Effect of feeding single-dam or pooled colostrum on maternally derived immunity in dairy calves

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ABSTRACT

The role of colostrum management in providing adequate immunological protection to neonatal calves has been widely investigated, and thresholds for colostrum quality, as well as optimum volume and timing for colostrum feeding have been established. However, limited information is available on the effect of colostrum source (single dam or pooled) on passive immunity, as well as subsequent antibody survival in the calf. This study aimed to assess the effect of feeding single-dam colostrum (own and other dam) or pooled colostrum on transfer of passive immunity, and also investigate the rate of depletion of disease-specific antibodies among dairy calves. In total, 320 cows and 119 dairy heifer calves were enrolled in the study. Calves were blood-sampled immediately after birth and received either own-dam, other-dam, or pooled colostrum. Calves were blood-sampled at 24 h to assess serum IgG concentrations and at monthly intervals thereafter to document disease-specific antibody survival. Mean colostrum IgG concentration was higher for other-dam treatment group, whereas own-dam and pooled treatments were similar. For all treatment groups, the mean IgG concentration was >80 mg/mL, exceeding the quality threshold of 50 mg/mL. Mean calf serum IgG concentration was lower for calves fed pooled colostrum compared with those that received colostrum from a single cow. There was a negative association with 24-h serum IgG and calf birth bodyweight; calves <30 kg at birth had the highest 24-h serum IgG concentration. Survival of antibodies to bovine viral diarrhea, *Salmonella* infection, leptospirosis, bovine parainfluenza 3 virus, bovine respiratory syncytical virus, rotavirus, and coronavirus was not associated with colostrum source; however, antibodies to infectious bovine rhinotracheitis had a greater period of survival among calves fed own-dam colostrum. We found that feeding single-dam colostrum

can thus improve calf immunity through increased serum IgG levels and antibody survival rates. Furthermore, we hypothesize that immune exclusion may occur with pooled colostrum; therefore, providing pooled colostrum may still be a good practice as long as it can be ensured that enough antibodies are absorbed into the blood stream to deal with pathogens calves may encounter because different dams may have antibodies against different strains of viruses and bacteria, yielding cross protection.
Key words: survival, health, immunoglobulin, heifer, birthweight

INTRODUCTION

Neonatal bovines are dependent on transfer of passive immunity (**TPI**) to provide protection from infectious diseases in early life (Godden, 2008). Passive immunity can be acquired from colostrum, the first mammary secretion postparturition, which contains a wide range of immunological factors, such as immunoglobulins, cytokines, and antimicrobials (e.g., lactoferrin and lactoperoxidase; McGrath et al., 2016; Stelwagen et al., 2009). Immunoglobulins, also known as antibodies, are produced in the maternal bloodstream by lymphocytes in response to foreign antibodies, and they account for over 70% of the total protein content in colostrum (Larson, 1992). Immunoglobulins present in colostrum can be divided into the following 5 subclasses: IgM, IgA, IgG, IgE, and IgD. The most abundant immunoglobulin subclass in colostrum is IgG, which has the greatest specificity and affinity for target pathogens (Hurley, 2003). As a result, colostrum quality is determined mainly by IgG concentration, and a concentration of ≥50 mg/mL is considered as good quality (Kruse, 1970).

In calves, failure to achieve an adequate level of passive immunity is identified as a serum IgG concentration of <10 mg/mL at 24 h old, resulting in increased susceptibility to disease as well as an increased risk of mortality (Weaver et al., 2000; Stilwell and Carvalho, 2011; Chigerwe et al., 2015). Achieving adequate TPI

depends predominantly on the following 3 main factors: (1) the volume of colostrum provided to the calf (Godden et al., 2009; Conneely et al., 2013), (2) the timing within which colostrum is received by the calf after birth (Michanek et al., 1989; Morin et al., 1997), and (3) the quality of the colostrum (Beam et al., 2009).

When feeding colostrum, the recommended practice is to provide each calf with colostrum from their own dam once the colostrum is of sufficient quality (Godden, 2008). In Ireland, however, studies have found that feeding of nonmaternal colostrum is commonly practiced. Over 30% of farmers provide calves with colostrum that has been pooled from several cows, whereas almost 10% of farms provide colostrum produced by another cow (other dam; Barry et al., 2019). Feeding pooled colostrum, but also single-dam colostrum, can have negative implications for TPI when colostrum quality is low (Weaver et al., 2000). Direct comparisons between passive immunity of calves fed colostrum from maternal and nonmaternal colostrum (other dam, pooled), as well as the effects on antibody depletion, are required. Additionally, there is limited information on any negative implications of feeding either pooled or other-dam colostrum in situations where colostrum quality is relatively high. For example, within a herd that has a high mean value for colostrum IgG, does pooling colostrum still elicit a reduction in colostrum quality and present a risk to TPI (Godden, 2008)? Also, there remains a gap in the knowledge on which source of colostrum provides the most immunological protection to calves, not only for passive immunity, but also for disease-specific immunity during the first year of life. The range of disease-specific antibodies in colostrum will vary depending on immune function of the dam and exposure to pathogens; therefore, we hypothesized that pooling colostrum from several cows could provide more comprehensive immunity to calf as they receive antibodies to a wider range of diseases.

Antibodies acquired from colostrum have a limited survival time once present in the calf's circulatory system. The rate of antibody depletion can vary based on the animal, due to either individual differences or the level of passive immunity received, with high levels of maternally derived immunity resulting in a slower rate of depletion (Chase et al., 2008). When colostrum quality is high, and high rates of TPI are achieved, much slower rates of antibody depletion are expected. Differences in maternally derived immunity at the animal level could also negatively affect vaccine efficacy as studies have found no increase in vaccine-derived antibody titers when vaccines were administered in the presence of maternally derived antibodies to the same disease (Ellis et al., 1996; Kirkpatrick et al., 2001). As a result, vaccination protocols would need to be modified accordingly to avoid any window for susceptibility to disease, which calves may experience in the absence of vaccine-derived immunity.

By investigating the effects of feeding pooled or single-dam colostrum (own dam or other dam), and assessing passive immunity in addition to the depletion rate of disease-specific immunity, improved protocols for colostrum feeding and vaccine administration could be developed. This could enhance calf immunity, ultimately contributing to improved welfare among calves. Therefore, this study aimed to assess the effect of feeding single-dam colostrum, maternal or nonmaternal colostrum, or colostrum pooled from multiple cows of known immunological profiles on TPI and disease-specific immunity of calves, and also to investigate the rate of depletion of disease-specific antibodies among dairy calves.

MATERIALS AND METHODS

This study was carried out at Teagasc, Animal & Grassland Research and Innovation Centre (Moorepark, Fermoy, Co. Cork, Ireland) between January and October of 2016 and 2018. Ethical approval was received from the Teagasc Animal Ethics Committee (TAEC101/2015), and procedure authorization was granted by the Irish Health Products Regulatory Authority (AE19132/P044). Experiments were undertaken in accordance with the Irish Cruelty to Animals Act (Irish Statute Book, 1876, 2005) and the European Community Directive 86/609/EC.

In total, 320 cows and 120 dairy breed heifer calves were enrolled in the study. To determine the number of animals required, a sample size calculation was completed using IgG data from previous studies completed at Teagasc Moorepark (Conneely et al., 2014). Calculations indicated that to detect a significance of $P < 0.05$ at a power of 0.80, a minimum of 40 calves per treatment were required to detect a difference of 7 mg/mL in a 2-sample t-test. It should be noted that these calculations do not take account for the effects of doing mixed models or including numerous covariates in statistical models, all of which include the effect of reducing the effective sample size. The study was repeated over 2 yr to ensure data from a sufficient number of animals were available. In both January 2016 and January 2018, 160 cows (120 multiparous, 40 primiparous, each year) were enrolled. Cows were selected based on expected calving date (January–February). Blood sampling of cows was conducted in early January to ensure the immunity profile of cows was captured close to parturition and that serological results were received

before the commencement of calving. Blood samples were refrigerated at 4°C for <24 h, up to the point of dispatch to Enfer Scientific (Enfer Group), where a range of pathogen-specific serological ELISA were conducted. Pathogens investigated included bovine herpes virus-1 (**BHV-1**; IDEXX, BHV gE antibody test), bovine viral diarrhea virus (**BVDV**; IDEXX BVD p80 test kit), *Leptospira borgpetersenii* serovar Hardjo and *Leptospira interrogans* serovar Hardjo (Linnodee *Leptospira* Hardjo ELISA kit), *Salmonella enterica* serovar Dublin and *Salmonella enterica* serovar Typhimurium (PrioCHECK *Salmonella* Ab bovine), bovine rotavirus (Bio-X Bovine rotavirus ELISA), bovine coronavirus (SVANOVIR Bovine Coronavirus antibody test), bovine parainfluenza 3 virus (**PI-3**; SVANOVIR PIV3 Ab Bovine Parainfluenza virus type 3), and bovine respiratory syncytical virus (**BRSV**; SVANOVIR Bovine Respiratory Syncytical virus antibody test). Based on serum antibody results, cows were classified as having ELISA-positive or ELISA-negative serological status to each pathogen. To maintain health practices within the research herd and prevent an effect on the study, vaccination protocols for cows and calves remained identical across both years of study. Although 320 cows were blood-sampled for inclusion in the study, 119 eventually produced calves and colostrum compatible with the planned logistics of the study. Reasons for ineligibility of cows in the study included poor colostrum quality, low volume of colostrum produced, and birth to a stillborn or male calf. Of the cows used in the study, 46 were first parity, 25 were second parity, 25 were third parity, and 21 were fourth or greater parity. With regard to breed, 62 were Holstein-Friesian (**HF**) and 57 were HF crossed with another dairy breed (i.e., Jersey).

Using only heifer calves from cows that had been immunologically profiled, a complete randomized block design was applied. A total of 119 calves (59 in 2016, and 60 in 2018) were enrolled in the study. Of these, 88 were HF and 31 were HF × Jersey. Calves were balanced based on birthweight, breed, and date of birth. They were then assigned to 1 of the 3 treatments where they received (1) maternal colostrum (**MC**), (2) non-maternal colostrum (**NMC**), or (3) pooled colostrum (**PC**). The PC was prepared by combining colostrum at equal ratios from several cows (2 cows in 2016, and 4 cows in 2018). Within each of the treatment groups (MC, NMC, PC), calves were balanced based on the serological profile of the cow(s) supplying the colostrum. In terms of colostrum IgG concentration, efforts were made to ensure that similar quality colostrum was provided across each of the treatment groups. Colostrum quality was determined cow-side by Brix refractometry to satisfy the necessity of providing calves with colostrum in a timely manner (Bielmann et al., 2010).

Calf Management

All calving events were supervised by trained and experienced personnel. Following birth, calves were immediately separated from the dam as a standard biosecurity measure. Two identically numbered plastic ear tags were applied to each calf following birth, as required by the Irish Department of Agriculture, Food and the Marine. Each calf was weighed (TruTest XR3000, Tru-test Limited), and 10% iodine was applied to the remnants of the umbilical cord and the navel area. Calves were then placed in indoor individual pens (0.8 m × 1.2 m in size). Colostrum was collected either immediately post-calving using a single portable milking unit, or in the milking parlor when calving occurred within 1 h of a scheduled milking (0700 or 1430 h). Each calf received 8.5% of its birth BW in colostrum within 2 h of birth via bottle and teat or an esophageal tube feeder in the event the calf would not drink voluntarily. Colostrum was either fed immediately or stored under refrigeration (4°C) for a maximum of 48 h for use at a later date (Cummins et al., 2017). Colostrum that had been frozen or refrigerated was defrosted and warmed using tepid water and fed immediately at a temperature of approximately 37°C, as determined by visual thermometer. Colostrum quality was initially measured using a digital Brix refractometer (Milwaukee, MA871 Refractometer; Milwaukee Electronics Kft) and a sample (100 mL) collected for laboratory analysis to more accurately quantify IgG concentration (as described in Colostrum and Serum IgG Analyses section). Only colostrum with a Brix value of ≥22% was used (Bielmann et al., 2010). Colostrum that failed to satisfy the Brix value requirement was not used in this study or provided to any calves as a first feed. When pooling colostrum, a sample was taken of each individual colostrum contribution before pooling, as well as a sample once the pool had been assembled. Each colostrum sample was stored at −20°C until analysis for IgG was conducted by radial immunodiffusion (**RID**) assay (Triple J Farms).

In 2016, following a single feed of colostrum, all calves received milk replacer (**MR**; 26% CP, 16% crude fat; Heiferlac Instant) thereafter. In 2018, calves received colostrum and 5 feeds of transition milk from the same origin (i.e., a calf that received own-dam colostrum then received 5 feeds of transition milk from their own dam; in the case of PC, transition milk from the same cows was pooled and fed). Following the fifth feed of transition milk, calves received MR (26% CP) thereafter. At 4 d of age, calves were transferred from their individual pen to a group pen (indoors; 7.5 × 4.7 m area with deep straw bedding; 15 cm) containing up to 15 calves. Calves from each treatment group were evenly distributed across group pens. Once a group was

created, calves remained in their allocated group until after weaning. The maximum difference in birthdate within groups was 2 wk.

Automatic feeding stations (Vario Smart Powder, TAP5–VS1–50: Förster–Technik GmbH) were located within each group pen and provided MR according to a predesigned feeding program. Calves entering the group pen commenced an individual feeding program, initially receiving 4 L of MR per day, which increased gradually to 6 L of MR per day over a 7-d period. This volume of MR per day remained constant for the duration of time taken to reach the minimum target weaning weight (95 kg for HF, 80 kg for HF × Jersey). The feeding program was then modified to achieve gradual weaning over a period of 14 d. At all stages of the feeding program, the daily MR allowance was delivered over 3 equal feeds, prepared fresh at 37°C in 1-L portions at a concentration of 15%. During the housing period, fresh clean drinking water was available in automatic drinking bowls. Hay and concentrates (18% CP, 10.4% crude fiber, 6.2% crude ash: Roches Feeds) were offered ad libitum. Postweaning, calves were given full-time access to pasture and offered 1 kg of supplementary concentrate feed per day, identical to that described above.

Blood Sample Collection

Before we sampled blood from precalving cows, an area at the tail base was sanitized using an alcohol swab. From this area, 10-mL blood samples were collected via the coccygeal vein using 21 gauge (0.75 × 25 mm) vacutainer needles (BD Vacutainer PrecisionGlide Multiple Sample Needle, Becton, Dickson and Company) and plain serum tubes (10-mL BD Vacutainer, BD).

Within 1 h of birth, and before colostrum feeding, one 10-mL blood sample was taken from each calf via the jugular vein using 21 gauge (0.75 × 25 mm) vacutainer needles (BD Vacutainer PrecisionGlide Multiple Sample Needle, Becton, Dickson and Company) and a plain serum tube (10-mL BD Vacutainer, BD). A second blood sample was taken from each calf 24 h postcolostrum feeding to determine serum IgG.

Calves were blood-sampled at monthly intervals thereafter, and the resultant serum samples were analyzed for antibodies against the same range of pathogens as each dam (i.e., BHV-1, BVDV, *Leptospira* Hardjo, *Salmonella* species, bovine rotavirus, bovine coronavirus, PI-3, and BRSV; Enfer Scientific, Ireland). These were collected to examine the depletion rate of pathogen-specific antibodies in calves of known colostrum status. Blood samples were refrigerated at 4°C for 24 h before serum separation by centrifugation (3,000 × g for 30

min) at 4°C. Following centrifugation, serum samples were collected and frozen at −20°C until submitted for IgG or serological analysis.

Colostrum and Serum IgG Analyses

Calf serum samples (0 and 24 h) and colostrum samples were defrosted at 4°C and analyzed using RID kits (Triple J Farms). Each kit contained a 24-well test plate and 3 reference sera samples. Kits were stored at 4°C and allowed to reach room temperature (20–24°C) 30 min before use. Colostrum samples were prepared at a 1:3 dilution using distilled water, whereas 24-h serum samples were prepared at a 1:2 dilution using the same diluent. The 0-h serum samples did not require any dilution before analysis because IgG concentrations at this time point would be negligible (Stott et al., 1979). On each test plate, wells 1 to 3 contained kit reference sera (196 mg/dL, 1,402 mg/dL, 1976 mg/dL, respectively), well 4 was blank, and the remaining wells contained serum or colostrum samples. Each sample was tested in duplicate. Plates were incubated at room temperature for 24 h. Diameters of precipitate rings surrounding each well were measured to the nearest 0.01 mm using digital vernier calipers. Ring diameter values of the 3 reference sera samples were squared and plotted against their known concentrations, producing a standard reference curve. This was then used to determine the IgG concentration of the serum and colostrum samples present on the plate. Samples that produced a value beyond the range of the reference curve were re-analyzed at an increased dilution rate.

Apparent Efficiency of Absorption

The ratio of IgG absorbed into the neonatal circulatory system relative to the IgG mass ingested (i.e., the apparent efficiency of absorption; **AEA**) was calculated using the following set of formulas (Quigley et al., 1998):

$$AEA = \frac{\text{serum IgG}\,(g/L) \times \text{plasma volume}\,(L)}{\text{total IgG intake}\,(g)} \times 100$$

and

$$\text{plasma volume (L)} = 0.089 \times \text{birth BW (kg)},$$

where total IgG intake = [IgG] of colostrum received × (birth BW × 0.085), assuming a feeding rate of 8.5% of birth BW (Conneely et al., 2014). When calculating AEA, samples (colostrum and 24-h calf serum) for

4 calves produced a value of >100%. As this would not be biologically possible, these values were excluded from the data set.

Health Scoring and Performance Recording

From birth to 4 wk postweaning, individual animal health scores were assigned to each calf on a twice-weekly basis. This was carried out by a single trained observer using a modified calf health–scoring system that combined health-scoring systems developed by the School of Veterinary Medicine, University of Wisconsin-Madison and Teagasc (Dillane et al., 2020; Sayers et al., 2016). Calves were scored on 11 different aspects of health as follows: demeanor, mobility, interest in surroundings, temperature, ears, eyes, nasal discharge, fecal consistency, cough, naval, and joints. Each individual aspect received a score from 0 to 3; 0 represented normal and 3 represented the most severely affected. Following each health-scoring assessment, results were relayed to the farm manager to allow treatments to be provided if necessary. Any illness episodes were treated appropriately by farm personnel or a veterinarian, depending on the severity or complexity of the illness. Type of treatment and duration of administration were recorded. Calves were weighed weekly preweaning and at 2-wk intervals postweaning.

Statistical Analyses

All statistical analyses were conducted using SAS (SAS version 9.4; SAS Institute Inc.). Normality of dependent variables was assessed using PROC UNIVARIATE through an assessment of residual skewness combined with a visual examination of the distribution pattern. All dependent variables were normally distributed. Significant associations were confirmed when $P < 0.05$; in both models, interactions were examined between significant variables and least squares means assessed. Factors associated with colostrum and calf serum IgG concentration were assessed using linear mixed models in PROC MIXED, as were calf disease-specific serological responses at 1-mo of age and treatment group. Factors considered in the model investigating colostrum IgG included dam breed (HF, HF cross), year (2016, 2018), parity (1, 2, 3, ≥4), and dam pathogen-specific serological status. For calf 24-h serum IgG concentration, factors considered in the model included colostrum feeding treatment (MC, NMC, PC), dam parity, calf breed, birth BW, IgG concentration of colostrum fed, year, and dam pathogen-specific serological status. Birth BW was included in the model as a continuous variable, but also in a separate model

as a categorical variable, based on quartiles of the data set. Birth month nested within year was also included in the model, and interactions between treatment and year were investigated. Factors considered in the model investigating disease-specific antibody levels in 1-mo-old calves included colostrum feeding treatment, breed, IgG concentration of colostrum, year, birthweight, and dam serological status and antibody levels to the same disease. A separate model was used for each of individual pathogen status (BVDV, *Salmonella* species, BHV-1, *Leptospira* Hardjo, PI-3, BRSV, bovine rotavirus, and bovine coronavirus).

Survival Analyses

Survival of maternally derived pathogen-specific antibodies in heifer calves was documented through analyses of serum samples collected on a monthly basis. Survival rate in the present study refers to the time point from birth to the month in which half of all samples produce a seronegative result to the specific pathogen. The effect of colostrum feeding treatment (MC, NMC, PC), IgG concentration of colostrum received, birth BW, and serum IgG concentration on survival of antibodies against a specific pathogen was analyzed by the Cox proportional hazards model using PROC PHREG. Kaplan-Meier survival functions were estimated for treatment and year using PROC LIFETEST. A separate model was applied for each specific disease when investigating factors affecting antibody depletion.

RESULTS

Colostrum IgG

No interaction existed between treatment and year with regard to colostrum IgG concentration. Mean colostrum IgG concentration was 84.2 mg/mL and ranged from 52.5 to 164.9 mg/mL. No difference was detected between the overall IgG concentration of PC and the mean IgG concentration of individual samples used to create the pool. Treatment, year, calving month, and parity were all associated ($P < 0.05$) with colostrum IgG concentration. Mean colostrum IgG concentration was highest for the NMC treatment group, whereas MC and PC yielded similar values (Table 1). For all treatment groups, the mean IgG concentration was >80 mg/mL, exceeding the quality threshold of 50 mg/mL. Mean colostrum IgG concentration in 2016 was 71.2 mg/mL (range = 34.9–108.9 mg/mL), which was significantly lower ($P < 0.001$) than 2018 (mean = 97.0 mg/mL; range = 54.8–164.92 mg/mL).
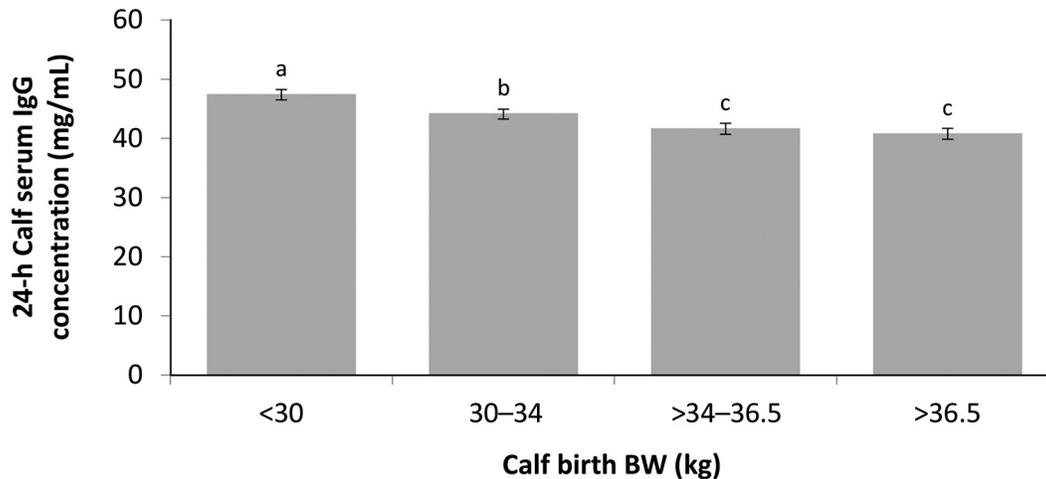
Figure 1. Least squares means (1 ± SE) for 24-h calf serum IgG concentration based on birth BW (when included as categorical variable). Categories were based on quartiles of the data set and include <30 kg (n = 30), 30 to 34 kg (n = 36), >34 to 36.5 kg (n = 24), and >36.5 kg (n = 28). Bars with different letters (a–c) indicate a significant difference ($P < 0.05$) in means.

Calf Serum IgG

Mean calf serum IgG concentration was 43.7 mg/mL (range = 18.7–105.4 mg/mL). Calves that received PC had a lower mean 24-h serum IgG concentration than calves fed colostrum from their own dam or another single dam (Table 1). For calf 24-h serum IgG concentration, factors considered in the model included colostrum feeding treatment (MC, NMC, PC), dam parity, calf breed, birth BW, IgG concentration of colostrum fed, year, and dam pathogen-specific serological status. Mean serum IgG concentration in 2016 was 32.6 mg/mL, ranging from 18.7 to 59.8 mg/mL. In 2018, mean serum IgG concentration was 58.4 mg/mL, ranging from 31.3 to 105.3 mg/mL. For BW, a negative association was identified with regard to calf 24-h serum IgG (Figure 1); calves with a birth BW of <30 kg had the highest 24-h serum IgG concentration (47.5 mg/mL). Calves that received highest quality colostrum (i.e., containing >100 mg/mL of IgG) had the highest serum IgG concentration at 24 h. When calf serum IgG concentration at 24 h was plotted against IgG concen-tration of colostrum received (Figure 2), this produced a regression equation of $y = 0.5454x - 0.3835$, with a coefficient of determination of 0.38.

Associations With Calf Serological Status at 1 mo

Calf BVDV serological status at 1 mo of age was associated ($P < 0.05$) with colostrum feeding treatment, which was highest among MC calves. Calf PI-3 serological status was associated ($P < 0.05$) with treatment, which was lowest among the PC treatment group. Calf BRSV serological status at 1 mo of age was associated ($P < 0.05$) with dam BRSV antibody levels. No further significant associations were identified with regard to these pathogens.

With regard to *Leptospira* Hardjo, *Salmonella* Dublin/Typhimurium, BHV-1, bovine rotavirus, and bovine coronavirus serological status of calves at one month of age, no association was identified with colostrum feeding treatment, calf breed, IgG concentration of colostrum received, year, birthweight, and dam serological status to the same pathogen.

Table 1. Mean colostrum IgG concentration and calf serum IgG concentration at 24 h as determined by radial immunodiffusion assay for calves fed own dam (MC), nonmaternal dam (NMC), and pooled colostrum (PC)

Item	Treatment			SEM	*P*-value
	MC	NMC	PC		
Colostrum IgG (mg/mL)	88.55[a]	94.18[b]	87.41[a]	1.66	<0.01
24-h serum IgG (mg/mL)	44.53[a]	43.84[a]	40.50[b]	0.64	<0.01
AEA[1] (%)	55.39[a]	54.07[a]	51.52[b]	0.76	<0.01

[a,b]Different superscript letters indicate significant difference between treatments ($P < 0.05$).

[1]AEA = apparent efficiency of absorption.

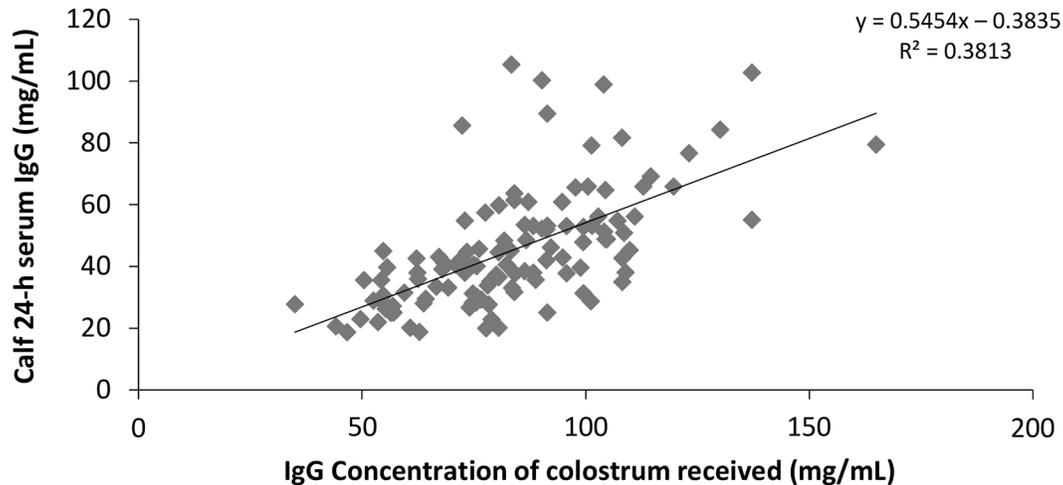$$y = 0.5454x - 0.3835$$
$$R^2 = 0.3813$$

Figure 2. Immunoglobulin G concentration of colostrum received by 119 dairy calves, plotted against the 24-h serum IgG concentration of the same calves, as determined by radial immunodiffusion assay.

Maternally Derived Antibody Survival

Antibody survival rates for the diseases of interest, in both 2016 and 2018, are summarized in Table 2. For BVDV, *Leptospira* Hardjo, *Salmonella* Dublin/Typhimurium, PI-3, BRSV, bovine rotavirus, and bovine coronavirus, no associations were detected between survival of antibodies and colostrum feeding treatment, IgG concentration of colostrum received, year, or calf serum IgG concentration at 24 h. With regard to BHV-1, colostrum feeding treatment ($P < 0.05$) and IgG concentration of colostrum ($P < 0.05$) were associated with antibody survival. Antibody survival for infectious bovine rhinotracheitis (**IBR**) was lower for calves fed PC compared with calves fed other-dam colostrum. No difference was identified in IBR antibody survival for calves fed PC and MC colostrum, or calves fed MC and NMC colostrum. A linear association was detected between anti-BHV-1 antibody survival and IgG concentration of colostrum. Higher IgG colostrum resulted in an increased duration of survival of anti-BHV-1 antibodies.

DISCUSSION

The role of colostrum management in providing adequate immunological protection to neonatal calves has been widely investigated, and thresholds for colostrum quality, as well as optimum volume and timing for colostrum feeding have been established (Godden, 2008; Conneely et al., 2013). However, limited information is available on the effect of colostrum source (individual dam or pooled) on passive immunity, as well as on maternally derived antibody survival in the

calf. Research shows that on commercial Irish dairy farms, colostrum is fed from several different sources (MC, NMC, or PC; Barry et al., 2019), often to reduce the farm labor requirements that increase dramatically during the calving season. This study aimed to assess the effect of feeding individual dam and PC on passive immunity in neonatal calves, and to investigate associations with pathogen-specific antibody survival over an 8 mo period.

Colostrum Quality

Mean colostrum IgG concentration obtained in this study was high, well exceeding the 50 mg/mL threshold for good quality (Godden, 2008). Although colostrum quality in the present study was not as high as previously identified among Irish dairy research herds (Conneely et al., 2013), it was similar to that found during a recent study on commercial Irish dairy farms (85 mg/mL; Barry et al., 2019). This indicates that findings from the present study could be reflective of expected outcomes for passive immunity that would be experienced on commercial dairy farms when the same management practices are applied. Large variation existed in colostrum quality both between and within years. Based on existing literature, such variation would be expected because colostrum quality is influenced by a large number of factors including breed, parity, prepartum nutrition, dry period length, and time of colostrum collection (Godden, 2008). Although parity and breed of the cohort of dams from which colostrum was used was similar in both years, and strict guidelines applied for timing of colostrum collection, prepartum nutrition and dry period length were not controlled, and could

Table 2. Maternally derived disease-specific antibody survival in dairy heifers during the first 8 mo of life, based on colostrum feeding treatment and provision of colostrum only (2016) or colostrum and transition milk (2018)

Item[1]	Antibody survival analysis[2] (mo)															
	BVD		Salmonella infection		IBR		Leptospirosis		PI-3		BRSV		Rotavirus		Coronavirus	
	2016	2018	2016	2018	2016	2018	2016	2018	2016	2018	2016	2018	2016	2018	2016	2018
MC	4.5	5.4	4.6	5.4	4.3	4.9	4.1	5.1	5.7	7.4	6.3	6.9	4.3	6.9	—[3]	5.9
NMC	4.9	5.5	4.6	5.6	4.4	4.8	4.1	5.3	5.8	7.4	6.3	6.9	4.1	6.9	—	3.0
PC	4.5	5.5	4.9	5.5	4.4	4.6	4.1	4.9	5.9	7.1	6.3	6.7	4.0	6.4	—	3.0

[1]MC = maternal colostrum; NMC = nonmaternal dam colostrum; PC = pooled colostrum.

[2]BVD = bovine viral diarrhea; IBR = infectious bovine rhinotracheitis; PI-3 = bovine parainfluenza 3 virus; BRSV = bovine respiratory syncytial virus.

[3]Right-censored data indicate >50% of samples were positive at sampling end-point.

potentially be a source of variation. Other factors such as dam health (Dardillat et al., 1978) and mineral status (Weiss et al., 1990) can also influence colostrum quality, which could also be potential sources of variation both between and within years. Previous studies have found similar variation in colostrum quality; however, few studies have examined colostrum quality within a herd across several different years. The present study demonstrated that colostrum quality within a herd can vary between years, which further emphasizes the need for year on year assessment of colostrum quality to avoid any negative implications of such variation, such as failed TPI.

Pooling Colostrum

Failure to find a statistical difference between IgG concentration of PC and the mean IgG concentration of MC indicated that the process of pooling does not have a negative effect on IgG concentration. However, in the present study, only colostrum that produced a value of ≥22% on a Brix refractometer was used and combined in equal proportions to create the pool. In situations where colostrum quality is not assessed before pooling and mixed at different proportions, it could negatively affect overall IgG concentration (Weaver et al., 2000) and increase the risk of failed TPI occurring. A recent study by Barry et al. (2019) described how over 30% of Irish dairy farmers reported feeding PC. The same study also found that less than 15% of Irish dairy farmers actively measure colostrum quality. Results from the present study demonstrate that when strict procedures are followed (i.e., using only good quality colostrum, combining in equal proportions, storing correctly), PC can contain an IgG concentration capable of providing adequate TPI. However, this study was conducted within a research herd where high herd health standards were achieved through comprehensive vaccination programs, regular disease screening, prompt isolation or culling of any animals suspected of harboring infectious disease, and meticulous hygiene standards. The associated risk of disease transfer within a herd when pooling colostrum is extremely difficult to overcome (Godden et al., 2009), and pooling colostrum should not be practiced in situations where diseases transmissible through colostrum, such as Johnes disease, are present (Nielsen et al., 2008).

Serum IgG Concentration and AEA

The difference in calf 24-h serum IgG concentration between treatments indicated that colostrum from a single-dam (MC, NMC) results in significantly improved TPI compared with feeding PC. However, colostrum

quality for the pooled treatment group was similar to that of the MC treatment group. This suggested that although the pooling process did not affect IgG concentration of colostrum, it could affect antibody uptake when fed to calves. This was further supported as AEA was lower among calves that received PC compared with single-dam colostrum (own and other dam). The absorption of IgG, as well as other immunoglobulins, occurs in the small intestine of the calf by a passive process, pinocytosis, whereby immunoglobulins are transferred across the intestinal epithelium (Quigley et al., 2002). Differences in AEA in the present study indicated that colostrum source can influence the efficiency with which this process occurs.

We postulated that a phenomenon of immune exclusion may be occurring when using PC; this is when immunoglobulins, in conjunction with the mucus lining of the gut, bind to pathogens to prevent their entry into the body (Everett et al., 2004; Ulfman et al., 2018). During immune exclusion, the pathogens as well as the immunoglobulins remain confined to the intestinal lumen, and the immunoglobulins prevent adhesion to intestinal epithelium (Ulfman et al., 2018). Providing PC from several sources may increase the array of pathogens that are removed, which would decrease serum IgG to some degree, similar to that reported in this study. This may explain the lack of difference in colostrum IgG content between the 3 treatments investigated (MC, NMC, PC) and also explain the reduced immunoglobulin absorption and lower serum IgG of the PC calves. More detailed exploration, such as testing fecal samples for IgG (Ulfman et al., 2018), may be warranted to confirm this. In general, pooling colostrum is discouraged (Godden, 2008); however, evidence from human medicine shows that bovine IgG binds to many human pathogens and allergens, neutralizes experimental infection of human cells, and limits gastrointestinal inflammation (Ulfman et al., 2018). This may suggest that once high-quality colostrum is used (i.e., >50 mg/mL IgG or >22% Brix; Godden, 2008; Bielmann et al., 2010), pooling colostrum may be a good practice, as long as it can be ensured that enough antibodies are absorbed into the blood stream to deal with pathogens calves may encounter, as different dams may have antibodies against different strains of viruses and bacteria yielding cross protection.

The association identified in the present study between calf serum IgG concentration at 24 h and dam parity and birth BW suggests that calves <30 kg at birth have better antibody absorption capabilities. In contrast, Hopkins and Quigley III (1997) reported no difference in calf serum IgG at 24 h based on birth BW; however, BW was used as a covariate of intake in the study. Lower calf BW at birth is associated with lower rates of dystocia, and Berry et al. (2007) reported the probability of calving dystocia for a third parity dam was 1, 2, 5, and 15% for a calf weighing 20, 30, 40, and 50 kg, respectively, born to cows with a calving BW of approximately 440 kg. This association occurs because corticosteroid levels in the neonatal calf are directly related to antibody absorption from colostrum. Following a difficult birth, cortisol release in the calf is inhibited, resulting in lower levels of antibody absorption (Chase et al., 2008). Dystocia-induced acidosis (respiratory or metabolic acidosis) could also be a factor, as studies have demonstrated a negative association with IgG absorption (Boyd, 1989; Besser et al., 1990).

Greater vigor among calves with a lower birth BW could also be a factor, as could the relative absorptive surface area available in a small calf being greater than a large calf. To the authors knowledge, this has not been previously investigated. Murray et al. (2015) reported lower AEA among calves that did not achieve sternal recumbency within 15 min of birth. This supports the hypothesis that calf vigor is a determining factor for AEA, and further investigation is warranted on AEA and interactions between birthweight and calf vigor.

Findings from the present study indicated that particular attention must be given to large calves, and also to those that experience difficult births, to ensure they achieve adequate TPI, while additional efforts are made to minimize calf exposure to infectious diseases. Based on findings from the present study, feeding PC to calves that experience a difficult calving could increase the risk of failed TPI occurring if low-quality colostrum is fed.

Calf Immunity at 1 mo

Failure to find associations between calf immunity at 1 mo of age for leptospirosis, *Salmonella* infection, IBR, BRSV, rotavirus, and coronavirus and calf breed, treatment, IgG concentration, and year indicated that similar levels of passive immunity are achieved by calves to these diseases. Yang et al. (2015) reported a faster development of the immune defense mechanism when a higher quality of colostrum was fed; given there were minimal differences in colostrum quality offered to calves in the present study, it was unsurprising there was a lack of difference in calf immunity at 1 mo of age. Furthermore, levels of circulating IgA and IgG do not reach significant levels in calves until 16 to 32 d after birth, with adult levels only being achieved approximately 4 mo after birth (Chase et al., 2008).

Calf antibody levels to BVD were highest among the MC treatment group, which also had the highest serum IgG concentration at 24 h. This may suggest that in herds where BVD is an issue, best practice would be to

feed calves their own dams' colostrum. Calf immunity levels to PI-3 were lower among calves that received PC, and although all calves were ELISA-positive for PI-3 at 1 mo of age, NMC provided increased immunity to the disease relative to PC. Although no biological difference existed for PI-3 antibody levels at 1 mo of age, feeding single dam (other or own dam) colostrum could result in greater immunity to PI-3 relative to PC. Despite the differences across treatments in terms of antibodies to respiratory viruses, which require more investigation, the study did show that respiratory diseases were associated with colostrum treatments. Failure to identify an association between colostrum treatment and gut pathogens may be related to the theory of immune exclusion discussed earlier; however, further investigation is required.

Establishing links between colostrum and disease-specific immunity among calves is a relatively new field of research, and this study adds to the limited knowledge even though there are limitations. In the present study, as levels of disease-specific antibodies within colostrum were not assessed, this could not be accounted for, and differences in 1-mo immunity could occur due to differences in specific antibodies within the colostrum rather than as a result of the source of colostrum fed. Determining disease-specific antibody levels in colostrum would prove difficult due to the physical characteristics of the colostrum (yellow color, high viscosity), which could interfere with ELISA performance. Also, maternally derived antibodies and those produced as a result of an innate immune response could not be differentiated in the survival analysis. To determine true survival of maternally derived antibodies, labeling of disease-specific maternally derived antibodies would be required.

Maternally Derived Antibody Survival

Antibody survival rates in the present study indicated that calves had comprehensive immunity in the early stages. Fulton et al. (2004) reported that most maternally derived antibodies have a decay half-life of 16 to 28 d, although the duration of this immunity is dependent on the mass of antibodies ingested and absorbed by the calf in the first 24 h of life. Given the high 24-h serum IgG concentrations identified across each of the treatment groups (>40 mg/mL), this suggests a high level of antibody absorption that would result in a lower rate of antibody depletion (Chase et al., 2008). A study by Muñoz-Zanzi et al. (2002) assessed the survival rate of maternal antibodies to BVD and identified a survival rate of 141 d (4.7 mo), which is similar to that of the present study (4.6 and 5.5 mo in 2016 and 2018, respectively). A study on beef calves by Fulton

et al. (2004) reported estimated survival times of 6.3 and 6.2 mo for antibodies to PI-3 and BRSV, which are similar to survival rates identified in the present study.

High survival among antibodies to some diseases in 2018 relative to 2016 could have occurred due to the higher concentration of IgG in colostrum during 2018. It could also be because calves were fed 5 feeds of transition milk in 2018, whereas they were fed no transition milk in 2016. Failure to identify an effect of treatment for survival of antibodies to all diseases, except IBR, indicated that in high health status herds, which screen colostrum before feeding, colostrum source will not influence disease-specific immunity in early life. Although colostrum is essential to protect the calf in early life before it develops its own immunity, there may be interference from the maternally derived antibodies that inhibit the calf's ability to mount an immune response to antigens from vaccination or natural infection (Morein et al., 2002).

For IBR, feeding MC increased antibody survival. Calves within the MC treatment group also had the highest serum IgG concentration. Given the positive association between passive immunity levels and antibody survival, this was not unexpected. Dunn et al. (2018) reported the importance of feeding colostrum from vaccinated dams to provide calves with passive protection against IBR. The cows used in this study were vaccinated for IBR, but given that a low proportion of farmers in Ireland vaccinate to provide protection against IBR (Barry et al., 2020), a review of calf vaccination protocols for IBR could be required. To the authors' knowledge, survival of maternal antibodies to *Salmonella* infection and leptospirosis has not been previously documented in female dairy calves. Therefore, the present study provided valuable information on calf immunity to *Salmonella* infection and leptospirosis, which will also be important for future studies that aim to improve calf immunity to both diseases (*Salmonella* infection and leptospirosis). As part of a herd health protocol, all dams used in the present study received a vaccine for rotavirus and coronavirus to enhance passive immunity of calves to these pathogens. The present study indicated that this protocol is successful because calves had immunity to both rotavirus and coronavirus during the period of susceptibility (first 3 mo of life; Mayameei et al., 2010). Given the increased antibody survival rate for coronavirus in 2018, it may also suggest that feeding transition milk is a contributory factor to improved immunity.

CONCLUSIONS

Our results showed that feeding colostrum from a single dam (own or other dam) resulted in higher

passive immunity among calves, relative to those that received PC. However, it may suggest that immune exclusion occurs with PC. Providing PC may be a good practice as long as it can be ensured that enough antibodies are absorbed into the blood stream to deal with pathogens calves may encounter because different dams may have antibodies against different strains of viruses and bacteria, yielding cross protection. However, this needs to be confirmed. Maternal antibody survival has been documented in the present study and indicated that once calves receive a sufficient amount of high-quality colostrum, they achieve comprehensive immunity to a range of diseases. Although there were differences across treatments in terms of antibodies to respiratory viruses, which require more investigation, the study did show that respiratory diseases were associated with colostrum treatments. Although no association was identified between colostrum treatments and gut pathogens, further investigation is warranted in which the level of disease-specific antibodies in colostrum is quantified.

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