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Rapid assessment of bovine colostrum quality: How reliable are transmission infrared spectroscopy, and digital and optical refractometers?

I. Elsohaby,*†[1] J. T. McClure,* M. Cameron,* L. C. Heider,* and G. P. Keefe*
*Department of Health Management, Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, Prince Edward Island, Canada C1A 4P3
†Infectious Diseases, Department of Animal Medicine, Faculty of Veterinary Medicine, Zagazig University, Zagazig City 44511, Sharkia Province, Egypt

ABSTRACT

The objectives of this study were to evaluate the performance of the transmission infrared (IR) spectroscopic method and digital and optical Brix refractometers for measurement of colostral IgG concentration and assessment of colostrum quality of dairy cows. Colostrum samples (n = 258) were collected from Holstein cows on 30 commercial dairy farms in Nova Scotia and Newfoundland, Canada. Colostral IgG concentrations of 255 samples were measured by the reference radial immunodiffusion (RID) assay and IR spectroscopy. The Brix scores were determined on 240 of these samples using both the digital and optical Brix refractometers. Approximately half (48%) of the colostrum samples had RID IgG concentrations <50 g/L, which was the cut-point for poor quality. The correlation between RID and IR IgG concentrations was 0.88. The correlations between RID IgG concentration and Brix scores, as determined by the digital and optical refractometers, were 0.72 and 0.71, respectively. The optimal cutoff levels for distinguishing good- and poor-quality colostrum using IR spectroscopy, and digital and optical Brix refractometers were at 35 g/L and 23% Brix, respectively. The IR spectroscopy showed higher sensitivity (90%) and specificity (86%) than the digital (74 and 80%, respectively) and optical (73 and 80%, respectively) Brix refractometers for assessment of colostrum quality, as compared with RID. In conclusion, the transmission-IR spectroscopy is a rapid and accurate method for assessing colostrum quality, but is a laboratory-based method, whereas Brix refractometers were less accurate but could be used on-farm.

Key words: infrared spectroscopy, Brix refractometer, immunoglobulin G, radial immunodiffusion assay

INTRODUCTION

Colostrum is a critical source of immunoprotection and nutrition for newborn calves (Bielmann et al., 2010). Ingestion of good-quality colostrum during the first 24 h of life is essential for the future health and performance of dairy calves (Rauprich et al., 2000). Insufficient ingestion or absorption of colostral IgG results in failure of transfer of passive immunity (Calloway et al., 2002; Godden, 2008). Calves with failure of transfer of passive immunity (serum IgG <10 g/L) are more susceptible to infectious diseases and have higher morbidity and mortality rates (Robison et al., 1988; Donovan et al., 1998; Virtala et al., 1999). Thus, it is important to assess colostrum quality before feeding to calves. Only 53% of US dairy operations routinely evaluate colostrum quality as part of their replacement heifer management, and 45% of these operations depend on visual appearance to assess colostrum quality (USDA, 2016).

Several methods have been developed to evaluate colostrum quality by measuring the colostral IgG concentration either directly or indirectly, but few of them are applicable to farm or field conditions. The radial immunodiffusion (**RID**) assay is regarded the most accurate reference method for directly measuring bovine colostral IgG content (McBeath et al., 1971; Oyeniyi and Hunter, 1978; Fleenor and Stott, 1980). However, this assay is a laboratory-based method, requires 18 to 24 h to obtain results, has high cost, lacks automation, and uses reagents with a short shelf-life (Riley et al., 2007; Bielmann et al., 2010). Thus, the RID assay is not practical for timely, routine on-farm monitoring of colostrum quality.

The colostrometer and Brix refractometer are the most common indirect methods for evaluating colostrum quality on-farm. Colostrometers measure the specific gravity of colostrum and provide an estimate of relative quality, not precise IgG concentrations (Fleenor and

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[1]Corresponding author: ielsohaby@upei.ca

Stott, 1980; Pritchett et al., 1991). Results from the colostrometer are affected by colostrum temperature and total content of all solids, not just IgG (Mechor et al., 1992; Morin et al., 2001); moreover, it is fragile and needs to be carefully cleaned before use (Bielmann et al., 2010; Bartier et al., 2015). Only 4% of farms in Quebec and 11% of farms in the United States have been reported to use the colostrometer consistently (Vasseur et al., 2010; USDA, 2016). Brix refractometers, either digital or optical, can be used to estimate colostral IgG levels (Bielmann et al., 2010; Bartier et al., 2015; Morrill et al., 2015). Previous studies reported a strong correlation between Brix scores and IgG concentrations, as determined by RID (Chigerwe et al., 2008; Bielmann et al., 2010; Bartier et al., 2015). Unlike the colostrometer, the Brix refractometer is not sensitive to ambient temperature (Bielmann et al., 2010; Bartier et al., 2015) and is less fragile. Thus, Brix refractometers have been promoted by the dairy industry as an effective on-farm tool to estimate colostral IgG and colostrum quality (Thornhill et al., 2015). However, only 4% of US dairy producers routinely use Brix refractometers to evaluate colostrum quality (USDA, 2016).

Recently, infrared (**IR**) spectroscopy, in combination with multivariate data analysis, has emerged as an alternative technique for assessing colostrum quality (Rivero et al., 2012; Elsohaby et al., 2016b). Infrared spectroscopy has several advantages that overcome the drawbacks associated with other methods. It is a rapid, low-cost test, requires minimal sample preparation, and 1 spectrum can be used for quantitative analysis of several components (Shaw et al., 1998; Shaw and Mantsch, 1999, 2000). A small, compact, portable infrared spectroscope has been manufactured that is ideally suited for field use on the farm, veterinary clinic, or small laboratory (Santos et al., 2013). Infrared spectroscopy, in combination with partial least squares (**PLS**) regression, has been widely used for quantifying human serum total protein and glucose (Ward et al., 1989; Shaw et al., 1998), bovine milk protein, lipid, and lactose analysis (Rutten et al., 2011a,b), as well as serum IgG in bovines (Elsohaby et al., 2014, 2016a), equines (Riley et al., 2007), and camelids (Burns et al., 2014). To our knowledge, no study has validated the use of IR spectroscopy for quantifying colostral IgG and assessing colostrum quality in dairy cows. Thus, the objectives of our study were (1) to investigate the utility of previously built PLS models (Elsohaby et al., 2016b) for quantifying colostral IgG concentration and assessing colostrum quality in dairy cows using IR spectroscopy, and (2) to determine and compare the diagnostic test characteristics of IR spectroscopy and digital and optical Brix refractometers for assessing colostrum quality.

MATERIALS AND METHODS

Colostrum Sample Collection

Colostrum samples (n = 258) were collected between April and September 2015 from Holstein dairy cows on 30 commercial dairy farms in Nova Scotia (n = 24) and Newfoundland (n = 6), Canada. Herds were selected by veterinary clinics in the study area and were asked to provide 10 colostrum samples. Colostrum was collected by farm staff between 1 and 15 h after calving, and each farm delivered between 4 and 12 samples. At each farm, 50 mL of colostrum were collected in a vial labeled with the farm name, cow identification number, and date of collection, and then stored on the farm at −20°C until transportation to the Maritime Quality Milk Laboratory, University of Prince Edward Island. All samples arrived frozen and were placed at −20°C for later analysis. This study was conducted in accordance with the Canadian Council on Animal Care guidelines (CCAC, 2009) under a protocol approved by the Animal Care Committee at University of Prince Edward Island.

Colostrum Sample Analysis

RID Assay. A commercial RID assay (Bovine IgG RID Kit; Triple J Farms; Bellingham, WA) was used as the reference method for measuring colostral IgG concentrations. Colostrum samples were thawed at room temperature (20–24°C) and vortexed for 10 s. Thawed samples were diluted (1:4) with deionized sterile water and mixed by vortexing at a maximum of 2,700 rpm for 10 s. After dilution, the RID assay was performed according to manufacturer's instructions, using 5 μL of diluted colostrum in each well, and tested alongside the manufacturer's standard. The RID plates were incubated at room temperature for 18 to 24 h and the precipitating ring diameter surrounding the well was measured using a hand-held caliper. Colostrum samples with IgG concentration greater than the manufacturer's stated performance range for the assay (>30 g/L) were diluted (1:6) with deionized sterile water and retested. Samples that still showed IgG concentrations above the standard range were considered good-quality colostrum for diagnostic tests, but they were excluded from the correlation analysis (n = 3). To account for plate-to-plate variation, the same IgG standard (same lot) was used on all RID assays, and each of the assay standards and colostrum samples was tested in duplicate. The average of the assay standards was used to build a calibration curve to determine the IgG concentration for each colostrum sample. The final IgG concentration for each sample was determined by calculating the average