# Comparison of beta-hydroxybutryate determination by Fossomatic milk analysis to milk and blood tests in postpartum dairy cattle

**D.J. Wilson**,<sup>1</sup> DVM, MS, PhD, DACVPM; **G.M. Goodell**,<sup>2</sup> DVM <sup>1</sup>Utah State University, Logan, UT 84322 <sup>2</sup>The Dairy Authority, Greeley, CO 80634

## Introduction

A common method for detection of ketosis in early postpartum dairy cattle, which is usually associated with negative energy balance, is the measurement of the ketone beta-hydroxybutyrate (BHB) in blood or milk. The primary objective of this study was to compare the agreement among 4 test methods used to detect BHB in blood and milk samples and classify whether the cow in question has ketosis. The test methods assessed were BHB blood test strips tested in a Precision XTRA® meter, milk test strips (Keto-Test®), DHIA milk meter-collected samples tested with a Fossomatic milk analyzer (Foss), and hand-stripped milk samples tested with a Foss. The secondary objective of this study was to compare milk-test methods for classifying the ketosis status of postpartum dairy cows. Fossomatic milk BHB testing is not commercially available in the US.

## **Materials and Methods**

A preliminary study found a negligible number of ketotic cows after 14 days-in-milk (DIM); therefore, no cows >14 DIM were studied. On 5 commercial dairy herds, blood and milk samples were obtained from 444 Holstein cows from 1 to 14 DIM on the same day as the DHIA test milk samples (DHIA milk samples were unavailable for one 80-cow herd, and hand-stripped milk samples were unavailable for an additional 28 cows). Cows were restrained in lock-up stalls and bled from the coccygeal vein. Milk samples were collected during the next milking. A small number of milk samples were of insufficient volume to complete all milk testing.

Three test methods produced continuous results, while the milk test strip produced categorical results of 0, 50, 100, 200, 500, 1000 µmol of BHB/L. All 4 BHB tests utilized a specific threshold to classify cows categorically as ketotic or nonketotic. Analyses compared each pair of BHB tests' agreement (concordant [C]) or disagreement (discordant [D]) in categorizing the ketosis status of each cow. The extent of agreement beyond chance between each possible pair of test methods was assessed with the kappa ( $\kappa$ ) statistic.

## Results

Cows were classified as ketotic (K) or non-ketotic (N) by use of test-specific threshold values of  $\geq 1,200$  $\mu mol$  of BHB/L for the blood test strips, and  $\geq 200 \ \mu mol$ of BHB/L for each of the other 3 test methods (milk tests strips and DHIS milk meter-collected and handstripped milk samples analyzed with a Foss). Of the 446 blood samples analyzed with blood test strips, 53 cows were classified as ketotic and 393 cows were classified as nonketotic. Of the 442 milk samples analyzed with the milk test strips, 34 cows were classified as ketotic and 408 cows were classified as nonketotic. Of the 425 DHIA milk meter-collected milk samples analyzed with Foss, 29 cows were classified as ketotic and 396 were classified as nonketotic. Of the 338 hand-stripped milk samples analyzed with Foss, 41 cows were classified as ketotic and 297 cows were classified as nonketotic.

When the blood strip test results were compared with the milk strip test results, there were 15 cows classified as ketotic by both tests, 371 cows classified as nonketotic by both tests, 37 cows classified as ketotic by the blood test strip and nonketotic by the milk test strip, 19 cows classified as nonketotic by the blood test strip and ketotic by the milk test strip, and 87% and 13% of cows had concordant and discordant test results, respectively. When blood strip test results were compared with the milk meter Foss results, there were 10 cows classified as ketotic by both tests, 357 cows classified as nonketotic by both tests, 42 cows classified as ketotic by the blood test strip and nonketotic by the milk meter Foss, 19 cows classified as nonketotic by the blood test strip and ketotic by the milk meter Foss, and 86% and 14% of cows had concordant and discordant test results, respectively. When blood strip test results were compared with the hand-stripped Foss results, there were 10 cows classified as ketotic by both tests, 266 cows classified as nonketotic by both tests, 31 cows classified as ketotic by the blood test strip and nonketotic by the hand-stripped Foss, 31 cows classified as nonketotic by the blood test strip and ketotic by the hand-stripped Foss, and 82% and 18% of cows had concordant and discordant test results, respectively. When milk strip test results were compared

© Copyright American Association of Bovine Practitioners; open access distribution

with the hand-stripped Foss results, there were 16 cows classified as ketotic by both tests, 287 cows classified as nonketotic by both tests, 10 cows classified as ketotic by the milk strip test and nonketotic by the hand-stripped Foss, 25 cows classified as nonketotic by the milk strip test and ketotic by the hand-stripped Foss, and 90% and 10% of cows had concordant and discordant test results, respectively. When milk meter Foss results were compared with the hand-stripped Foss results, there were 5 cows classified as ketotic by both tests, 272 cows classified as nonketotic by both tests, 20 cows classified as ketotic by the milk meter Foss and nonketotic by the hand-stripped Foss, 33 cows classified as nonketotic by the milk meter Foss and ketotic by the hand-stripped Foss, and 84% and 16% of cows had concordant and discordant test results, respectively.

Kappa evaluation of test agreement was considered poor for the comparisons of blood strip test with milk meter Foss ( $\kappa = 0.18$ ), blood strip test with hand-stripped Foss ( $\kappa = 0.14$ ), milk strip test with milk meter Foss ( $\kappa =$ 0.08), and milk meter Foss with hand-stripped Foss ( $\kappa =$ 0.07); fair for the comparison of blood strip test with milk strip test ( $\kappa = 0.28$ ); and moderate for the comparison of milk strip test with hand-stripped Foss ( $\kappa = 0.42$ ). Sensitivity (Se) and specificity (Sp) are relative to the "gold standard" test. With the blood test strips used as the gold standard for "true" ketosis status, the Se and Sp of the other 3 tests were as follows: milk test strips, Se = 29%, Sp = 95%; milk meter Foss, Se = 19%, Sp = 95%; hand-stripped Foss, Se = 24%, Sp = 90%. With milk test strips used as the gold standard, the Se and Sp for the milk meter Foss were 21% and 94%, respectively, and those for the hand-stripped Foss were 62% and 92%, respectively.

## Significance

Results of this study indicated that measurement of BHB in milk classified fewer postpartum cows as ketotic, compared with measurement of BHB in blood, which is in agreement with results of other studies. The results of the test methods evaluated in this study agreed well for classification of nonketotic cows, but did not agree as well for classification of ketotic cows. Investigation of improved calibration for Fossomatic testing of BHB in milk is indicated.