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Suitability of capillary blood obtained by a minimally invasive lancet technique to detect subclinical ketosis in dairy cows by using 3 different electronic hand-held devices

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ABSTRACT

The objective of this study was to evaluate the suitability of capillary blood obtained by a minimally invasive lancet technique to detect subclinical ketosis in 49 prepartum and 191 postpartum Holstein-Friesian cows using 3 different electronic hand-held devices [FreeStyle Precision (FSP, Abbott), GlucoMen LX Plus (GLX, A. Menarini), NovaVet (NOV, Nova Biomedical)]. The β -hydroxybutyrate (BHBA) concentration in serum harvested from coccygeal blood samples was analyzed in a laboratory and used as a reference value. Capillary samples were obtained from the skin of the exterior vulva by using 1 of 3 different lancets. In all samples, the concentration of BHBA was immediately analyzed with all 3 hand-held devices used in random order. All lancets used in the study were eligible for capillary blood collection but differed in the total number of incisions needed. Spearman correlation coefficients between the BHBA concentrations in capillary blood and the reference test were highly significant with 83% for the FSP, 73% for the NOV, and 63% for the GLX. Using capillary blood, the FSP overestimated the mean BHBA concentration compared with the reference test (+0.08 mmol/L), whereas the GLX and NOV underestimated the mean concentration (−0.07 and −0.01 mmol/L). When a BHBA concentration of 1.2 mmol/L in serum was used to define subclinical ketosis, the corresponding analyses of receiver operating characteristics resulted in optimized thresholds for capillary blood of 1.1 mmol/L for the NOV and GLX devices, and of 1.0 mmol/L for the FSP. Based on these thresholds, sensitivities (Se) and specificities (Sp) were 89 and 84%

for the NOV, 80 and 89% for the GLX, and 100 and 76% for the FSP. Based on a serum BHBA concentration of 1.4 mmol/L, analyses of receiver operating characteristics resulted in optimized cut-offs of 1.4 mmol/L for the FSP (Se 100%, Sp 92%), 1.3 mmol/L for the NOV (Se 80%, Sp 95%), and 1.1 mmol/L (Se 90%, Sp 85%) for the GLX. Using these optimized thresholds for the specific hand-held meters, no significant differences between the devices in Se and Sp to detect subclinical ketosis in coccygeal blood were observed. Calculated test characteristics for analyzing capillary blood using the hand-held devices were numerically smaller compared with blood obtained from a coccygeal vessel, but overlapping confidence intervals indicate no statistical difference between the origin of the sample. Hence, this procedure seems to be suitable for ketosis monitoring in dairy cows, but further validation with more data from different farms is recommended.

Key words: cow, ketosis, capillary blood, β -hydroxybutyrate, diagnostic test

INTRODUCTION

Subclinical ketosis (SCK) is defined as a metabolic disorder with an increased ketone body concentration in the absence of clinical symptoms of ketosis (Andersson, 1988; Duffield et al., 1998; Rollin et al., 2010). Commonly used thresholds to define SCK are BHBA concentrations in blood of 1.2 and 1.4 mmol/L (Geishauser et al., 1998; Duffield et al., 2009). At herd level, a target incidence of SCK below 20% (using a cut-off of the BHBA concentration in serum of 1.4 mmol/L) was recommended by Cook et al. (2006).

The occurrence of SCK in dairy herds in the periparturient period, caused by a negative energy balance, represents an important challenge for dairy farmers. Several studies have shown that SCK is associated with an increased risk for the occurrence of secondary dis-

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eases such as clinical ketosis, displaced abomasum, metritis, mastitis, and lameness (Geishauser et al., 1998; Duffield et al., 2009; Suthar et al., 2013). Additionally, decreased milk yield (Dohoo and Martin, 1984; Duffield et al., 1997) and impaired reproductive performance (Walsh et al., 2007a,b; Chapinal et al., 2012) are associated with the occurrence of SCK. Based on BHBA concentrations in blood of ≥ 1.2 mmol/L, Suthar et al. (2013) reported an overall prevalence of SCK for 10 European countries of 21.8%, ranging from 11.2 to 36.6%, within 2 wk after calving. Reported prevalence for North American dairy herds ranged from 8.9 to 43.2% within the first 2 mo of lactation (Dohoo and Martin, 1984; Geishauser et al., 1998; McArt et al., 2012). Recently, an increased BHBA concentration before calving has been associated with a detrimental effect on milk yield and animal health (Chapinal et al., 2011, 2012). Animals showing a BHBA concentration in serum ≥ 0.7 mmol/L within the last week of gestation were at higher risk of early culling (Roberts et al., 2012).

Considering the abovementioned aspects, monitoring of dairy herds for SCK is reported to be an appropriate measure for disease prevention and improvement of stock management efficiency in dairy farming (Cook et al., 2006). In several studies, the determination of BHBA concentrations in serum or plasma with standard laboratory methods was defined as a reference test for diagnosing of SCK (Duffield et al., 1998; Geishauser et al., 2000). This method, however, is inconvenient for a broader surveillance because of its costs in terms of time and money. The possible delay in treatment of animals suffering from SCK, because of shipping and analyzing a blood sample at an external laboratory, might have a negative effect on animal welfare as well. Routine testing of animals at risk for SCK by the farmer can be used on a cow level (to guide individual treatment) as well as on a herd level (to evaluate transition management and feeding).

Within the last 2 decades, several point-of-care tests have been developed and were evaluated for dairy cows to detect ketones in urine (Carrier et al., 2004; Krogh et al., 2011), milk (Geishauser et al., 1998, 2000; Carrier et al., 2004; Krogh et al., 2011), and whole blood (Iwersen et al., 2009, 2013; Mahrt et al., 2014b). Cow-side ketone tests for blood may be preferred because they are most close to the reference test for SCK based on BHBA concentrations in serum or plasma (Geishauser et al., 1998, 2000; Carrier et al., 2004; Iwersen et al., 2009, 2013). Only few studies, however, performed a direct comparison between different hand-held meters for detection of SCK (Iwersen et al., 2013).

To our knowledge, only venous or arterial blood samples or both have been evaluated for monitoring

of ketosis using electronic hand-held devices. A disadvantage of this testing method is its more invasive sampling technique compared with milk- and urine-based systems. Additionally, in many countries (e.g., Germany, Switzerland, and the Netherlands), national legislation prohibits conventional blood sampling by laypersons (e.g., farmers). Capillary blood might be an alternative, as sampling is considered less invasive and easier to achieve compared with the conventional blood-sampling procedures. The permission of obtaining capillary blood by the farmer using a minimally invasive technique for diagnostic purposes is already in consideration by the authorities in Austria, for instance.

The primary objective of this study was to test whether capillary blood obtained from the skin of the exterior vulva by using a minimally invasive lancet technique is suitable to detect SCK in pre- and postpartum dairy cows. Secondary objectives were to test 3 different lancets for obtaining the capillary blood and to test 3 commercially available hand-held devices within the same experiment. The BHBA concentrations in coccygeal blood were analyzed in a laboratory to be used as reference value and assessed with the hand-held devices to distinguish between the effect of the device and the type of blood used for the cow-side testing.

MATERIALS AND METHODS

Experimental Design

The study was approved by the institutional ethics committee of the University of Veterinary Medicine, Vienna, and the national authority according to § 26 of the Law for Animal Experiments, Tierversuchsgesetz 2012 – TVG 2012 (GZ 68.205/0007-II/3b/2014) as well as by the Slovakian Regional Veterinary Food Administration (428/2014). The study was conducted on 4 sampling dates between March and April 2014 on a Slovakian dairy farm, keeping approximately 2,700 Holstein-Friesian cows and additional youngstock. Cows were housed in freestall barns with high bed cubicles. Rubber mats with dried slurry separator material were used as cubicle bedding. The average ECM yield (based on 4.0% butterfat and 3.4% protein) was 9,165 kg in 2013.

In total, 240 primi- and multiparous cows predominantly within the transition period were enrolled in this study. Each cow was only tested at one sampling date within the study period. At each of the 4 farm visits, samples of approximately 50 randomly selected animals in the fresh cow pen and of approximately 12 randomly selected cows in the close-up pen were taken.

The final data set used for the statistical analyses consisted of 34 (14.2%) animals in first lactation, 97

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