

An Evaluation of The PortaSCC® Test As A Measure of Udder Health Status Dairy Cows (An Excerpt From A Technical Report)*

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Introduction

Mastitis is one of the most costly diseases of dairy cows (DeGraves and Fetrow, 1993; Fetrow, 2000). The National Mastitis Council (NMC) has developed and extended udder health management programs involving strategies to both eliminate and prevent intramammary infections (IMI), as well as to monitor udder health status (National Mastitis Council, 2002). Determination of udder health status has traditionally involved the use of time-consuming laboratory tests such as bacteriological culture and somatic cell count (SCC) determination. These tests are very reliable and relatively accurate (Radostitis et al., 1994). The expenses incurred, and logistic considerations involved, render these tests inconvenient on an individual cow basis. Yet, alternative options for effective, rapid, inexpensive on-farm evaluation of udder health status have been limited. Therefore, the California Mastitis Test (CMT) was often used on-farm as a low-cost method to assess of udder health status during lactation. Recent studies have examined the sensitivity and specificity of the CMT as a means of identifying infected cows in the immediate postpartum period (Sargeant et al., 2001; Dingwell et al., 2003). Most optimal results were reported from samples taken on day 3 (Sargeant et al., 2001) and day 4 (Dingwell et al., 2003). However, in both studies, the ability of the CMT to detect intramammary infection (IMI) was marginal.

A new rapid, cow-side test has been developed for the detection of sub-clinical mastitis (PortaSCC®). A color change in the test pad provided with this test correlates to the level of white blood cells in the milk sample. The PortaSCC® test has been reported to have high sensitivity and specificity when SCC was used as the gold standard (Thacker et al., 2000).

The CMT and PortaSCC® are management tools for the assessment of udder health status that may fill a critically important role in the near future. There is a growing concern regarding the widespread use of antibiotics in food production systems and the issue of antimicrobial resistance. As a result, there has been increased emphasis on the selective use of dry cow antibiotic therapy. New strategies to prevent IMI, such as the use of external teat sealants and internal teat sealers have been developed and tested (Huxley et al., 2002; Godden et al., 2003). Successful use of an internal sealer at drying off, without the use of antibiotic therapy, would depend on knowledge of udder health status prior to application. The sealer would not be expected to be effective in eliminating an existing infection. Therefore a rapid, cow-side test with excellent sensitivity and specificity may aid producers in the selection of cows to receive dry cow therapy on the day of milk cessation.

It was hypothesized that the PortaSCC® test would provide an accurate assessment of udder health status at a minimal cost. Thus, the primary objective of this study was to determine the test characteristics of the PortaSCC®, using SCC as the gold standard. An additional objective was to assess the ease of use, and potential for implementation of this test in udder health management programs.

Methods

Quarter or composite milk samples were aseptically collected from cows at various stages of lactation as part of a routine udder health management surveillance program in herds serviced by the University of Guelph Ruminant Field Service Clinic. Animals were selected for this study on the basis of elevated, or changing SCC from the individual cow SCC results from the Ontario Dairy Herd Improvement (DHI) Corporation. In the same situations, the CMT was used to select specific quarters for further evaluation. This study was conducted from May to July 2002.

All samples were analyzed for SCC using a Bentley SomaCount 300 (Bentley Instruments Inc., Chaska, Minnesota). For the purposes of this study the SCC cut-point that defined disease was > 200, 000 cells/ml (Dohoo and Leslie, 1991). Each sample was then tested using the PortaSCC®.

Following manufacturers guidelines, a single-use pipette was used to place one to two drops of the milk sample onto the absorptive area of the test. Once the milk was completely absorbed, four drops of wash solution were individually placed and absorbed into the test area. Once the final drop was added and absorbed, the test was incubated at room temperature for ten minutes. Following incubation, the color of the test area was observed and recorded. If no color change was observed, the result was considered negative, or zero. If the test area showed a light blue color, the result was recorded as 'one'. If the test area changed to an aqua color the result was considered 'two'. Finally, a color change to royal blue was recorded as 'three'. The recorded values of one, two and three are designed to correlate to an SCC value of 200,000 cells/ml, 500,000 cells/ml and 1,000,000 cells/ml, respectively. Any reaction greater than or equal to one was considered a positive test result. Any change in colour of the test pad (indicating a cell count > 200, 000 cells/ml) was considered a positive PortaSCC® result.

Results

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A total of 622 quarter or composite milk samples from 200 cows in 10 herds were available for evaluation and analysis. Of these, 615 samples were evaluated using both the PortaSCC® test and electronic SCC determination.

The PortaSCC® test results compared to the gold standard SCC determination by the electronic Bentley Cell Counter. The PortaSCC® provided negative results on 482 samples and positive results on the remaining 133. Similarly, the gold standard SCC determination indicated 138 samples to be considered positive ($> 200,000$ cells/ml) and the remaining 477 to be negative. From a comparison of these results, it was calculated that 33 out of 482 negative samples were false negative according to the PortaSCC® test results. Additionally, 28 out of 133 positive samples from the PortaSCC® test were incorrectly classified as positive (false positive).

Thus, the calculated test characteristics of PortaSCC® using a cut-point of $> 200,000$ cells/ml on the standard SCC determination as the gold standard. The sensitivity and specificity of the PortaSCC® were 74% and 94%, respectively. The predictive value (PV) of a negative test was 93%. In addition, the positive predictive value was determined to be 79%.

The Spearman rank association test revealed the $\ln(\text{SCC})$ and the PortaSCC® test results were highly correlated ($R_2 = 0.63$, $P = < 0.001$).

Discussion

The sensitivity of the PortaSCC® was defined as the ability of PortaSCC® to detect the presence of a SCC $> 200,000$ cells/ml. This is calculated as the proportion of quarters that had a positive PortaSCC® test out of all of the quarters with a SCC $> 200,000$ cells/ml. The specificity of the PortaSCC® is the ability to detect quarters that have a SCC $< 200,000$ cells/ml. This is calculated as the proportion of low SCC quarters ($< 200,000$ cells/ml) that had a negative PortaSCC® test. In combination, these two test characteristics indicate how well the PortaSCC® can discriminate between low SCC and high SCC quarters (Martin et al., 1987). In general, it is ideal that screening tests have a high sensitivity to avoid false negative reactions. This would ensure that all high SCC quarters were identified appropriately. However, it is also important to couple high sensitivity with high specificity. A highly specific test would avoid false positives, and thus minimizing taking action on cows unnecessarily.

Based on these data, the calculated sensitivity was marginal at 76%. Thus suggesting, that 24% of tested quarters would be falsely considered negative. However, it is noteworthy to mention that many of these milk samples were either extremely thick or demonstrated significant clotting. It was clearly visible that these milk samples prevented proper absorption into the test pad area of the PortaSCC® test. It is likely that improper absorption would result in inaccurate test function and a false negative result. The average SCC for these false negative samples was 680,097 cells/ml, while the average of the true positive samples was 780,481 cells/ml. These findings reflect the possibility that severe mastitis may cause inaccurate test readings. However, in an on-farm situation, it is likely that this test need not be run for cows with obvious signs of mastitis.

The specificity of the PortaSCC® was determined to be 94%; suggesting that 6% of tested quarters would falsely be considered as positive. This is considered to be a high specificity. However, these results could hypothetically mean that antibiotic treatment may be administered to cows that truly were negative. The average SCC for the false positive samples was 123,286 cells/ml, while the average SCC for the true negatives was 41,054 cells/ml. These numbers illustrate that the PortaSCC® test works well when detecting very low SCCs, but has a narrow grey zone for detecting borderline SCCs. Regardless, this high specificity would be beneficial to a producer looking to utilize this test for selecting negative quarters.

The sensitivity and specificity of the test, as well as the prevalence of the disease within the population, influence the calculated predictive values (Martin et al., 1987). Therefore, it is difficult to examine these test characteristics stand-alone without considering the associated variables. Regardless, the predictive value of a negative PortaSCC® test was very high, at 93%. This high negative predictive value ensures that cows or quarters that are indeed healthy ($< 200,000$ somatic cells/ml) are appropriately identified. Conversely, the positive predictive value was 76%. Although this seems marginal, it must be emphasized that a number of thick and clotted samples that were obviously positive for elevated SCC did not allow the filter pad to function, and affected test performance. Therefore, it is hypothesized that the on-farm use of this test, where cows with obvious signs of mastitis would not be evaluated, would produce a better positive predictive value.

In summary, the PortaSCC® test is easy to use, (provided the test package includes single-use plastic pipettes) and it provides accurate, relatively rapid results for estimation of SCC in milk. With the resurgence in research examining the CMT test as a cow-side method of determining udder health status (Wallace et al., 2004; Dingwell et al., 2003; Sargeant et al., 2001), and the increased emphasis on production of low SCC milk, there is a need for highly sensitive and specific diagnostic tools to predict SCC. The PortaSCC® shows considerable promise as a useful management tool to monitor individual cows for intramammary inflammation.

Conclusions

The PortaSCC® test is a fast and accurate method of SCC estimation that can be used on-farm. As with actual electronic SCC, it is not an extremely accurate method for determining bacterial IMI. While some adjustments need to be made in its packaging (inclusion of pipettes), the PortaSCC® is both sensitive and specific in its test characteristics. The tests high predictive value for a negative test also enhances its potential future use by dairy producers in their dry cow treatment decision-making protocol.

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