated by a  
203 correlation coefficient of 0.623. This relationship also displayed a relatively weak  $R^2$  value,  
204 0.388 (Figure 4). Although this  $R^2$  value is low, the significant p-value still indicates that this  
205 relationship is significant. The low  $R^2$  value may indicate that cfu counts are not a good predictor  
206 of Brix values of colostrum, however, other factors may be affecting the Brix value not  
207 determined in this study.

208 The quality of the colostrum fed during the first feeding, as measured by the Brix, did not  
209 have a significant relationship with the ADG of the calves ( $p=0.3801$ ). The relationship between  
210 the Brix value of the first feeding of colostrum and the likelihood of the calf developing a fever  
211 within the first 14 days of life was also not significant ( $p=0.2639$ ). There was no significant

212 relationship between the sum of the aerobic counts and a calf developing a fever within the first  
213 14 days of life ( $p=0.1477$ ) or between the total sum of the coliform counts and developing a  
214 fever within the first 14 days of life ( $p=0.0501$ ). However, there was a significant relationship  
215 between the ADG and the sum of the aerobic cfu counts, as well as the ADG and the total sum of  
216 the coliform counts with p-values of 0.0191 and 0.0229 and correlation coefficients of 0.689 and  
217 0.674, respectively . These both show moderate positive linear correlations (Figure 5).

## 218 **DISCUSSION**

### 219 ***Bacterial and immunoglobulin colostrum content***

220 There was a significant relationship between the bacterial count (cfu/mL) of each of the  
221 different types of bacteria and the Brix (%) of the colostrum. All of the bacterial counts had a  
222 moderate negative linear correlation with the Brix colostrum values. In each of the graphs, there  
223 was a group of samples that clustered well above the regression line and had extremely high  
224 cfu/mL counts (Figure 1 A-D). These extremely high cfu data points that had evident grouping in  
225 Figure 1A-D were all from the same samples of colostrum. If these samples that were classified  
226 as having extremely high cfu/mL counts were excluded from the data set, the association  
227 between the bacteria counts and immunoglobulin content changes and there is no longer a  
228 significant correlation between the two variables. Although these few samples are different and  
229 have extremely high cfu counts, they should not be eliminated from the data at this time since  
230 they make up a relatively large portion of the samples collected (approximately 22%) and it is  
231 not clear that they are biological outliers. There was a great amount of variation in the bacterial  
232 counts of different colostrum samples, so these samples with extremely high cfu counts could  
233 just be part of the natural variation seen across samples. Although both bacterial counts and Brix  
234 values are very important ways to evaluate colostrum, the relationship between these two

235 variables has not been explored in the literature. However, some studies have shown that  
236 colostrum fed to calves on dairy farms in New Zealand followed a similar pattern to the  
237 colostrum collected, having sub-optimal Brix values, and high bacteria counts (Denholm et al,  
238 2016). Although it is unclear why there is a significant relationship between the bacterial counts  
239 and the Brix readings for the colostrum, there are likely other factors involved that were not  
240 analyzed in this study that may be causing this.

241 There was a significant relationship between the sum of the coliform cfu/mL counts and  
242 the sum of the lactic acid bacteria cfu/mL counts ( $p=0.0060$ ) of the first two feedings of  
243 colostrum which had a strong linear correlation (Figure 2). These results go against what would  
244 be expected. Lactic acid bacteria have been proposed as a type of probiotic (Ljungh and  
245 Wadstrom, 2006). They have been shown to exert inhibitory effects on the growth of  
246 enteropathogenic bacteria such as *Escherichia coli* (Kawakami et al, 2010). Some coliform  
247 bacteria are enteropathogenic, therefore it was thought that the lactic acid bacteria count and the  
248 coliform count would have a negative relationship as Kawakami et al (2010) have shown. This  
249 may not have been the case in this study since in general, the bacteria counts were very high for  
250 all types of bacteria. Since all of the counts were elevated, this could potentially have appeared  
251 as a false positive relationship. With elevated counts of all types of bacteria including the  
252 enteropathogenic bacteria or coliform bacteria, the lactic acid bacteria present may not have been  
253 able to inhibit these coliform bacteria since there may have been too many coliform bacteria  
254 present for the amount of lactic acid bacteria present to overcome.

### 255 ***Effect on calf health***

256 Guidelines from the University of Wisconsin-Madison state that the normal body  
257 temperature for a bovine is between 100- 102.5°F. A calf that has a fever that exceeds 103°F for

258 one to two days in a row would be considered a sick calf which would typically warrant  
259 treatment (McGuirk, n.d.). It was determined that the relationship between a calf having a  
260 temperature greater than 103°F for at least one day within the first 14 days of life and the average  
261 daily gain of the calf was significant and calves who had a fever during this time period ended up  
262 having a lower average daily gain within the first 8 weeks of life (Figure 3). Only one calf  
263 (Dallas) was treated during the 8-week study due to a high fever (105.9°F). However, he was  
264 treated on day 22 of life which did not interfere with our temperature analysis which examined  
265 temperatures during the first 14 days of life. Calves that are morbid or have a disease such as  
266 respiratory disease have been shown to have a decreased dry matter intake, resulting in decreased  
267 average daily gain (Galyean and Hubbert, 1995). In this study, the exact cause of the increased  
268 temperature in these calves was not explored, but the type of illness they were affected by may  
269 put them at an even higher risk for having a lower average daily gain. For example, if the calves  
270 suffered from a gastrointestinal illness such as cryptosporidiosis, which is a common cause of  
271 diarrhea in calves, they may have a decreased average daily gain since they are not as able to  
272 absorb the nutrients in the food they are receiving.

273         There was a significant, moderate positive correlation between the sum of the lactic acid  
274 bacteria counts of the colostrum in the first two colostrum feedings and the ADG of the calves  
275 over the 8 weeks of the study (Figure 4). In other studies, oral administration of lactic acid  
276 bacteria to calves has resulted in improved body weight gain (Abe et al, 1995) which does  
277 support the results found in this study. One mechanism that may be involved in this is that  
278 probiotics, such as lactic acid bacteria, produce water soluble B vitamins which may improve  
279 nutrient metabolism in the gut, leading to an increased ADG (Roodposhti and Dabiri, 2012).  
280 There was also a significant relationship between the ADG and the total sum of the aerobic

281 counts, as well as the relationship between the ADG and the sum of the coliform counts. This  
282 positive linear relationship is not what would be expected. It was expected that the calves  
283 consuming colostrum with higher bacterial loads would experience decreased ADG. Studies  
284 have shown that increased bacteria present in colostrum can result in decreased serum IgG levels  
285 in calves (Johnson et al, 2007; Godden et al, 2012). This reduced serum IgG linked to increased  
286 bacterial loads could be the result of failure of passive transfer due to lack of immunoglobulins  
287 able to be transferred to the calf, which has been linked to reduced growth rate in calves  
288 (Robison et al, 1988). Although the results of this study suggest the opposite, this could be due to  
289 other factors not considered within this study.

#### 290 ***Recommendation to management***

291 Many of the refractometer readings did not match the colostrometer readings when  
292 evaluating the colostrum, and many of these evaluation disagreements occurred with colostrum  
293 fed during the calf's first feeding, which is the most critical feeding due to the short window in  
294 which passive transfer can occur. Passive transfer of immunoglobulins can only occur during the  
295 first 24 hours following birth, with transfer being optimal in the first 4 hours (Stott et al, 1979).  
296 Feeding colostrum of unknown quality could pose problems since the calves may actually be  
297 receiving lower quality colostrum than originally thought, resulting in potential failure of passive  
298 transfer. One reason for this disagreement between the colostrometer and Brix refractometer  
299 readings could be due to measuring the colostrum at an improper temperature. Colostrometers  
300 are temperature dependent and should be used to measure samples at room temperature. If the  
301 colostrum was measured at a temperature greater than room temperature, the colostrometer  
302 would underestimate the colostrum IgG and if it was measured at a temperature colder than room  
303 temperature, the colostrometer would overestimate the IgG content (Mechor et al, 1991). At the

304 Miller Research Center, there is currently no temperature protocol for using the colostrometer.  
305 One way to improve colostrum evaluation methods on farm would be to update the current  
306 colostrometer protocol to adjust for temperature. Another method which could potentially  
307 improve the evaluation methods would be to switch from using a colostrometer on farm, to a  
308 Brix refractometer. Brix refractometers function independently of colostrum temperature, yet are  
309 still user friendly (Bartier et al, 2015). Studies have shown that Brix refractometers provide a  
310 more accurate assessment as well as offer more precision in terms of repeatability than  
311 hydrometers do when assessing colostrum quality (Bartens et al, 2016).

312         The aerobic and coliform bacterial counts of most colostrum samples were much higher  
313 than the recommended bacterial counts. These high bacterial counts indicate that bacterial  
314 contamination is a serious issue on the farm. This could put the calves consuming this colostrum  
315 at an increased risk of becoming sick. The bacteria can come from either an infected mammary  
316 gland, or fecal contamination from the environment from post-harvest contamination. Studies  
317 have shown that the most significant bacterial contamination occurs during the harvest process.  
318 Bacteria counts were shown to be very low in colostrum samples collected directly from the  
319 udder of cows (Stewart et al, 2005). High levels of bacteria in colostrum are worrisome since  
320 these infectious agents could directly cause diseases such as enteritis or septicemia. It is also  
321 believed that increased levels of bacteria in the small intestine at the time of feeding of colostrum  
322 could interfere with systemic absorption of Ig molecules (Stewart et al, 2005). One possible  
323 mechanism for this could be competition between the intestinal microbes and Ig molecules to  
324 bind to common receptors on intestinal epithelial cells (Staley and Bush, 1985).

325         Feeding clean colostrum of good quality is very important to calf health and therefore  
326 measures should be taken to decrease contamination during the harvest and feeding process.

327 Possible sources of contamination include teat skin, milking cup liners, hoses, or floor buckets  
328 that the colostrum may be collected in (Stewart et al, 2005). In order to decrease the  
329 contamination of colostrum the procedure for sanitizing the udder prior to milking should be  
330 improved. The sanitation procedure of the milking equipment should also be improved, making  
331 sure to sanitize all of the components of the system including buckets and hoses. Methods could  
332 also be developed on farm to evaluate the cleanliness of equipment to see if cleaning procedures  
333 are working well. One tool commonly used in evaluating surface sanitation is an ATP Meter.  
334 Adenosine triphosphate (ATP) is present in all types of organic matter. An ATP meter can detect  
335 ATP levels via ATP bioluminescence which provides an indication of the level of surface  
336 contamination (Moore et al, 2010). This tool can obtain results in real time which would make it  
337 very easy to use in a farm setting. The rapid results that this test shows are also helpful because it  
338 will allow for easy monitoring of sanitation procedures and problems within the regimen can be  
339 identified and corrected quickly. This tool could be used to swab and test collection buckets,  
340 hoses, along with the other milk collection equipment.

#### 341 *Limitations and future prospects*

342 One limitation to evaluating the data was the temperature of the colostrum samples were  
343 not recorded when colostrometer readings were taken at the Miller Research Center. Therefore,  
344 the colostrometer readings were not able to be corrected for temperature. At the Miller Research  
345 Center, in order to make a confident recommendation on how exactly to improve colostrum  
346 evaluation procedures, future studies should be completed analyzing batches of colostrum  
347 samples using both evaluation methods (refractometers and colostrometers) and comparing their  
348 readings to the results obtained by the gold standard method, RID, which has been explored in  
349 previous studies (Bartier et al, 2015). RID is an assessment that is performed in a laboratory and





373 health of the calves. In order to further determine the best colostrum evaluation method for the  
374 Miller Research Center, the colostrum measurement tools discussed in this study could be  
375 directly tested against the gold standard, RID. Further studies could also be performed to  
376 determine the exact sources of contamination and pathogens present in the environment at the  
377 farm that could potentially compromise the health of the calves. Additional studies with a larger  
378 sample size should also be carried out to re-evaluate the relationship between bacterial counts  
379 and colostrum IgG content along with average daily gain to get more conclusive results.

380

381

382

383

384

385

386

387

388

389

390

391

392

393

394

395

396  
397  
398  
399  
400  
401  
402  
403  
404  
405  
406  
407  
408  
409  
410  
411  
412  
413  
414  
415  
416  
417  
418

## ACKNOWLEDGMENTS

I would like to thank Dr. John Barlow for all of his advice and guidance throughout this process. I would like to acknowledge Charlotte Callon for assisting with sample collection as well as with some of the experimental procedures in the lab. I would also like to acknowledge the CREAMers at the UVM Miller Research Center for assisting in colostrum sample collection. Furthermore, I would like to thank Dr. Norman Purdie and Dr. Julie Smith for evaluating my thesis and contributing revisions to improve my manuscript and statistical analysis of the data. Lastly, I would like to thank the Honors College, and the entire DUR committee for granting me this opportunity.

## REFERENCES

- 419
- 420 Abe, F., N. Ishibashi, and S. Shimamura. 1995. Effect of administration of bifidobacteria and  
421 lactic acid bacteria to newborn calves and piglets. *J Dairy Sci* 78(12):2838-2846.
- 422 Arsenopoulos, K., A. Theodoridis, and E. Papadopoulos. 2017. Effect of colostrum quantity and  
423 quality on neonatal calf diarrhoea due to *Cryptosporidium* spp. infection. *Comp Immunol*  
424 *Microbiol Infect Dis* 53:50-55.
- 425 Balthazar, E., E. Doligez, O. Leray, and Y. Le Cozler. 2015. A comparison of thawing methods  
426 on IgG1 concentration in colostrum of dairy cows. Vol. 166.
- 427 Bartens, M. C., M. Drillich, K. Rychli, M. Iwersen, T. Arnholdt, L. Meyer, and D. Klein-Jöbstl.  
428 2016. Assessment of different methods to estimate bovine colostrum quality on farm.  
429 *New Zealand Veterinary Journal* 64(5):263-267.
- 430 Bartier, A. L., M. C. Windeyer, and L. Doepel. 2015. Evaluation of on-farm tools for colostrum  
431 quality measurement. *Journal of Dairy Science* 98(3):1878-1884.
- 432 Beam, A. L., J. E. Lombard, C. A. Koprak, L. P. Garber, A. L. Winter, J. A. Hicks, and J. L.  
433 Schlater. 2009. Prevalence of failure of passive transfer of immunity in newborn heifer  
434 calves and associated management practices on US dairy operations. *Journal of Dairy*  
435 *Science* 92(8):3973-3980.
- 436 Biemann, V., J. Gillan, N. R. Perkins, A. L. Skidmore, S. Godden, and K. E. Leslie. 2010. An  
437 evaluation of Brix refractometry instruments for measurement of colostrum quality in  
438 dairy cattle. *Journal of Dairy Science* 93(8):3713-3721
- 439 Brady, M. P., S. M. Godden, and D. M. Haines. 2015. Supplementing fresh bovine colostrum  
440 with gut-active carbohydrates reduces passive transfer of immunoglobulin G in Holstein  
441 dairy calves. *Journal of Dairy Science* 98(9):6415-6422.

442 Denholm, K. S., J. C. Hunnam, E. L. Cuttance, and S. McDougall. 2017. Associations between  
443 management practices and colostrum quality on New Zealand dairy farms. *New Zealand*  
444 *Veterinary Journal* 65(5):257-263.

445 Donovan, G. A., I. R. Dohoo, D. M. Montgomery, and F. L. Bennett. 1998. Associations  
446 between passive immunity and morbidity and mortality in dairy heifers in Florida, USA.  
447 *Preventive Veterinary Medicine* 34(1):31-46.

448 Galyean, ML, Perino LJ, Duff, GC. 1999. Interaction of cattle health/immunity and nutrition. *J*  
449 *Anim Sci.*;77:1120-1134.

450 Gelsinger, S. L., C. M. Jones, and A. J. Heinrichs. 2015. Effect of colostrum heat treatment and  
451 bacterial population on immunoglobulin G absorption and health of neonatal calves.  
452 *Journal of Dairy Science* 98(7):4640-4645.

453 Godden, S. 2008. *Colostrum Management for Dairy Calves*. Vol. 24.

454 Godden, S. M., D. J. Smolenski, M. Donahue, J. M. Oakes, R. Bey, S. Wells, S. Sreevatsan, J.  
455 Stabel, and J. Fetrow. 2012. Heat-treated colostrum and reduced morbidity in preweaned  
456 dairy calves: Results of a randomized trial and examination of mechanisms of  
457 effectiveness. *Journal of Dairy Science* 95(7):4029-4040.

458 Halleran, J., H. J. Sylvester, and D. M. Foster. 2017. Short communication: Apparent efficiency  
459 of colostral immunoglobulin G absorption in Holstein heifers. *Journal of Dairy Science*  
460 100(4):3282-3286.

461 Heinrichs, J., & Jones, C. M. 2016. *Colostrum Management Tools: Hydrometers and*  
462 *Refractometers*. N.p.: PennState Extension.

463 Heinrichs, J., & Jones, C. M. 2016. *Monitoring Dairy Heifer Growth*. N.p.: PennState Extension.

464 Johnson, J. L., S. M. Godden, T. Molitor, T. Ames, and D. Hagman. 2007. Effects of Feeding

465 Heat-Treated Colostrum on Passive Transfer of Immune and Nutritional Parameters in  
466 Neonatal Dairy Calves. *Journal of Dairy Science* 90(11):5189-5198.

467 Kawakami, S., Yamada, T., Nakanishi, N., & Cai, Y. 2010 . Feeding of Lactic Acid Bacteria and  
468 Yeast on Growth and Diarrhea of Holstein Calves. N.p.: *Journal of Animal and*  
469 *Veterinary Advances* 9 (11): 1112-1114.

470 Ljungh, A. and T. Wadstrom. 2006. Lactic acid bacteria as probiotics. *Current issues in intestinal*  
471 *microbiology* 7(2):73-89.

472 McGuirk, S. M. n.d. *Managing the Young Calf*. N.p.: University of Wisconsin-Madison School  
473 of Veterinary Medicine.

474 McGuirk, S. n.d. *Sick Calf Protocols*. N.p.: Food Animal Production Medicine University of  
475 Wisconsin-Madison.

476 McGuirk, S. M. and M. Collins. 2004. Managing the production, storage, and delivery of  
477 colostrum. *The Veterinary clinics of North America. Food animal practice* 20(3):593-  
478 603.

479 Mechor, G., Y. Grohn, and R. Van Saun. 1991. Effect of Temperature on Colostrometer  
480 Readings for Estimation of Immunoglobulin Concentration in Bovine Colostrum. Vol.  
481 74.

482 Metges, C. C., J. Flor, J. Steinhoff-Wagner, U. Schönhusen, and H. M. Hammon. 2013.  
483 LACTATION BIOLOGY SYMPOSIUM: Role of colostrum and colostrum components  
484 on glucose metabolism in neonatal calves<sup>1,2</sup>. *Journal of Animal Science* 91(2):685-695.

485 Moore, Dale A., and William M. Sischo. 2015. *Use of Petrifilm for Milk or Colostrum Total*  
486 *Plate and Coliform Counts*. USDA.

487

488 Moore, G., D. Smyth, J. Singleton, and P. Wilson. 2010. The use of adenosine triphosphate  
489 bioluminescence to assess the efficacy of a modified cleaning program implemented  
490 within an intensive care setting. *American Journal of Infection Control* 38(8):617-622.

491 Robison, J. D., G. H. Stott, and S. K. DeNise. 1988. Effects of Passive Immunity on Growth and  
492 Survival in the Dairy Heifer<sup>1, 2</sup>. *Journal of Dairy Science* 71(5):1283-1287.

493 Roodposhti, P. M. and N. Dabiri. 2012. Effects of probiotic and prebiotic on average daily gain,  
494 fecal shedding of *Escherichia coli*, and immune system status in newborn female calves.  
495 *Asian-Australasian journal of animal sciences* 25(9):1255-1261.

496 Schaefer, A. L., N. J. Cook, J. S. Church, J. Basarab, B. Perry, C. Miller, and A. K. W. Tong.  
497 2007. The use of infrared thermography as an early indicator of bovine respiratory  
498 disease complex in calves. *Research in Veterinary Science* 83(3):376-384.

499 Staley, T. E. and L. J. Bush. 1985. Receptor Mechanisms of the Neonatal Intestine and Their  
500 Relationship to Immunoglobulin Absorption and Disease<sup>1, 2</sup>. *Journal of Dairy Science*  
501 68(1):184-205.

502 Stewart, S., S. Godden, R. Bey, P. Rapnicki, J. Fetrow, R. Farnsworth, M. Scanlon, Y. Arnold, L.  
503 Clow, K. Mueller, and C. Ferrouillet. 2005. Preventing Bacterial Contamination and  
504 Proliferation During the Harvest, Storage, and Feeding of Fresh Bovine Colostrum.  
505 *Journal of Dairy Science* 88(7):2571-2578.

506 Stott, G. H., D. B. Marx, B. E. Menefee, and G. T. Nightengale. 1979. Colostral immunoglobulin  
507 transfer in calves I. Period of absorption. *J Dairy Sci* 62(10):1632-1638.

508 Stott, G. H., D. B. Marx, B. E. Menefee, and G. T. Nightengale. 1979. Colostral immunoglobulin  
509 transfer in calves. III. Amount of absorption. *J Dairy Sci* 62(12):1902-1907.

510

511 Tyler, J. W., D. D. Hancock, S. E. Wiksie, S. L. Holler, J. M. Gay, and C. C. Gay. 1998. Use of  
512 Serum Protein Concentration to Predict Mortality in Mixed-Source Dairy Replacement  
513 Heifers. *Journal of Veterinary Internal Medicine* 12(2):79-83.

514 Research Animal Resources and Compliance. (n.d.). Normative Data for Cattle. University of  
515 Wisconsin-Madison.

516 Urie, N. J., J. E. Lombard, C. B. Shivley, C. A. Koprak, A. E. Adams, T. J. Earleywine, J. D.  
517 Olson, and F. B. Garry. 2018. Preweaned heifer management on US dairy operations:  
518 Part V. Factors associated with morbidity and mortality in preweaned dairy heifer calves.  
519 *J Dairy Sci* 101(10):9229-9244

520 USDA. 2016. Dairy 2014, “Dairy Cattle Management Practices in the United States,  
521 2014”USDA–APHIS–VS–CEAH–NAHMS. Fort Collins, CO#692.0216

522 Vinson, M. C., R. C. Ketring, W. D. George, R. W. Godfrey, and S. T. Willard. 2014.  
523 Relationship among eye and muzzle temperatures measured using digital infrared thermal  
524 imaging and vaginal and rectal temperatures in hair sheep and cattle1. *Journal of Animal*  
525 *Science* 92(11):4949-4955.

526 Weaver, D. M., J. W. Tyler, D. C. VanMetre, D. E. Hostetler, and G. M. Barrington. 2000.  
527 Passive Transfer of Colostral Immunoglobulins in Calves. *Journal of Veterinary Internal*  
528 *Medicine* 14(6):569-577.

529 Zanton, G. I. and A. J. Heinrichs. 2005. Meta-Analysis to Assess Effect of Prepubertal Average  
530 Daily Gain of Holstein Heifers on First-Lactation Production\*. *Journal of Dairy Science*  
531 88(11):3860-3867.

532



533 **Table 1.** Colostrum sample evaluation results and comparison of colostrometer and  
 534 refractometer readings of colostrum fed to calves during the first and second feedings. The  
 535 highlighted rows indicate that there was not agreement between the colostrometer and Brix  
 536 refractometer readings. Data that has been entered as “n/a” indicates missing values.

Calf Name	Colostrum Feeding Number	Colostrum Brix %	Colostrometer Reading	Agreement between evaluation methods
Lenox	1	20.7	green	no
Phoebe	1	22	green	yes
Louise	1	24.2	green	yes
Sadie	1	21.5	green	no
Leo	1	23	green	yes
lavender	1	13	green	no
Linus	1	24	green	yes
Jinx	1	16.5	red	yes
Lennox	2	20	green	no
Phoebe	2	21.9	green/red	no
Sadie	2	22	green	yes
Louise	2	15.5	n/a	n/a
lavender	2	18.5	green	no
Dolores	2	21	yellow	yes
Jemima	2	12.5	n/a	n/a
goose	2	14	yellow/red	yes
Linus	2	26.5	n/a	n/a
Jinx	2	16	red	yes

537  
 538  
 539  
 540  
 541  
 542  
 543  
 544  
 545

546 **Table 2.** Coliform bacteria count and Brix refractometer readings of colostrum fed to calves in  
 547 the first and second feedings. Highlighted values indicate the cfu/mL is greater than the  
 548 recommended industry level of 10,000 cfu/mL. Data that has been entered as “n/a” indicates  
 549 missing values.

Cow that produced colostrum	Calf that received	Coliform cfu/mL	Colostrum Brix %	Feeding Number
Leena	Lennox	870	20.7	1
Leena	Lennox	34000	20	2
primrose	Phoebe	280	22	1
Dakota/Jubilee	Phoebe	1100000	21.9	2
Dakota	Sadie	3000	21.5	1
Dakota	Sadie	10500	22	2
Leena	Louise	340	15.5	2
Genevieve	Louise	1200	24.2	1
Leena/shadow	Lavender	13900000	13	1
Dakota	Lavender	670000	18.5	2
Genevieve	Leo	3600	23	1
n/a	Dolores	86000	21	2
Lakota	Jemima	8700	12.5	2
Genevieve/shadow	Goose	4600000	14	2
Lily	Linus	1000	24	1
n/a	Linus	390	26.5	2
Jubilee	Jinx	64000	16.5	1
Jubilee	Jinx	137000000	16	2

550

551

552

553

554

555

556

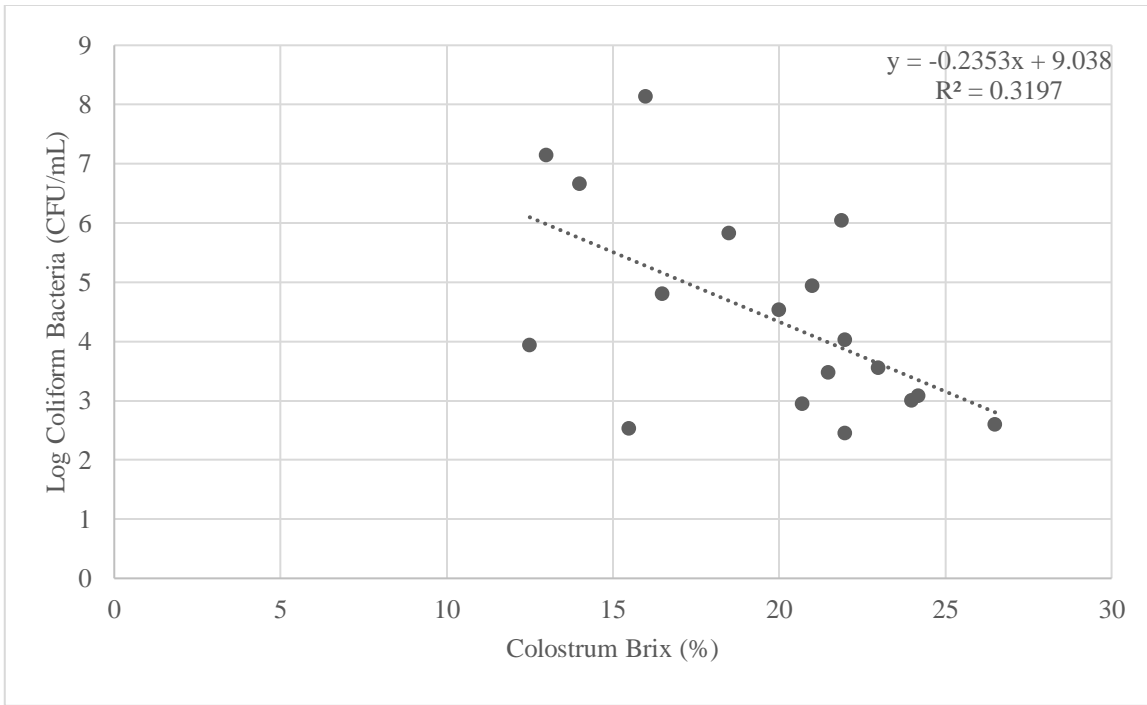
557

558

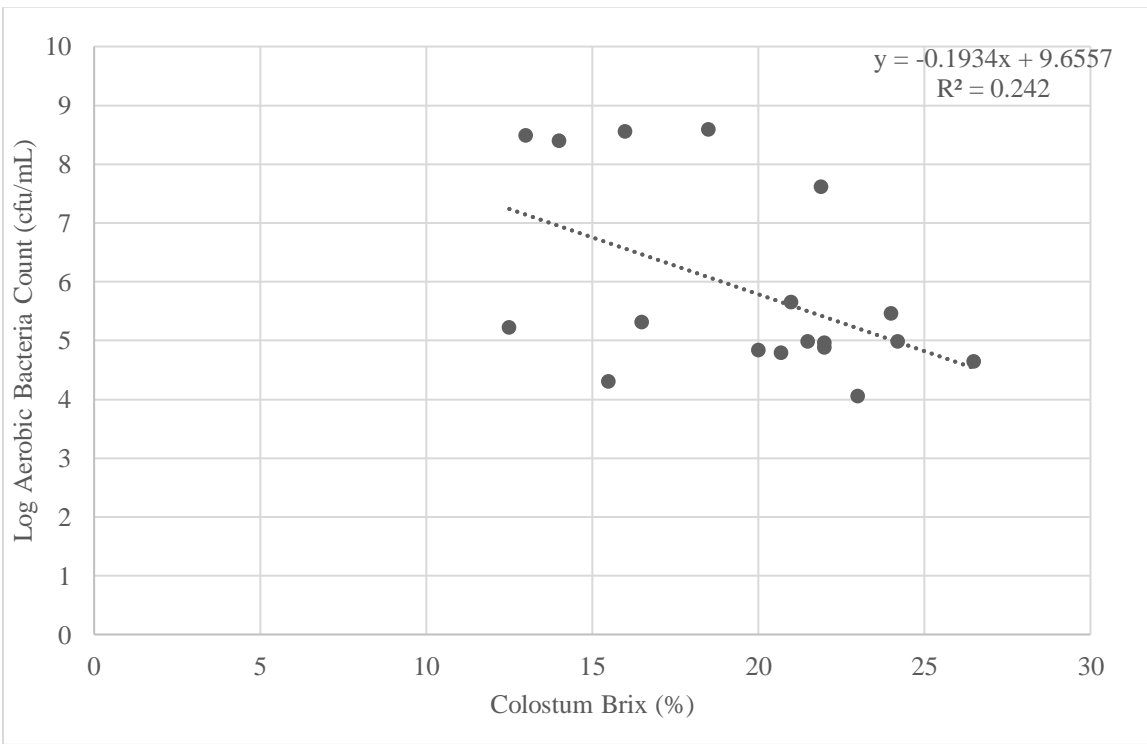
559 **Table 3.** Aerobic bacterial counts and Brix refractometer readings of colostrum fed to calves in  
 560 the first and second feedings. Highlighted values indicate the cfu/mL is greater than the  
 561 recommended industry level of 100,000 cfu/mL. Data that has been entered as “n/a” indicates  
 562 missing values.

Cow that produced colostrum	Calf that received	Aerobic Count cfu/mL	Colostrum Brix %	Feeding Number
Leena	Lennox	60000	20.7	1
Leena	Lennox	67000	20	2
primrose	phoebe	89000	22	1
Dakota/jubilee	phoebe	40000000	21.9	2
Dakota	Sadie	95000	21.5	1
Dakota	Sadie	74000	22	2
Leena	Louise	19500	15.5	2
Genevieve	Louise	95000	24.2	1
Genevieve	Leo	11000	23	1
Leena/shadow	lavender	30000000	13	1
Dakota	lavender	38000000	18.5	2
n/a	Dolores	440000	21	2
Lakota	Jemima	164000	12.5	2
Genevieve/shadow	goose	24400000	14	2
Lily	Linus	286000	24	1
n/a	Linus	43000	26.5	2
Jubilee	Jinx	199000	16.5	1
Jubilee	Jinx	35700000	16	2

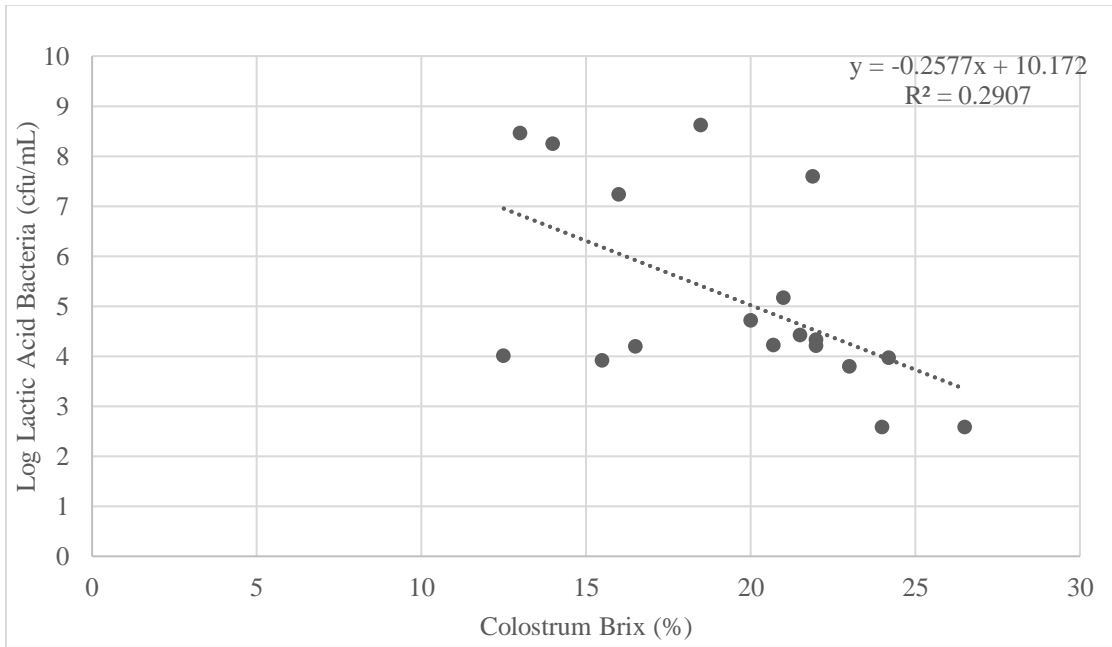
563  
 564  
 565  
 566  
 567  
 568  
 569  
 570  
 571



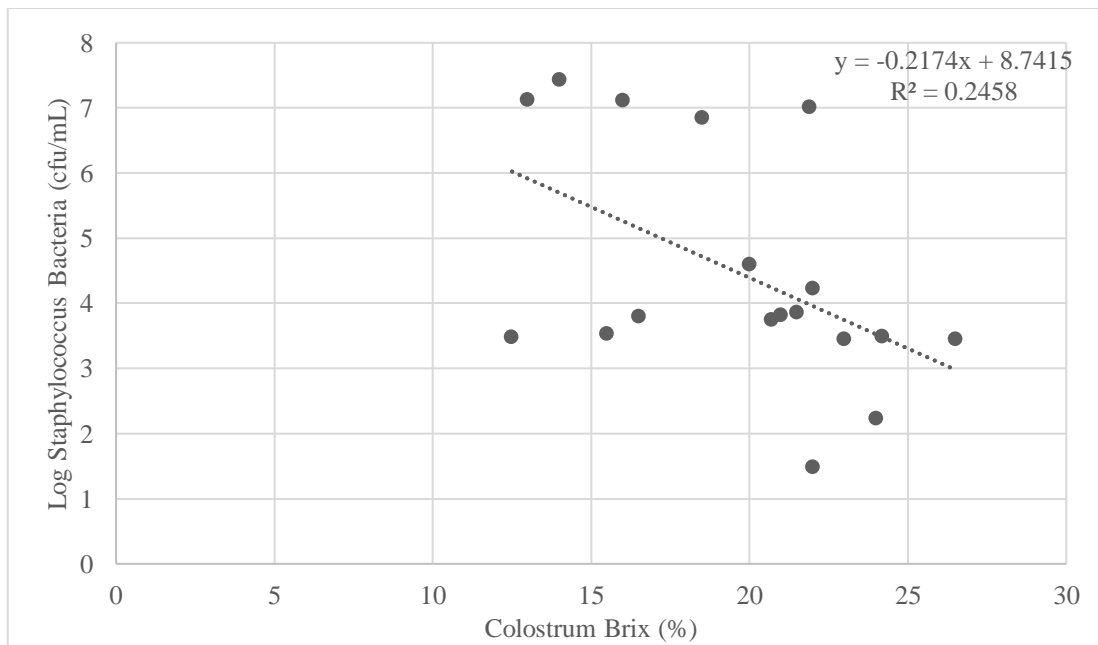
572 A)



573 B)



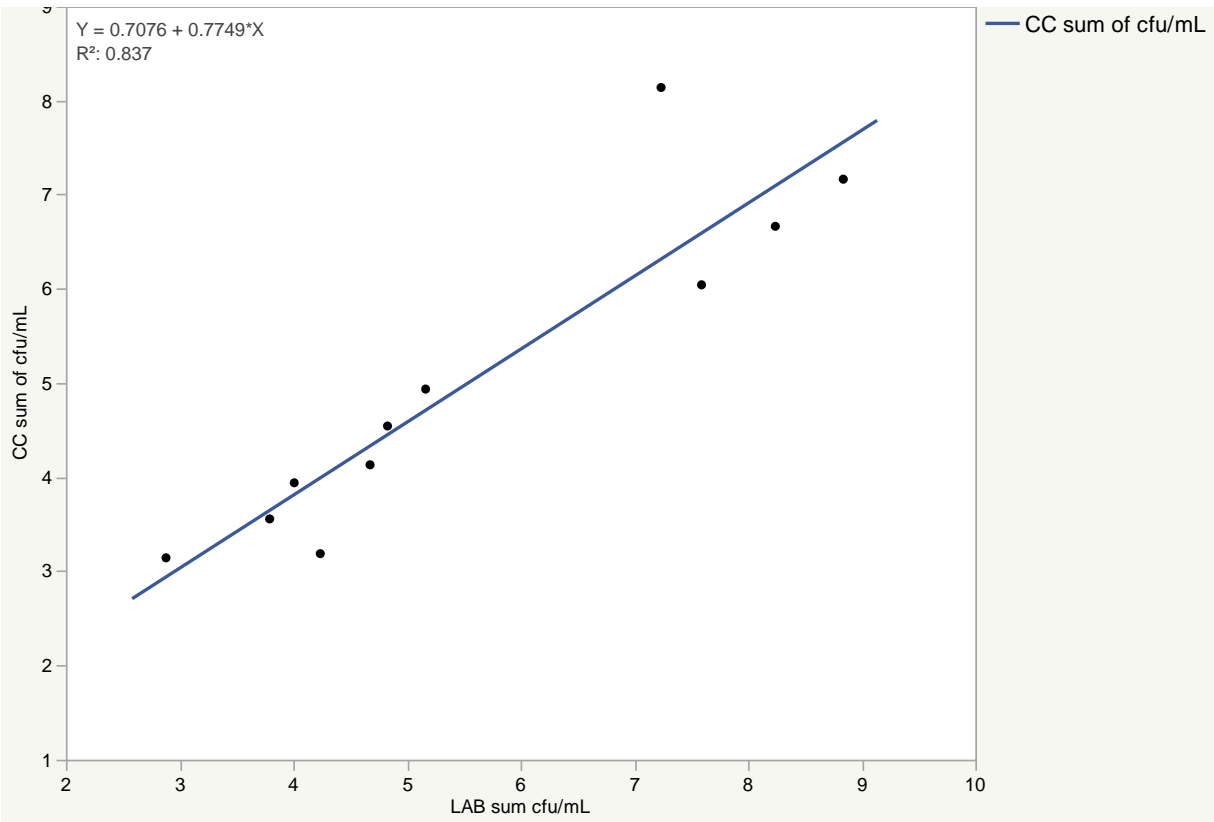
574 C)



575 D)

576 **Figure 1.** Relationship between the four types of bacterial counts —coliform bacteria count (A),  
 577 aerobic bacteria count (B), lactic acid bacteria count (C) and *Staphylococcus* bacteria count (D)  
 578 — and the colostrum Brix value for the colostrum samples fed during both the first and second  
 579 feedings to the calves.

580

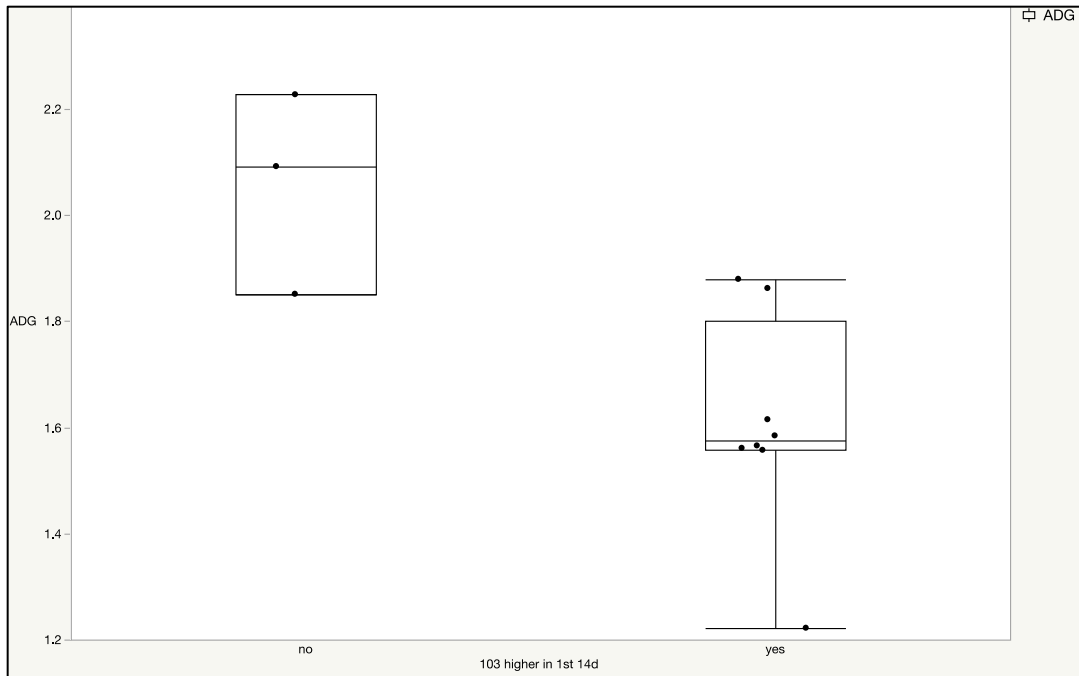


581

582 **Figure 2.** Relationship between the sum of the coliform bacteria counts (CC) of the first and  
 583 second feeding and the sum of the lactic acid bacteria (LAB) counts of the first and second  
 584 feeding.

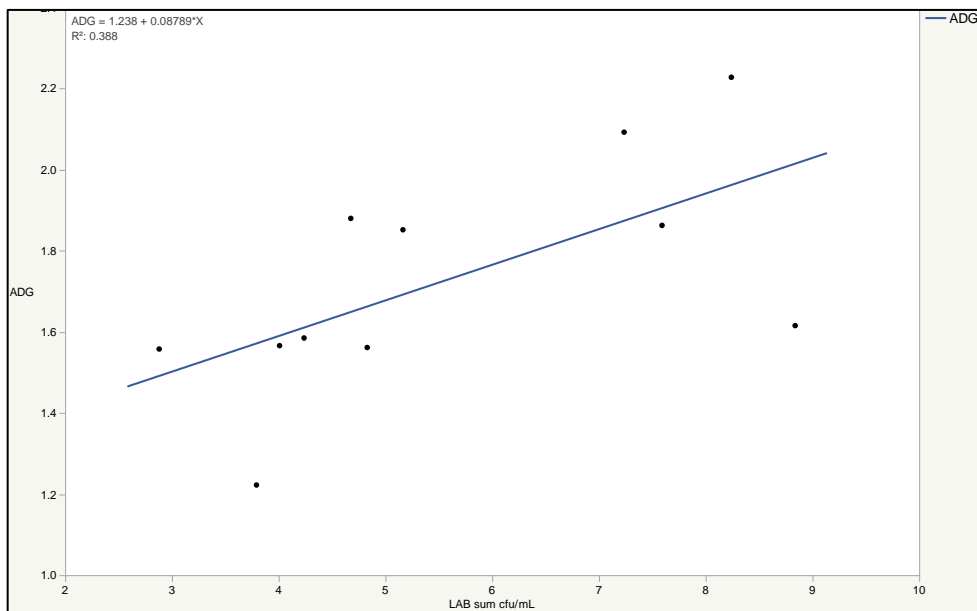
585

586



587

588 **Figure 3.** Relationship between a calf developing a fever (rectal temperature above 103°F)  
 589 within the first 14 days of life and average daily gain (ADG)  $p=0.0092$ . Calves who developed a  
 590 fever during the first 14 days of life had a decreased ADG.

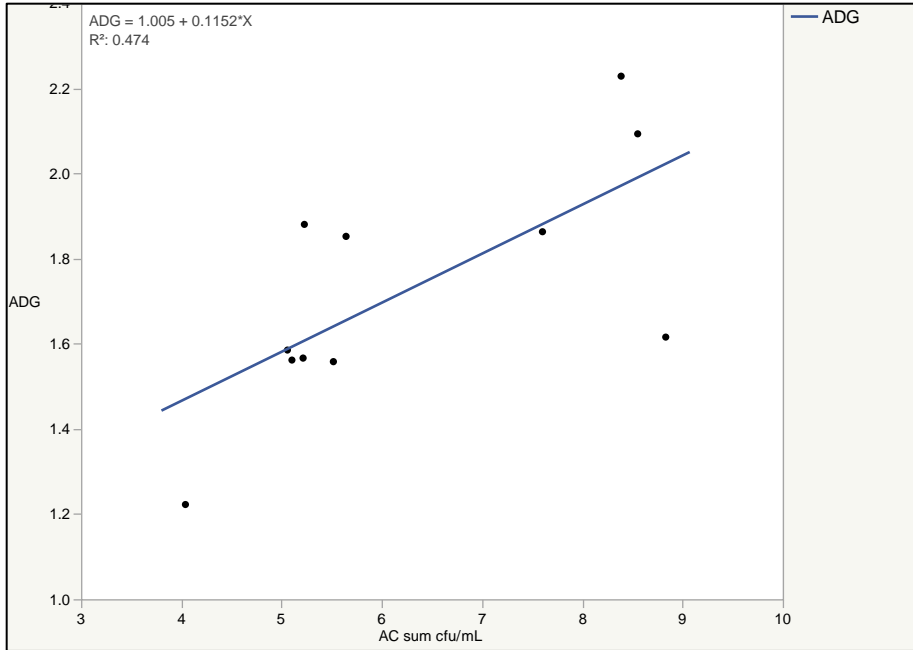


591

592 **Figure 4.** Relationship between the average daily gain (ADG) of the calves and the sum of the  
 593 lactic acid bacteria (LAB) present within the colostrum fed during the first and second feedings.

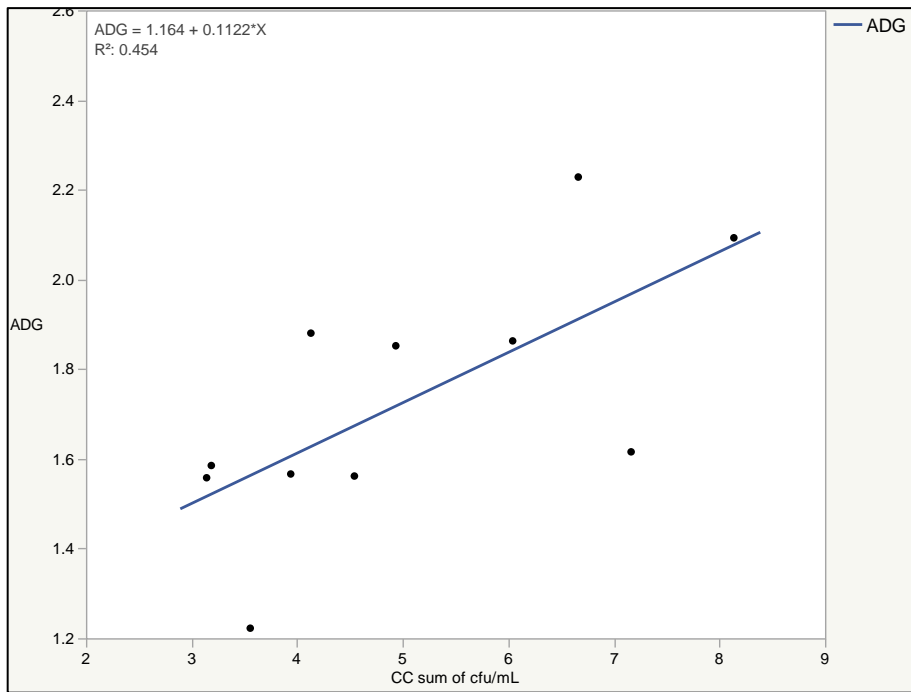
594

A.



595

B.



596 **Figure 5.** Relationship between the average daily gain (ADG) of the calves and the sum of the  
597 aerobic count (AC) bacteria present in both the colostrum from the first and second feedings (A)  
598 and the relationship between the average daily gain (ADG) of the calf and the sum of the  
599 coliform (CC) bacteria present within the colostrum from the first and second feedings (B).



