this study was to investigate the effect of time passed from parturition to milking, and from these data propose a scheme that can be used at smaller farms to increase the likelihood of harvesting colostrum with a high content of antibodies.

Materials and Methods

Twenty-one farms with 100 to 1250 dairy cows/farm participated in the study in September through November 2013, delivering between 1 and 23 colostrum samples from each farm. The included dairy farms were positioned <2 h drive from the University Laboratory (AU-Foulum, Denmark). Colostrum was harvested as the farmer would normally do it, and at the latest 24 h after calving. For each sample, the time for calving and milking was noted by the farmer; 92% of the samples were collected at latest 13 h after calving. Colostral IgG concentration was determined with ELISA (Bovine IgG ELISA quantitation Set, Cat. No. E10-118; Bethyl Laboratories Inc, Montgomery, TX). The results were expressed as IgG concentration in g/L.

Results

The results showed that the IgG content decreased as time from calving to milking (R^2=0.15) increased. Only 51% of the colostrum collected later than 5 hours after calving contained more than 50 g IgG/L, whereas 82% of the samples harvested during the 5 hours following calving contained the recommended level of IgG (Lokke et al, 2016). In practice, however, it is difficult to milk all fresh cows within 5 hours of calving. To get around this, fresh cows can be milked right after startup of the milking system and cows that give birth during the milking period can be milked at the end. In the period after the milking system is turned off until 5 hours before next milking, a mobile milking unit can be used.

Significance

The results emphasize the importance of milking as soon as possible after calving. By changing routines at the farm it is possible to milk most fresh cows within 5 hours after calving, thereby increasing the likelihood of collecting high-quality colostrum from 50% to around 80%.

Reference

Lokke MM, Engelbrecht R, Wiking L. Covariance structures of fat and protein influence the estimation of IgG in bovine colostrum. *J Dairy Res* 2016; 83:58-66.

Development and validation of the VetMAX-Gold MAP Detection Kit

A. Burrell, *BS, MS*; **I. Leyva Baca**, *DVM*; **R. Shah**, *BS*; **D. Kephart**, *PhD*
Thermo Fisher Scientific, Austin, TX 78744

Introduction

Mycobacterium avium subspecies *paratuberculosis* (MAP) is the causative agent for Johne's disease (chronic granulomatous enteritis of the small intestine) in cattle. Johne's disease causes severe economic losses in the cattle industry due to reduced productivity, reproductive losses, and the eventual death or culling of the infected animal. We have validated a MAP testing workflow consisting of high-throughput nucleic-acid purification and MAP detection from both individual and pooled bovine fecal samples. The VetMAX-Gold MAP Detection Kit is a real-time PCR assay for the rapid in vitro detection of MAP DNA purified from bovine feces. The assay targets a unique sequence element in the MAP genome to provide highly sensitive and specific results. The purpose of this study is to determine the performance characteristics of the VetMAX-Gold MAP Detection Kit in detecting MAP DNA from nucleic acid extracted from individual and pooled bovine fecal samples.

Materials and Methods

The VetMAX-Gold MAP Detection Kit workflow was evaluated with 126 individual MAP-positive and 134 individual MAP-negative bovine fecal samples. The MAP status of each sample was confirmed with culture (MGIT culture system, Herrold's Egg Yolk (HEY), or TREK ESPTM Culture System II) prior to the start of the study. MAP samples were sourced from diverse geographic regions (9 states) and represented a range of MAP infectivity (19 to 21% heavy shedder, 10 to 13% moderate shedder, 10 to 27% light shedder). The feasibility of pooling up to 5 bovine fecal samples into a single nucleic-acid extraction and detection test was evaluated by testing 51 MAP-positive pools and 24 MAP-negative pools. All pools consisted of 5 individual fecal samples. Forty-nine positive pools were created by combining 1 MAP-positive sample with 4 MAP-negative samples. Two positive pools were created by combining 2 MAP-positive samples with 3 MAP-negative samples. For both individual and pooled

sample testing, a collaborator laboratory purified nucleic acid from bovine fecal samples using the MagMAX Total Nucleic Acid Isolation Kit. MAP bacterium was physically and chemically lysed by homogenizing the fecal supernatant using the FastPrep®-24 homogenizer in the presence of lysis solution. Five thousand copies/reaction of XenoDNA Control was spiked into the lysis solution of each purification to monitor for inhibition. Samples were then processed using the MagMAX Express-96 Deep Well Magnetic Particle Processor. 8 µL of extracted nucleic acid was tested using the VetMAX-Gold MAP Detection Kit on the 7500 Fast Real-Time PCR system according to the Instructions for Use.

Results

The results of testing show the VetMAX-Gold MAP Detection Kit produced diagnostic sensitivity and specificity values of 96.2% and 96.4%, respectively, when testing individual fecal samples as compared to culture. For individual samples, the predictive value of a positive test was 93.8% and predictive value of a negative test was 99.3%. Pooled sample testing with the VetMAX-Gold MAP Detection Kit resulted in diagnostic sensitivity and specificity values of 96.2% and 100%, respectively, as compared to culture. For pooled samples, the predictive value of a positive test was 100% and predictive value of a negative test was 96.0%.

Significance

This study indicates that DNA isolated from a diagnostic bovine fecal sample, tested with the VetMAX™-Gold MAP Detection Kit, provides an economical and rapid solution for MAP detection from both individual and pooled fecal samples. The results of this study are under review by APHIS' Center for Veterinary Biologics to support Thermo Fisher Scientific's Biological Product License application.

Effect of Acetate Ringer's solution with or without dextrose intravenously administered to diarrheic calves

K. Tsukano, *DVM*[1]; **S. Kato**, *DVM*[2]; **K. Suzuki**, *DVM, PhD*[1]; **Y. Nishi**, *DVM*[1]; **S. Sarashina**, *DVM, MS*[2]; **T. Shimamori**, *DVM*[1]
[1]*The School of Veterinary Medicine, Rakuno Gakuen University, 582 Midorimachi, Bunkyoudai,Ebetsu, Hokkaido, 069-8501, Japan*
[2]*Donan-NOSAI, Donan Agricultural Mutual Relief Association, 25-16 Misugityo, Yakumo, Futami-gun, Hokkaido, 049-3114, Japan*
[3] *The Department of Veterinary Clinical Sciences, Ohio State University, Columbus, OH 43210*

Introduction

Neonatal diarrhea remains the most common cause of death in beef and dairy calves. Intravenous (IV) fluid therapy is an important method for decreasing mortality associated with diarrhea in calves. Although Acetated Ringer's (AR) solution is superior for correcting moderate metabolic acidosis and circulation volume in calves with dehydration, it cannot improve the negative energy balance. Fluids using AR with 5% dextrose (ARD) may be useful for the treatment of calf diarrhea in order to prevent catabolism. The objectives of this study were to evaluate the effects of ARD on rehydration and metabolism restoration in calves with diarrhea.

Materials and Methods

A total of 16 diarrheic calves with a mean age of 9.6 ± 3.1 days (from 5 to 14 days old) were used in this study. *Cryptosporidium parvum* was isolated from 81.3% (13/16) of the diarrheic calves. A 14-gauge catheter was inserted into the right jugular vein for fluid infusion. The calves were randomly assigned to the AR (n=8; IV infusion of AR) or ARD groups (n=8; IV infusion of ARD). Calves received 100 ml/kg of 1 of the fluids, at a flow rate of 25 ml/kg/hr. The initiation of the infusion of the fluid was designated as time 0. Venous blood samples were collected at 0 (pre-infusion), 0.5, 1, 2, 4, and 24 hours after initiation of fluid infusion. Venous blood samples were anaerobically collected in a heparinized 1 ml syringe from the left jugular vein, and the tips of the syringes were capped after collection. The blood samples were analyzed for β-hydroxybutyrate (BHBA) by an automatic analyzer (Precision Xceed, Abbott Japan), and for blood gases, Ht, BE, Hb, sodium concentration, potassium chloride, and glucose by an automatic analyzer (i-STAT 1, USA). Changes in relative plasma volume (rPV) were calculated from Hb and Ht using accepted formulas. The data are expressed as the means ± standard deviation. Within groups, mean values for each dependent variable were compared with the pre-values, us-