Title

Evaluation of cow-side meters to determine somatic cell count in individual cow quarter and bulk-tank milk samples

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Evaluation of cow-side meters to determine somatic cell count in individual cow quarter and bulk-tank milk samples

By

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THESIS

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ABSTRACT

Intramammary infections, which cause mastitis, can increase treatment and labor costs, decrease milk production, and affect milk quality. Meters that measure quarter somatic cell count (SCC) could be used to make more informed dry cow therapy decisions. The objective of this study was to compare the RT-10 iPhone adapter (RT-10; Dairy Quality Inc., Newmarket, Canada), DeLaval Cell Counter (DSCC; DeLaval, Graiguecullen, Carlow), Porta Check Quick Test (PortaCheck, NJ), California Mastitis Test (ImmuCell, Portland, ME), pH meter (Hanna Instruments, RI), electrical conductivity meter (OHAUS, NJ), and the dual laser infrared temperature thermometer (Klein Tools, IL) for measuring SCC in individual quarters in comparison to a reference standard, the Fourier Transform Spectrometer 600 Combi System (Combi; Bentley Instruments, MN). Meters were evaluated using individual cow quarter samples and bulk-tank samples to measure SCC. To test individual quarter SCC, individual quarter milk samples from 160 cows from 4 commercial dairy herds (40 cows per herd) were collected just before cessation of milking and tested within 4 h of collection. To test bulk-tank SCC, 100 bulk-tank milk samples (25 mL) were collected from UC Davis VMTRC Milk Quality Lab. Meter SCC values were regressed on observed Combi SCC using PROC GLM (SAS Institute, 2021 v. 9.4). Then goodness of fit was evaluated by partitioning of the mean square predicted error (MSPE). For individual quarter SCC, RT-10 had the highest coefficient of determination (R^2 = 0.86), lowest mean square predicted error and highest proportion of MSPE due to random variation (96 %). Both the RT-10 and DSCC had the highest sensitivity and specificity for identifying quarter SCC above and below 200,000 cells/mL. For bulk-tank SCC, DSCC had the highest coefficient of determination (R^2 = 0.45), lowest mean square predicted error, and highest proportion of MSPE due to random variation (80 %). The RT-10 and DSCC could be used to measure individual quarter SCC to determine which cows to treat at cessation of lactation potentially reducing antibiotic use.

KEYWORDS: mastitis, milk meter, somatic cell count

Literature Review

Introduction

Intramammary infections (IMI), which cause mastitis, are an extensive problem in the dairy industry as these diseases can greatly decrease milk production and milk quality, affect cow health, and increase treatment and labor costs (Halasa, 2007). Somatic cell count (SCC) is a simple way to detect these infections, as a high SCC can suggest a subclinical (SCM) or clinical (CM) infection. The National Mastitis Council has stated that a healthy, or an uninfected quarter, is around 100,000 cells/ml, whereas an SCC of 200,000 cells/ml or greater is a subclinical or clinical infection of the quarter (NMC, 2001). Producers evaluate SCC on an individual cow level and, through bulk-tank SCC, predict their milking herd's overall average SCC. A Grade "A" Pasteurized Milk Ordinance was implemented in 1993 by the United States Department of Agriculture (USDA), which established a threshold of 750,000 cells/ml for bulk-tank SCC and since then the United States has vastly decreased the overall SCC (USDA, 2019). A common farm practice to monitor overall herd risk of intramammary infections (IMI) and milk quality is to implement monthly testing of bulk-tank SCC by using local milk quality laboratories. Throughout the years, bulk-tank SCC has gradually been decreasing due to improved herd management and milking procedure practices, as well as better milk testing technologies and availability. Silva-del-Rio and Collar (2010) reported bulk-tank SCC mean of 199,000 cells/mL in California. Monitoring both individual cow SCC and bulk-tank SCC can provide an opportunity for dairy producers to improve milk quality, production, cow health, and lower drug use and treatment costs.

Producers have multiple dry-off options that are available to combat IMI, such as the use of blanket and selective therapies. Blanket dry cow therapy, the practice of treating every cow at dry-off with antibiotics, has been the most prominent dry cow therapy strategy used, with 94.2 percent of large dairy operations using this particular method (USDA, 2014). The objective of a selective dry cow therapy program is to selectively provide therapy to cows identified with increased risk of IMI during the dry-off period and in the next lactation, while maintaining cow health and milk production for the herd. Cows with a current infection status, low milk production, and those who have exceeded days in milk (DIM) are all eligible for selective dry cow treatment. The use of selective dry cow strategy could eliminate up to 60% of antibiotics used for dry cows without adversely affecting milk production or health outcomes (Dairy Herd Management, 2018).

There are various cow-side tests that are available to the dairy industry that can evaluate and determine SCC at the individual cow level, which are both easy to handle and cost effective. Some common instruments developed for SCC detection are the Fourier Transform Spectrometer 600 Combi system, Fossomatic 400, bright-field microscopy, DeLaval Cell Counter, RT-10 iPhone adapter, pH meter, Electrical Conductivity meter, and a dual laser infrared temperature thermometer. Other common cow-side tests such as the California Mastitis Test, and PortaCheck Quick Test. The cow-side tests and instruments are described by meter cost, cost per sample, sensitivity (SE), and specificity (SP) in Table 1. Very few studies have examined the performance of these tests using the same cow quarter samples under the same conditions and across multiple herds (Adkins and Middleton, 2018; Albrechtesen *et al*., 2011; Enger *et al*., 2020; Kandeel *et al*. 2019 A; Kandeel *et al*. 2019 B; Norberg *et al*., 2004; Polat *et al*., 2010;

Rodrigues *et al*., 2009). The objective of this paper is to compare these cow-side tests to allow the dairy industry to better evaluate individual quarter and composite SCC to make more informed and beneficial dry cow therapy decisions for their herds. The following sections describe each of the most common cow-side tests and provides an accompanying technical review of each.

FTS 600 Combi System

The FTS 600 Combi System (Combi; Bentley Instruments, Chaska, Minnesota) is owned and operated by the Tulare Dairy Herd Improvement Association (DHIA) and is used for the determination of SCC and milk composition. The system is a combination of two instruments, a Fourier Transform Spectrometer and a Flow Cytometer. Infrared technology is used to provide an analysis of raw or preserved milk components and measures 500 samples per hour. The Fourier Transform Spectrometer module uses light to identify compounds within milk by measuring their spectrum for SCC determination. Milk quality laboratories are able to identify IMI causing pathogens within milk and record incidences using this technology.

The Flow Cytometer module uses a laser to determine SCC in milk. In this module, the milk sample is treated with a buffer solution that stains the cells within the milk with a fluorescent dye. These stained cells are then injected into a focusing window where a laser beam intersects the flow to have the cells emit a fluorescent light, which is then read by a computer. This technology can be used to detect, identify and count specific cells. Gunasekera *et al*. (2003) demonstrated that flow cytometry, along with flourescence techniques, provided both efficient and quality measuring abilities for milk analysis. Gunasekera analyzed raw and ultraheat-treated milk samples, and removed lipids by centrifugation as to not affect SCC analysis. The samples were then stained with a dual-fluorescence Gram Stain Kit (hexidium iodide, and SYTO 9)

(Moleculare Probes, Bioscientific), and were analyzed for SCC by using flow cytometery with the FACSCalibur (Becton Dickinson, Sydney, Austrailia), and the Fossomatic 400 (Foss Electric, Hillerooed, Denmark) for SCC comparison. The Fossomatic records the electrical pulse generated from SCC stained with ethidium bromide (Gonzalo et al., 1993). Gunasekera compared both the FACSCalibur and the Fossomatic to bright-field microscopy for direct cell counts for SCC determination. Results showed that there was no significant difference ($P \geq 0.05$) between the FACSCalibur and direct microscopy in the ultraheat-treated milk samples. Results for raw milk samples, however, showed a correlation coefficient of r = 0.88 between the FACSCalibur and direct microscopy. Good correlation (r = 0.98) was also shown between the FACSCalibur and the Fossomatic for SCC analysis (Gunasekera et al., 2003). By comparing flow cytometry with fluorescent technology, Gunasekera found that flow cytometers can be used for the determination of SCC, however recent research has shifted to cow-side instruments and tests as these can be more cost effective.

By using the both the Fourier Transform Spectrometer and the Flow Cytometer, milk quality laboratories can efficiently report IMI incidences, and SCC for dairy producers. However, it is not financially feasible for dairy producers to own the Combi to test their own milk samples as system is expensive, and the on-farm environment is not the ideal location to house this type of equipment as dusty, moldy, or damp environments can lead to false results or machine errors. Laboratories, such as DHIA, are available to test milk samples from multiple dairies in a time efficient manner and in a suitable environment. Therefore, cow-side tests that are accurate and cost effective are needed on-farm to identify cows that have a current mammary infection.

DeLaval Cell Counter & RT-10 iPhone adapter

The DeLaval Cell Counter (DSCC; DeLaval, Graiguecullen, Carlow), and the RT-10 iPhone adapter (RT-10; Dairy Quality Inc., Newmarket, Canada), use the same fluorescent technology, with the main difference being the RT-10's ability to use the camera of the iPhone to view and take pictures of the stained cells. These portable and efficient meters are available to producers for SCC evaluation. The DSCC and RT-10 both utilize a fluorescent reagent, propidium iodide, to stain the cell's nuclei to correctly identify somatic cells.

Gonzalo *et al*. (2006) compared the DSCC to the Fossomatic to evaluate the performance of the DSCC in diluted and undiluted ovine milk under various staining solutions and preservation conditions. Gonzalo sampled 29 composite ovine milk samples, and unpreserved milk sampled by the Fossomatic and the DSCC were highly correlated (r = 0.96 to 0.97). Results also showed that the accuracy of the DSCC was better suited to decreased SCC values, compared to the Fossomatic that had more accuracy with elevated SCC (Gonzalo, 2006).

pH meter

The Hanna Combo pH & electrical conductivity handheld meter (HC; Hanna Instruments, RI) is commercially available to dairy producers for evaluation of herd health status. Atasever *et al*. (2010) determined the relationship between pH and SCC by using raw milk samples. The SCC was determined using direct microscopy, which showed that as SCC increased, pH values declined, showing as a negative correlation between SCC and pH (r = - 0.523) (Atasever *et al*., 2010). Though this study showed a negative correlation between pH and SCC, the mean pH value of the raw milk samples was decreased, ranging between 6.20 and 6.67, when compared to the pH ranges reported by Ogola *et al*. (2004). The range of milk pH can vary between 6.63 to 6.81 and SCC generally appears to have no effect on the pH of quarter milk samples according to Ogola *et al*. (2004). This could be due to the variability in Na + within the milk because of the difference between positively and negatively charged ions within the plasma (Constable, 1997). While pH has been used previously to determine IMI, other instruments and tests may be better suited for SCC detection.

Electrical Conductivity meter

An Ohaus Starter Pen Meter ST10C-C (ECM; Ohaus, Parsippany, New Jersey) is a portable and waterproof meter that is available for IMI assessment. Electrical conductivity measures the concentration of anions and cations, such as Na + and Cl -, that are secreted into the mammary cells by active and passive transport. Tight junctions of the secretory cells and capillaries of the mammary gland can become damaged by physical trauma or IMI, which increases their permeability. The concentrations of these molecules are higher in blood than in milk (Wong, 1988), which could account for the increase in Na + and Cl – found within milk classified as SCM or CM (Kitchen, 1981). In cows classified as healthy, the normal electrical conductivity of milk has a range of 4.0 to 5.5 mS, but cannot account for the concentration of water within a milk sample and may not be able to detect SCM (Wong, 1988). Since then, other studies have found and updated the electrical conductivity range for healthy. Norberg *et al.* (2004) designated cows as healthy (quarter foremilk samples with no bacteria present, and no veterinary treatment), SCM (quarter foremilk samples with bacteria present, and no veterinary treatment), and CM (received veterinary treatment); and also classified different electrical conductivity traits within the milk. Contrary to Wong (1988), Norberg's study reported a milk electrical conductivity range of 5.5 to 6.5 mS for healthy cows. This elevated range could be due to an increased milk temperature, as electrical conductivity increases with the temperature of milk (Wong, 1988). It has been found that SCM infected cows have a larger conductivity range of 4.83 to 7.03 mS (Isaksson *et al*., 1987) and more recently, 6.45 to 6.85 (Woolford *et al*., 1998). In the case of CM infected cows however, electrical conductivity can range from 5.0 to 9.0 mS (Norberg *et al*., 2004; Hamann and Zecconi, 1998).

IR5 Dual Laser Infrared Temperature Thermometer

The IR5 Dual Laser Infrared Temperature Thermometer (IR5; Klein Tools, Lincolnshire, IL) is able to detect individual quarter temperatures by making use of the dual lasers, which provide a visable reference point on the individual quarter. The thermometer has a temperature range of -30ºC to 400ºC, with udder surface temperatures that range from 25°C to 39°C (Wollowskie *et al*., 2019) and an adjustable emissivity range of 0.10-1.00 based on the level of thermal radiation. This thermometer is non-invasive, as readings can be taken from a safe distance to lessen personal risk and animal stress. A study by Polat *et al*. (2010) evaluated the SCM detection ability of the IRT, and compared results with the CMT. Milk samples and udder skin surface temperatures were taken from 62 lactating Brown Swiss dairy cows. Polat minimized interferences of air flow, light, and humidity by moving cattle individually into a separate room where they were allowed to rest for 30 min before taking udder skin surface temperature measurements and then milk samples for CMT and SCC analysis. Results provided mean values of (ºC) of 33.23, 34.64, 35.73 for quarters that CMT scored + 1, + 2, + 3. A positive correlation was reported between udder skin surface temperature and SCC (r = 0.86), and was found that as CMT scores increased, so did the temperature scores (r = 0.75). This study also reported that SCM quarters had a 2.35ºC greater skin surface temperature than that of healthy quarters, and determined that IRT was sensitive enough to detect SCM through thermal changes on the udder skin (Polat *et al*., 2010).

Sathiyabarathi *et al*. (2016) has reported a narrower range of 37ºC to 38ºC compared to Wollowski *et al*. (2019), and a mean of 38ºC for both SCM and CM. Other versions of the IR5 have been used in previous studies to detect mastitis, have been found to be dependent on ambient temperature, and will compromise the end temperature result (Wollowski *et al.*, 2019; Castro-Costa *et al.*, 2014; Berry *et al.*, 2003;). This instrument has mainly been used by veterinary clinics for to distinguish injuries, and has not been applied to commercial dairies as of yet.

California Mastitis Test

The California Mastitis Test (CMT; ImmuCell, Portland, ME) a cow-side test, establishes SCC by reporting semi-quantitative results to monitor mastitis status and cow health. Leukocytes and neutrophils collect and build-up within the mammary gland to destroy mastitis causing bacteria. The CMT uses a reagent (bromocresol purple) that contains a detergent to disrupt these cell's membranes in order to gel the DNA within the cell's nucleous (University of Missouri, 1993). The CMT reports results based on an ordinal scale of Negative, Trace, + 1 (weak positive), + 2 (distinct positive), + 3 (strong positive), with each scale associated with a range of SCC. A negative result indicates 0 - 200,000 cells/mL, a trace result indicates 150,000 - 500,00 cells/mL, a + 1 result indicates 400,000 - 5,000,000 cells/mL, + 2 result indicates 800,000 - 500,000 cells/mL, and + 3 result indicates > 5,000,000 cells/mL (Schalm and Noorlander, 1956).

Sargeant *et al.* (2001), sampled 131 dairy cows from 3 herds by collecting quarter foremilk samples for SCC analysis by CMT and the Fossomatic. The study determined the CMT's SE and SP for identifying infected quartes at 66.2 (95% CI) and 66.4 (95% CI) respectively for this method, and found that as SCC levels increase, SE decreased and SP increased and found that the CMT can identify individual quarter SCC.

PortaCheck Quick Test

PortaCheck Quick Test (PC; PortaCheck, Moorestown, NJ) is a commercially available cow-side meter that provides semi-quantitative results for dairy producers to monitor cow health and SCC. PC uses a test strip that contains a dye, 3-(*N*-tosyl-lalanyloxy)-indol, which reacts with the enzymes found within the milk. Sampled milk is added to the test strip by using the included pipette, with 4 drops of milk being added directly onto the collection window. The dye then reacts to the enzymes found within in the milk, and after an incubation of 5 min, turns the strip a shade of blue, with the darker the shade of blue, the higher the concentration of SCC in the sample.

Kandeel *et al*. (2019) collected milk sample from cows at dry-off and at freshening to investigate IMI and SCM, and SE and SP by using cow-side tests from PortaCheck, a 45 min color test,the PortaSCC, and a 5 min test, the PC Quick Test. The PortaSCC uses the same dye found in PC, however has an incubation period of 45 min compared to the faster reaction time of 5 min for PC. Milk samples were also analyzed by the California Mastitis Test and by the DSCC for further SCC determination. Kandeel found that the PortaSCC, compared to PC, performed the best at determining SCM sensitivity (SE) and specificity (SP) at dry-off (SE = 0.91, SP = 0.81, respectively) and freshening (SE = 0.79, SP = 0.95, respectively). However, PC performed the best at determining IMI at dry-off (0.81, 0.78, respectively) (Kandeel *et al*., 2019). When comparing PC and CMT cow-side tests, the CMT was worse for determining IMI at dry-off (0.76, 0.60, respectively), but was equivalent to the PortaSCC at freshening. Kandeel found that although PC is clinically useful for identifying CM and SCM in quarter milk samples taken at dry-off and freshening, the cow-side test was not adequately accurate to be chosen over the CMT (Kandeel *et al*., 2019).

Conclusions

While there are many ways to detect SCC in milk, the instruments and tests chosen for this study were the DSCC, RT-10, PC, CMT, ECM, HC, and IR5. These instruments and tests are commercially available for producers, and comparing these on farm will provide a better understanding of their accuracy and precision for detecting IMI in cows, as well as which of these tests may be more practical and efficient for on-farm use. By comparing the applications successes and limitations, the producer will have more information for selecting cow-side SCC technology for guiding individual quarter dry-off treatment decisions which can potentially lead to decreased antibiotic use and good udder health.

Abbreviations

CMT = California Mastitis Test

CM = clinial mastitis

Combi = Fourier Transform Spectrometer 600 Combi System

DIM = day in milk

DSCC = DeLaval Cell Counter

ECM = Electrical Conductivity meter

HC = pH instrument

IMI = intramammary infection

IR5 = Dual Laser Infrared Temperature Thermomer

PC = PortaCheck Quick Test

SCC = somatic cell count

SCM = subclinical mastitis

SE = sensitivity

SP = specificity

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INTERPRETIVE SUMMARY

Meters for measuring somatic cells counts (SCC) such as the RT-10 and Delaval Cell Counter can be used to estimate individual quarter and bulk-tank milk samples SCC, respectively, to help dairy producers evaluate individual cow quarter and herd udder health to identify quarters for selective dry cow therapy potentially reducing antibiotic use and treatment costs at dry-off.

Title: **Evaluation of cow-side meters to determine somatic cell count in individual cow quarter and bulk-tank milk samples**

Running Title: Predict cow quarter SCC

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Declaration of Conflict of Interest

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INTRODUCTION

Dairy producers have multiple treatment options at cessation of milking to combat intramammary infections (**IMI**), such as the use of blanket and selective therapies. Blanket dry-cow therapy, the practice of treating every cow at dry-off with antibiotics, has been the most prominent dry-cow therapy used. In 2014, 80% of dairy herds (and 94.2 % of large dairy operations) used this method (USDA-NAHMS, 2014). However, only 35.6 % of cows at dry-off may have a prevalence of IMI (Pantoja *et al*., 2009). Dairy producers may be unnecessarily treating cows with antibiotics leading to increased treatment costs. Selective dry-cow therapy, treating only udders or quarters that are infected, is an alternative to blanket therapies (Østerås *et al*., 1991). This strategy has been shown to be effective in herds that already have a low risk of IMI (Robert *et al*., 2006). Hommels *et al*. (2021) found that blanket dry cow therapies were slightly more expensive than selective therapies, $ 54.7- $ 58.5 and $ 52.4 - $ 58.2 per cow, respectively and selective dry cow therapy showed a 29 % reduction of antibiotic use in comparison to blanket therapy. Selective dry-cow therapy has the potential to reduce the use of antibiotics used for dry-cows without adversely affecting milk production or health status (Robert *et al*., 2006).

Monitoring both individual cow somatic cell counts (SCC) and bulk tank milk (BTM) can provide an opportunity for dairy producers to improve milk quality, production, cow health, and lower treatment costs (Hillerton and Berry, 2005; Jones and Bailey, 2009). Commercially available cow-side meters can be quick and easy to use but to date no one has compared meter performance using the same samples from different dairies. Viguier *et al*. (2009) reviewed current SCC detection devices and alternative methods for the detection of IMI. Several studies have highlighted the need for tests that are sensitive for SCC detection, easy to use, and that can be measured cow-side (Rodrigues *et al*., 2009; Adkins and Middleton, 2018). Several studies evaluated two to three cow-side tests that can measure SCC (Albrechtesen *et al*., 2011; Enger *et al*., 2020), measure udder temperature (Polat *et al*., 2010), esterase activity (Kandeel *et al*. 2019 A), milk conductivity (Norberg *et al*., 2004), and pH, (Kandeel *et al*. 2019 B) but, few have examined all of these tests under the same conditions from multiple herds.

The objective of this study was to compare and evaluate 7 cow-side meters by using individual quarter milk samples obtained at dry-off, and BTM samples in comparison to a reference standard, the Fourier Transform Spectrometer 600 Combi System (**Combi**; Bentley Instruments, MN). Milk samples from dry-off were chosen because antibiotic treatment decisions for selective or blanket therapies for the cow are made at this time.

MATERIALS AND METHODS

All procedures involving animals were approved by the Animal Care and Use Committee of the University of California, Davis.

Cow management and housing

One hundred and sixty cows from 4 commercial dairy herds (n = 40 per herd) located in Tulare and Fresno County, California were enrolled between October 2020 and August 2021. Cows in Dairies 1 and 3 were housed in dry lots with headlocks. Cows in Dairies 2 and 4 were housed in free stall barns with headlocks. Dairies 1, 3, and 4 milked cows twice a day, whereas Dairy 2 milked cows 3 times a day. The cows selected were all Holsteins, and were enrolled prior to dry-off using each dairy's specific days carried calf (**DCC**) criteria, before receiving any dry-cow treatment, and before the cessation of milking.

Sample size determination

Minimum number of samples for regression is 8 (Jenkins and Quintana-Ascencio, 2020). This study used 40 cows per dairy as sample size and since dairy and cow were not significant contributor variables to the regressions, total sample size was 160 for individual quarter samples and 100 for BTM.

Individual quarter and BTM sampling

Individual quarter milk samples were collected before the morning milking on Dairies 1, 3, and 4. Individual quarter milk samples on Dairy 2 were collected 2 h after the morning milking. To collect each sample, teat ends were wiped with ethanol soaked gauze, and foremilk was discarded. Fifty mL of milk was hand collected into separate tubes for each quarter. Milk samples were then placed on ice, and transported to the lab. Then 25 mL of milk were separated into two different vials, one for bench top analysis and the other for analysis by DHIA using the Combi system.

Bulk-tank milk samples (n = 100) were provided by the UC Davis Veterinary Medicine Teaching and Research Center's Milk Quality Laboratory. No knowledge of the cows, location of dairy, or other management factors of these dairy herds were known, other than sample collection time, at AM or PM. Fifty mL of BTM milk were collected from the laboratory after bacteriological sampling had occurred. Twenty-five mL were aliquoted into separate vials, one for bench top analysis, and the other for DHIA. Only meters RT-10 adapter (**RT-10**; Dairy Quality Inc., Newmarket, Canada), DeLaval Cell Counter (**DSCC**; DeLaval, Graiguecullen, Carlow), PortaCheck Quick Test (**PC**; PortaCheck, NJ), electrical conductivity meter (**ECM**; OHAUS, NJ), and pH meter (**HC**; Hanna Instruments, RI) were used for BTM analysis as the samples were not obtained cow-side.

Meter SCC Analysis

All individual quarter and BTM samples were well mixed for the measurement of SCC. The Combi, which was designated as the reference standard test, was owned and operated by the Tulare Dairy Herd Improvement Association (**DHIA**), and was a combination of two modules, a Fourier Transform Spectrometer and a Flow Cytometer, for the determination of SCC and milk composition. Cells within the milk sample were stained with a fluorescent dye, exposed to a laser beam and the light is refracted in accordance to the amount of somatic cells.

The DSCC was a portable machine that uses cassettes to predict SCC. The RT-10 for the iPhone 5/s used the same cassettes to predict SCC. The cassettes contained a fluorescent reagent (propidium iodide) to stain the cell's nuclei. In the DSCC, the cassette's counting window was exposed to an LED light, a digital camera photographed a picture of the stained nuclei, and then counts them. The DSCC had a measuring range of 10,000 to 4,000,000 somatic cells/mL. In the RT-10, the camera of the iPhone was used to count SCC. There can be a high rate of cartridge failure if the counting window was damaged or smudged when handled, which can affect the accuracy of the device.

The PC used test strips that estimated the number of somatic cells by measuring the esterase enzyme present within white blood cells. The test strip was mixed with milk and an activator solution [3-(*N*-tosyl-lalanyloxy)-indol] (~ 80 µl) in the sample well, which causes a color change to blue, represened the amount of esterase in the sample. A color chart was then used to score somatic cell results in categories of ≤ 100,000; 250,000, 500,000, 750,000, 1,500,000, ≥ 3,000,000 cells/mL.

The California Mastitis Test (**CMT**; ImmuCell, Portland, ME) was a qualitative four-welled plastic paddle that is able to test a cow's individual quarters for SCC. A reagent that broke down cell membranes and contained a pH indicator (bromocresol purple) caused the milk to gel in accordance with the concentration of SCC. Milk and reagent were added to the paddle, and were rotated and tilted until the reaction was completed. Results were scored a Negative, Trace, + 1, + 2, or a + 3 according to the standards set by the manufacturer.

The ECM used electrodes to measure the resistance or density of milk, which was electrically positive. This model had a range of 0.00 – 19.99 S/m, and an accuracy of ± 2.5 % femtosecond.

The HC used an electrode to measure the pH of milk. Changes in milk pH were due to compositional changes, such as extracellular fluid components and blood that led to an increase in pH. This meter had a range of 0 - 14 pH, and an accuracy of ± 0.05 pH.

Individual quarter temperatures were taken 1 h after sampling using a dual infrared temperature thermometer (**IR5**; Klein Tools, IL). The device used dual laser beams as a focal point for the temperature sensor on the front part of the tool for individual quarter temperature determination. The emissivity level was set to 0.98, the emissivity value used to measure the temperature of human skin (Jones and Plassman, 2002). This meter was aimed at the caudal area of each quarter. Temperature readings were able to be taken from a safe distance (~ 1.5 m) from the cow due to the dual laser to lessen personal risk and cow stress.

A common practice for dairies to decide which cows to treat at dry-off is previous month SCC (**PSCC**; Lago *et al*. 2004) by using machines such as the Combi. However, if the days between the previous test date and the dry-off data (**DSLT**) are far apart, PSCC may change as infections may be hard to detect inbetween SCC test and dry-off (de Haas, *et al*., 2002). Dairy 1 PSCC and DSLT data were used to evaluate observed SCC because this dairy recorded PSCC.

Statistical Analysis

The unit of interest in this study was mammary individual quarter and BTM sample. To determine how well meters were able to measure individual quarter SCC measured by the Combi, SCC values of the other meters using PROC GLM in SAS (SAS v. 9.4; Statistical Analysis System, SAS Inst. Inc., Cary, NC) were each regressed on Combi SCC. The independent variable was Combi SCC, with dependent variables of RT-10, DSCC, PC, CMT, ECM, HC, or IR5 SCC. The same regression was run again with co-variates dairy, cow, milk protein, fat, lactose, solids non-fat (**SNF**), and milk urea nitrogen (**MUN**) included in the equations to determine if there were environmental or milk composition effects on meter performance. Regression covariates were then eliminated using backwards elimination. Milk protein and lactose improved agreement of Combi SCC for meters DSCC, PC, CMT, ECM, HC, and IR5.

To evaluate the practice of using previous month SCC at test date (**PSCC**) in Dairy 1, Combi SCC were regressed on days since last test (**DSLT**) and PSCC using the PROC GLM procedure in SAS. Combi SCC had to be averaged by cow for each monthly test date since PSCC is a composite sample from the mammary gland. Since DSLT was not a significant contributor to Combi SCC, it was dropped from the regression, so the dependent variable was Combi SCC, with independent variable PSCC.

To evaluate how well meters predicted BTM SCC, the SCC estimated by the meters RT-10, DSCC, ECM, and HC were regressed on Combi SCC using the PROC GLM procedure of SAS.

The dependent variable was Combi SCC, with the independent variables RT-10, DSCC, ECM, and HC SCC. Covariates were sample collection times (AM or PM), milk protein, fat, lactose, SNF, and MUN. Using backwards elimination, covariates that were significant contributors to the regression were milk protein, SNF, and MUN for meters RT-10, DSCC, PC, ECM, and HC.

For individual quarter and BTM, tests for goodness of fit and the coefficient of determination (**R^2**), mean bias (**MB** %), error due to mean absolute error (**MAE**), and partitioning of the mean square predictive error (**MSPE**) due to central tendency (**CT** %), unequal variation (**UEV** %), error due to random variation (**RV** %), error due to slope ≠ 1 (%) were evaluated according to Bibby and Toutenburg (1977).

Determination of diagnostic sensitivity (**SE**), specificity (**SP**), prevalence, accuracy, likelihood ratios positive (**LR+**) and negative (**LR-**), and predictive values positive (**PPV**) and negative (**NPV**) for each meter for both individual quarter and bulk-tank milk samples was completed by using MedCalc's diagnostic test evaluation calculator (MedCalc v. 20.027; MedCalc Software Ltd., Ostend, Belgium). Disease prevalence was calculated by the total number of positives divided by the total number of milk samples. Cell counts that valued ≥ 200,000 cells/mL for the Combi were defined as IMI as this classified infection status (NMC, 2001), and each meter was evaluated on its ability to identify IMI correctly. The SCC that valued ≥ 200,000 cells/mL on DSCC and RT-10 meters, of the category ≥ Trace on the CMT, of the category ≥ 250,000 cells/mL on the PC meter, of ≥ 5.0 m/S on the ECM meter, of ≤ 6.6 pH on the HC meter, of ≥ 37ºC on the Temp meter, and of ≥ 200,000 cells/mL for PSCC were considered a positive test. The SCC values used for meters DSCC, RT10, PC, and CMT were defined using the manufacturer's directions, whereas the values used for meters ECM, HC, and IR5 were defined using previously published research on IMI incidence.

RESULTS AND DISCUSSION

Detection of IMI is needed to improve milk production, cow health, and ultimately, reduce the use of antibiotics at dry-off. To decrease the risk of IMI in herds, dairy producers can employ different meters that assess the amount of SCC in individual cow quarter and bulk-tank milk. This study compares the performance of several common SCC meters to evaluate SCC at dry-off.

Enrolled cows (**Table 2**) followed similar trends as their respective dairy in **Table 1**. The 4 enrolled dairies used different herd management practices, with different herd sizes, milk yields, and milk components. For example, due to the low price of pregnant heifers, Dairy 4 purchased pregnant heifers to milk, and milked all cows as long as possible. Dairy 4 also did not subscribe to monthly milk testing so there were no milk yield or milk composition data available. They generally did not treat at dry-off and they had the highest average DIM, DIM at dry-off, and relatively lowest average parity. Dairies 2 and 3 also did not prophylactically treat cows at dry-off and had higher SCC. Dairy 1 treated all cows at dry-off, had the lowest average quarter SCC, lowest DIM at dry-off, lowest SCC, and relatively high milk production because reproductive management was a priority at this dairy.

Performance of meters on individual quarter milk samples

The RT-10 meter best predicted SCC in individual cow quarters compared to the other meters (**Table 3**). The regression of predicted RT-10 on observed Combi quarter SCC had a slope closest to 1, i.e., predicted equaling observed SCC (**Figure 1**), the highest coefficient of determination (**Table 3**), and one of the lowest MSPE. The MSPE was split into 3 different categories of error; bias of prediction (difference contained within the model's predicted values versus the observed), slope not equal to 1 (error associated with the slope not being equal to 1) and random variation (variation contained within the observed data. Most of the MSPE was due to random variation in the data. The residuals regressed on observed Combi quarter SCC had a low but consistent bias (**Figure 1**). The under prediction of SCC by the RT-10 had less of an impact when SCC are below 200,000 cells/mL. Therefore the RT-10 was the best meter for measuring SCC below 200,000 cells/mL but, as SCC increased, so did the error associated with the measurement.

The RT-10 and DSCC had similar SE, and compared to other SCC tests, had a greater percent ability to identify a quarter with an infection (SCC > 200,000 cells/mL), similar SP, a greater percent ability to identify a quarter without infection, a greater percent ability to differentiate between SCC above and below 200,000 cells/mL, and accuracy. Previous studies used the same level of 200,000 cells/mL to determine SE and SP for the RT-10 and DSCC. Compared to the previous study by Albrechtesen *et al*. (2011), the RT-10 and DSCC showed a greater ability to differentiate infection status in individual quarters and estimated an approximately 10 % higher SE and 4 % higher SP.

The DSCC meter and RT-10 meter used the same methods and cartridges to measure SCC. So, it would be expected that they would perform similarly. However, while the DSCC meter performed well in diagnostic measures, SE and SP, the regression of predicted DSCC on observed Combi quarter SCC had a larger intercept, much lower coefficient of determination, larger MSPE with a higher error due the slope not being equal to 1 (**Table 3**). The DSCC under predicted SCC to a greater extent especially above SCC of 100,000 cells/mL and the underestimation increased as SCC increased with a bias in the residuals (Figure 2). However, if milk lactose content was added to the regression of predicted DSCC on observed Combi SCC, the ability of the DSCC to predict SCC improved ($R^2 = 0.44$). This implies that the difference between the performance of the two meters is due to a better camera in the iPhone as the DSCC may lose some resolution of SCC with more lactose in the milk.

The only other meter that was able to more accurately predict SCC was the PC meter. This meter is a colorimetric meter only able to predict a range of SCC within specified SCC categories and so interpretation of results is not as precise as other meters since it relies on the ability of the user to match a color chart that designates a range of SCC values. The PC meter classifies SCC within these ordinal categories: ≤ 100,000, 250,000, 500,000, 750,000, 1,500,000, and ≥ 3,000,000 cells/mL. The predicted SCC by the PC meter vs observed Combi SCC regression slope was further from unity than the RT-10 and DSCC, had a higher coefficient of determination and lower MSPE than the DSCC and more of the proportion of error was due to random variation in the data (**Table 3**). The PC meter had a lower SE, SP and accuracy compared to the RT-10 and DSCC (**Table 4**). The previous studies also used an SCC of 250,000 cells/mL to determine SE, SP and estimated an 8 % higher SE and 11 % lower SP than in this study, possibly due to colorimetric SCC ranges being subjective. If a dairy were to use any of these meters to determine whether to treat a cow for mastitis, they should consider which meter will predict SCC well within their desired SCC cutoff to treat to decide which cows should be treated for their dry-cow treatment program.

Because many dairies use PSCC at the most recent DHIA test to identify cows to treat at dry-off, one more method of predicting SCC was evaluated. Dairy 1 PSCC were used to predict combi SCC because they had the lowest herd SCC and they treated all cows at dry-off. Since the Combi SCC was only measured on individual quarters, SCC from each quarter were averaged to estimate Combi SCC and compare to PSCC. The regression of predicted SCC using PSCC on observed SCC from Combi had a low coefficient of determination and low MSPE but most of the error was due to a poor model fit (high error due to slope not equaling 1 and bias in the predictions) between predicted SCC from PSCC and observed SCC from an average of Combi (**Table 3**). The predictability of Combi SCC using PSCC decreased as Combi SCC increased (**Figure 3**), indicated by a steady increase in the residuals as Combi SCC increased. This led to lower predictability and bias of prediction, and is not unexpected since infections that result in a high SCC for a short period of time may be harder to detect in-between monthly test days as these infections are associated with environmental pathogens (de Haas, *et al*. 2002).

The SE, SP and accuracy of PSCC were also low compared to the other meters (**Table 4**). Compared to the previous studies, the current study showed a 19 % lower SE and a 24 % higher SP. This analysis suggests that PSCC was a poor predictors of average Combi SCC. However, using averaged individual quarter SCC from Combi may not be representative of SCC collected as a complete sample from the udder particularly from udders with infected quarter. Typical monthly DHIA milk test samples are blended and it is assumed that each quarter contributed an equal amount of milk to the blended sample. However, if one of the quarters has a high SCC, the milk sample can become thicker and will not flow and contribute the same amount of milk to the blended sample, therefore the blended sample could under-represent SCC. In this study, quarter infection status was different among the 40 enrolled cows from Dairy 1 with 30 % of cows with an infection in all quarters (SCC > 200,000), 19 % in 3 quarters, 23 % in 2 quarters, and 26 % in one quarter. If the thickness of the high SCC milk sample affected the SCC in the blended sample, the blended sample SCC will never truly represent the SCC of the cow and will underpredict the level of mammary infection. This is particularly a problem when only 1 or 2 quarters are infected, which was almost 50% of the cows at Dairy 1.

Performance of meters on BTM samples

The meters RT-10, DSCC, PC, ECM, and HC were chosen for BTM analysis as these meters did not need to be used on fresh milk samples or in the presence of the cow. The DSCC meter and RT-10 meters best predicted BTM SCC the highest coefficients of determination, lowest MSPE and highest proportion of MSPE due to random variation (**Table 5**). Both the DSCC and RT-10 had the best SE, SP and accuracy (**Table 6**). Compared to the previous studies SE and SP (**Table 3**), RT-10 and DSCC estimated an approximate 6 % higher SE and 24 % lower SP. Because BTM samples are composites samples from the milking herd, the meters might have had similar issues with high SCC quarter samples being underrepresented in a blended sample. Samples were also not measured until 12 - 24 h after collection from the bulk tank and so the longer length of time may have affected the ability of the meters to estimate SCC. Settling of solids in the bulk tank may have increased solids in the sample which could interfere with meter function.

Meter measurement performance comparison

In relation to the prediction statistics and diagnostic test measures of performance, the RT-10, PC and DSCC meters were the best in their ability to predict SCC. The DSCC meter is large and heavy, costs $1,960.22 more than the RT-10, and analyzes samples 10 s faster than the RT-10 (**Table 7**). However, the DSCC meter performed poorly at the prediction of individual quarter SCC compared to the RT-10. While the RT-10 performed the best at the prediction of individual quarter SCC, the adapter is only fitted for the iPhone 5s, and has not been updated for newer versions of the iPhone nor has it been made available for the android phone. The RT-10 uses a Dairy Health Management application for record storage, which makes it easy to use compared to the DSCC. The management application allows users to collect and store SCC and cow health data, input cow numbers, and create and export excel spreadsheets with health and SCC data. For SCC analysis, the DSCC and the RT-10 can both be used cow-side or in a laboratory setting. The cartridges for both the DSCC and RT-10 cost about $2.10 per quarter, with producers able to test one cow for about $8.40 compared to $1.00 per quarter for DHIA samples using the combi. Since dry-cow treatments generally cost $5.00 per quarter, if a dairy is willing to selectively treat individual quarters within cows, using the meter could save $11.60 per cow not treated.

The PC test strip is available in different versions, one being a 45 min version and the other being a 5 min version, with both being able to be used on-farm. The 5 min version was used in this study because 45 min was too long to wait for results. There could be a possibility of missed IMI treatment in the dry-off period as the threshold for the detection of SCC is < 150,000 cells/mL for this meter, and it did not perform as well as SCC increased. The test kit includes 20 pouches with two test strips in each pouch, 10 pipettes, activator solution, and instructions on how to perform the test, coming out to about $1.01 per sample, with all 4 quarters tested for about $4.04. While this test can be used cow-side, it is more feasible to perform this test on a lab benchtop.

The meters CMT, ECM, HC, and IR5 all underperformed at the analysis of individual quarter and BTM samples. While the CMT can be used as an indicator for IMI, this meter is unable to accurately quantify SCC as this is a qualitative, categorical test. There is also an element of human error with the CMT, as not every person will read the results in the same way. The meters ECM and HC could measure samples relatively fast; however, the measurement time would not be consistent across samples, and the meters would often turn off in the middle of sample analysis. The IR5 was easy to use cow side but could not adequately predict quarter SCC.

CONCLUSION

Meters that can accurately measure individual mammary quarter SCC have the potential to reduce antibiotic treatment of non-infected quarters and to decrease treatment costs associated with dry-cow therapies. The RT-10 meter best predicted individual quarter SCC ≤ 200,000 cells/mL followed by the DSCC which also best predicted BTM SCC ≤ 100,000 cells/mL. It may be beneficial for dairies to use these cow-side meters instead of previous SCC. As PSCC increases, predictability and the bias of prediction to underpredict SCC increases. There may be differences due to the average of individual quarter samples not being equivalent to the SCC of composite samples, as there may be one high quarter being diluted by quarters with lower SCC in the composite sample.

TABLES AND FIGURES

Table 1. Differences in herd management practices and milk production parameters of dairies enrolled

Dairy Sampled[1]	Total cows milking	Average DIM	DIM at dry-off	Average Parity	305ME kg	SCC x 1000	Milk Fat %	Milk Protein %	Times milked
1	1,471	197	335	1.9	14,300	281	5.72	3.45	2
2	4,000	190	340	2.4	14,700	551	5.32	3.57	3
3	2,500	143	340	2.3	11,800	546	4.13	3.82	2
4[2]	2,732	318	430	2.1					2

[1]Dairies 1, 2, and 4, used dairy herd management software DairyComp 305 (Tulare, CA), and Dairy 3 used dairy herd management software DHI-Plus (Provo, UT). Data for dairies 1, 2, and 3 were obtained from monthly DHIA herd testing records.
[2]Dairy 4 does not employ monthly milk testing

Table 2. Dry-cow management practices of cows enrolled at each dairy

Dairy Sampled[1]	Pen Type[2]	Season Sampled	Average DSLT[3]	Average Parity	Average DIM at dry-off	Average DCC[4]	Average SCC x 1000	Times sampled	Quarters sampled
1	DL	Fall	15	1.9	287	224	287	7:00 AM	160
2	FS	Winter	20	2	233	233	551	8:00 AM	176
3	DL	Summer	17	1.8	337	236	546	7:30 AM	162
4	FS	Summer		1.7	336	203	337	7:00 AM	160

[1]Dairies 1, 2, and 4, used dairy herd management software DairyComp 305 (Tulare, CA), and Dairy 3 used dairy herd management software DHI-Plus (Provo, UT)
[2]Pen types where cow quarter milk samples were taken are dry lot (DL) or free stall (FS) using composted manure
[3]Average days since last test (DSLT)
[4]Average days carried calf (DCC)

Table 3. Regression of meter SCC values on observed SCC (Combi) from individual cow quarters

Descriptive Statistics[1]	DSCC[2] (n = 658)	RT-10 (n = 658)	PC[3] (n = 658)	CMT (n = 658)	ECM (n =658)	HC (n = 658)	IR5 (n = 658)	PSCC[4] (n = 40)
Observed mean, SCC x 1000	432	432	432	432	432	432	432	282
Predicted mean[5], SCC x 1000	432	432	250	400	573	435	432	286
Observed SD, SCC x 1000	859	859	859	859	859	859	859	316
Predicted SD, SCC x 1000	543	797	641	87	3203	38	0.240	281
Linear Regression[6]								
Intercept	178	-28.8	-79.3	570	-749	0	431	186
Slope	0.60	1.1	1.5	-56	210	64	0.010	0.49
Mean Square Error (MSE)	62806	102106	66829	190505	312121	198002	197907	82463
Coefficient of Determination (R^2)	0.40	0.86	0.56	0.010	0.080	0.0040	0.0	0.19
Mean Bias, %	-0.030	0.030	0.15	-0.11	-33	-0.48	0.030	0.35
Mean Absolute Error (MAE)	251	111	259	436	559	445	445	182
Mean Square Predicted Error (MSPE)	442154	101685	326367	729200	10986013	733223	736542	78341
Partition of MSPE, %								
Error due to bias of prediction, %	3	1	0	3	0	0	1	0
Error due to slope ≠ 1, %	23	4	14	82	50	92	99	61
Error due to random variation, %	77	96	86	18	50	8	0	39

[1]Fourier Transform Spectrometer 600 Combi System (**Combi**; Bentley Instruments, MN) values were regressed on SCC predictions by the other meters using PROC GLM in SAS (SAS v. 9.4; Statistical Analysis System, SAS Inst. Inc., Cary, NC)

[2]DeLaval Cell Counter (DSCC; DeLaval Graiguecullen, Carlow), RT-10 iPhone adapter (RT-10; Dairy Quality Inc., Newmarket, Canada), PortaCheck Quick Test (PC; PortaCheck, NJ), California Mastitis Test (CMT; Dairy Research Product, Inc., IN), electrical conductivity meter (ECM; OHAUS, NJ), pH meter (HC; Hanna Instruments, RI), dual laser infrared temperature thermometer (IR5; Klein Tools, IL

[3]Meters PC and CMT are discontinuous tests, as they only predict ranges of SCC. The largest range number that corresponded to the color change of the test were chosen as the values of SCC

[4]Previous test day SCC (PSCC) regression was completed using data from Dairy 1 as this dairy recorded PSCC, and individual quarter samples for the Combi were averaged by test month for the regression analysis.

[5]Meters PC and CMT are discontinuous tests, the predicted means were chosen using the average SCC range values of the meters

[6]Goodness of fit were evaluated according to Bibby and Toutenburg (1977)

Table 4. Diagnostic tests evaluations of meter measurements of SCC for individual cow quarters

Diagnostic Test[1]	DSCC[2,3]	RT-10	PC	CMT	ECM	HC	IR5	PSCC[3]
SE, %	92.54	91.53	73.95	97.63	86.44	26.78	0	67.19
95 % CI	(88.93 – 95.27)	(84.74 – 94.44)	(69.07 – 78.73)	(95.17 – 99.04)	(82.00 – 90.13)	(21.81 – 32.22)	(0.00 – 1.24)	(54.31 – 78.41)
SP, %	90.08	90.36	89.70	16.25	33.88	69.15	100	74.49
95 % CI	(86.53 – 92.96)	(86.85 – 93.19)	(85.70 – 92.89)	(12.61 – 20.46)	(29.03 – 39.01)	(64.11 – 73.86)	(0.00 – 0.00)	(64.69 – 82.76)
Prevalence[4], %	50.30	50.15	58.97	44.83	81.30	61.85	44.83	54.94
Accuracy, %	91.19	90.88	81.16	52.74	57.45	50.15	55.17	71.60
LR +, %	9.33	9.49	7.18	1.17	1.31	0.87	0	2.63
LR -, %	0.08	0.09	0.29	0.15	0.4	1.06	1	0.44
PPV, %	88.35	88.52	89.49	48.65	51.52	41.36	0	63.24
NPV, %	93.7	92.92	74.38	89.39	75.46	53.76	55.17	77.66
Previously Published								
SE, %	82.00	82.00	81.00	57.40	43.50	83.00	95.60	86.00
SP, %	86.00	86.00	78.00	72.30	92.90	29.00	93.60	50.00
References	Albrechtesen *et al*. (2011)	Albrechtesen *et al*. (2011)	Kandeel *et al*. (2019 A)	Sargeant *et al*. (2001)	Norberg *et al*. (2004)	Kandeel *et al*. (2019 B)	Polat *et al*. (2010)	Kristula *et al*. (1992)

[1]Sensitivity (Se), specificity (Sp), lower and upper bounds of the 95% CI, accuracy, positive (LR +) and negative (LR -) likelihood ratios, and positive (PPV) and negative (NPV) predictive values were calculated by using the diagnostic test evaluation calculator by MedCalc (MedCalc v. 20.027; MedCalc Software Ltd., Ostend, Belgium)

[2]DeLaval Cell Counter (DSCC; DeLaval Graiguecullen, Carlow), RT-10 iPhone adapter (RT-10; Dairy Quality Inc., Newmarket, Canada), PortaCheck Quick Test (PC; PortaCheck, NJ), California Mastitis Test (CMT; Dairy Research Product, Inc., IN), electrical conductivity meter (ECM; OHAUS, NJ), pH meter (HC; Hanna Instruments, RI), dual laser infrared temperature thermometer (IR5; Klein Tools, IL), and previous SCC (PSCC). This diagnostic for PSCC was completed using only data from Dairy 1 as this dairy recorded PSCC.

[3]DSCC, RT-10, CMT, ECM, HC, IR5, and PSCC used SCC 200,000 cells/mL and above as positive infection, PC used 250,000 cells/mL and above as as a positive infection

[4]Prevalence was calculated by the total number of positives divided by the total number of milk samples

Table 5. Statistical summary of results from regression of predicted on observed bulk-tank SCC values

Descriptive Statistics[1]	DSCC[2] (n = 100)	RT-10 (n = 100)	PC[3] (n = 100)	ECM (n = 100)	HC (n = 100)
Observed mean, SCC x 1000	182	182	182	182	182
Predicted mean, SCC x 1000	182	182	100	182	182
Observed SD, SCC x 1000	70	70	70	70	70
Predicted SD, SCC x 1000	40	36	19	12	10
Linear Regression[5]					
Intercept	92.87	109.44	137.48	210.67	-118.12
Slope	0.40	0.31	0.26	-7.05	41.51
Mean Square Error (MSE)	1834	1983	2970	3408	3406
Coefficient of Determination (R^2)	0.33	0.26	0.08	0.03	0.02
Mean Bias, %	-4.00	-2.5	1.8	-6.8	1.22
Mean Absolute Error (MAE)	43	45	54	58	58
Mean Square Predicted Error (MSPE)	3200	3546	4426	4663	4700
Partition of MSPE, %					
Error due to bias of prediction, %	2	6	3	3	1
Error due to slope ≠ 1, %	27	32	57	72	75
Error due to random variation, %	73	68	43	28	25

[1]DeLaval Cell Counter (DSCC; DeLaval Graiguecullen, Carlow), RT-10 iPhone adapter (RT-10; Dairy Quality Inc., Newmarket, Canada), PortaCheck Quick Test (PC; PortaCheck, NJ), California Mastitis Test (CMT; Dairy Research Product, Inc., IN), electrical conductivity meter (ECM; OHAUS, NJ), pH meter (HC; Hanna Instruments, RI), dual laser infrared temperature thermometer (IR5; Klein Tools, IL

[2]Combi values were regressed on SCC predictions by the other meters using PROC GLM in SAS (SAS v. 9.4; Statistical Analysis System, SAS Inst. Inc., Cary, NC)

[3]Meter PC is a discontinuous test, as it only predicts ranges of SCC. The largest range number that corresponded to the color change of the test were chosen as the value of SCC

[4]Meter PC is a discontinuous test, the predicted mean was chosen using the average SCC range values of the meter

[5]Goodness of fit were evaluated according to Bibby and Toutenburg (1977)

Table 6. Summary of diagnostic test evaluations on meters using bulk-tank milk samples

Diagnostic Test[1]	DSCC[2]	RT-10	PC	ECM	HC
SE, %	85.71	88.10	59.52	21.43	0
95 % CI	(71.46 – 94.57)	(74.37 – 96.02)	(43.28 – 74.37)	(10.30 – 36.81)	(0.00 – 100)
SP, %	70.69	62.07	58.62	62.07	100
95 % CI	(57.27 – 81.91)	(48.37 – 74.49)	(44.93 – 71.40)	(48.37 – 74.49)	(93.84 – 100)
Prevalence[3], %	59.00	64.00	66.00	64.00	42.00
Accuracy, %	77.00	73.00	59.00	45.00	58.00
LR +, %	2.92	2.32	1.44	0.56	0
LR -, %	0.2	0.19	0.69	1.27	1
PPV, %	67.92	62.71	51.02	29.03	0
NPV, %	87.23	87.80	66.67	52.17	58.00

[1]Sensitivity (SE), specificity (SP), lower and upper bounds of the 95% CI, accuracy, positive (LR +) and negative (LR -) likelihood ratios, and positive (PPV) and negative (NPV) predictive values were calculated by using the diagnostic test evaluation calculator by MedCalc (MedCalc v. 20.027; MedCalc Software Ltd., Ostend, Belgium)

[2]DeLaval Cell Counter (DSCC; DeLaval Graiguecullen, Carlow), RT-10 iPhone adapter (RT-10; Dairy Quality Inc., Newmarket, Canada), PortaCheck Quick Test (PC; PortaCheck, NJ), electrical conductivity meter (ECM; OHAUS, NJ), pH meter (HC; Hanna Instruments, RI)

[3]Prevalence was calculated by the total number of positives divided by the total number of milk samples

Table 7. Comparison of ease of use and costs of meters

	Combi[1]	DSCC	RT-10	PC	CMT	ECM	HC	IR5
Meter cost[2], $	350000	3497	1533	40	15	61	150	60
Cost per sample[3], $	1.00	2.10	2.10	1.01	0.04	0.00	0.00	0.00
Measurement time[4]	7 s	50 s	60 s	5 m	10 s	10 s	30 s	2 s
Volume of milk sampled	6 mL	60 µl	60 µl	80 µl	3 mL	10 mL	10 mL	0
Sample environment	Lab	Both	Both	Lab	Cow-side	Lab	Lab	Cow-side

[1]A FTS 600 (Combi; Bently Instruments, MN), RT-10 iPhone adapter (RT-10; Dairy Quality Inc., Newmarket, Canada), DeLaval Cell Counter (DSCC; DeLaval Graiguecullen, Carlow), PortaCheck Quick Test (PC; PortaCheck, NJ), California Mastitis Test (CMT; Dairy Research Product, Inc., IN), electrical conductivity meter (ECM; OHAUS, NJ), pH meter (HC; Hanna Instruments, RI), dual laser infrared temperature thermometer (IR5; Klein Tools, IL)

[2]Meter cost and volume of milk sampled was determined by the set cost and directions for use from the manufacture

[3]Cost per sample was calculated by dividing Meter Cost ($) by the number of samples included in the kits

[4]Measurement time was determined when the meter’s reading became consistent

Figure 1. Plot of Predicted SCC using the RT-10 vs. Observed SCC from Combi based on individual milk samples

Meter Data (•), Residuals (▲), Regression (▬), Reference Regression line where slope = 1, intercept = 0 (▬), Regression of Residuals on Observed (– – – –) $P < 0.01$

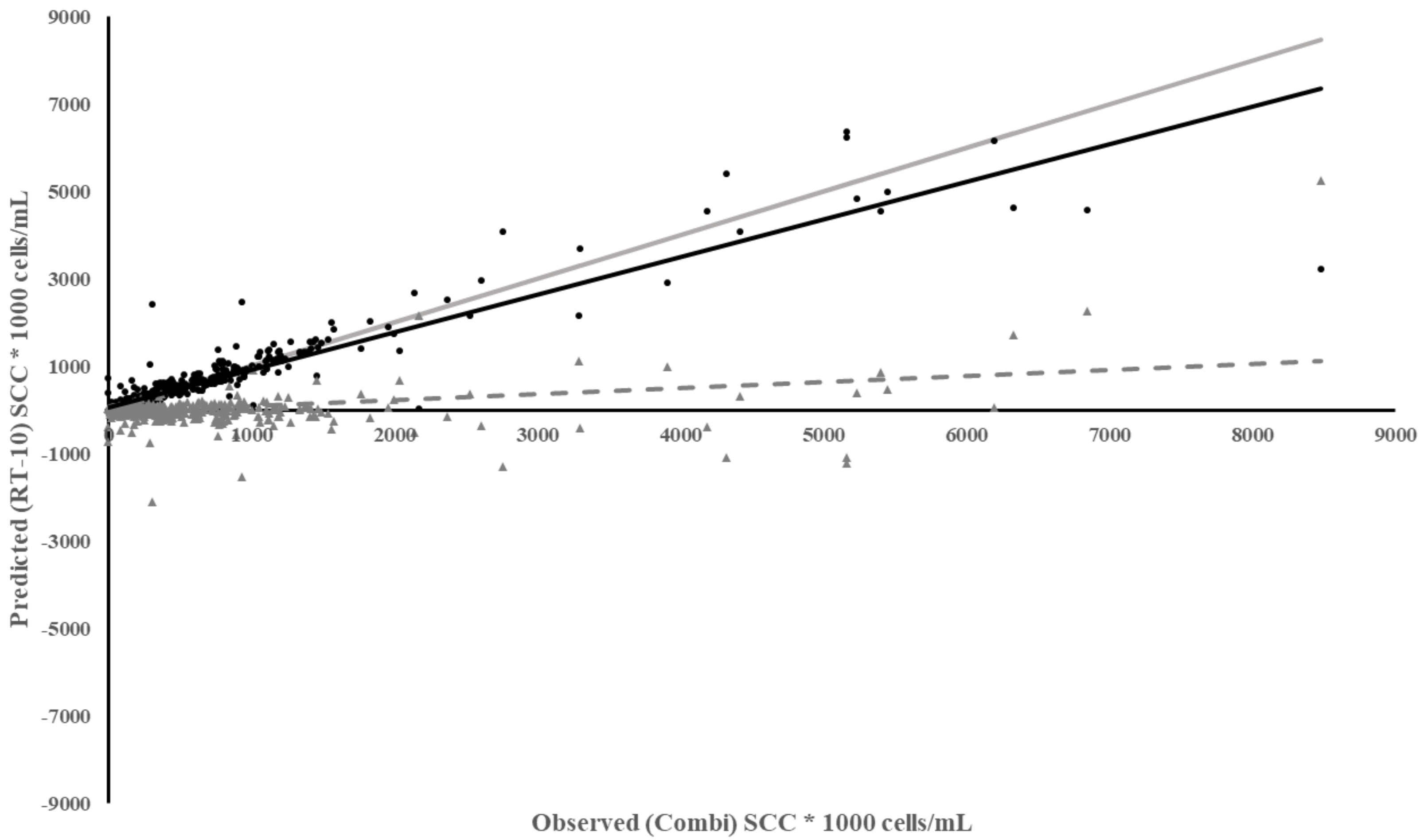


Figure 2. Plot of Predicted SCC using the DSCC vs. Observed SCC from the Combi based on individual milk samples

Meter Data (•), Residuals (▲), Regression (▬), Reference Regression line where slope = 1, intercept = 0 (▬), Regression of Residuals on Observed (– – – –) $P < 0.01$

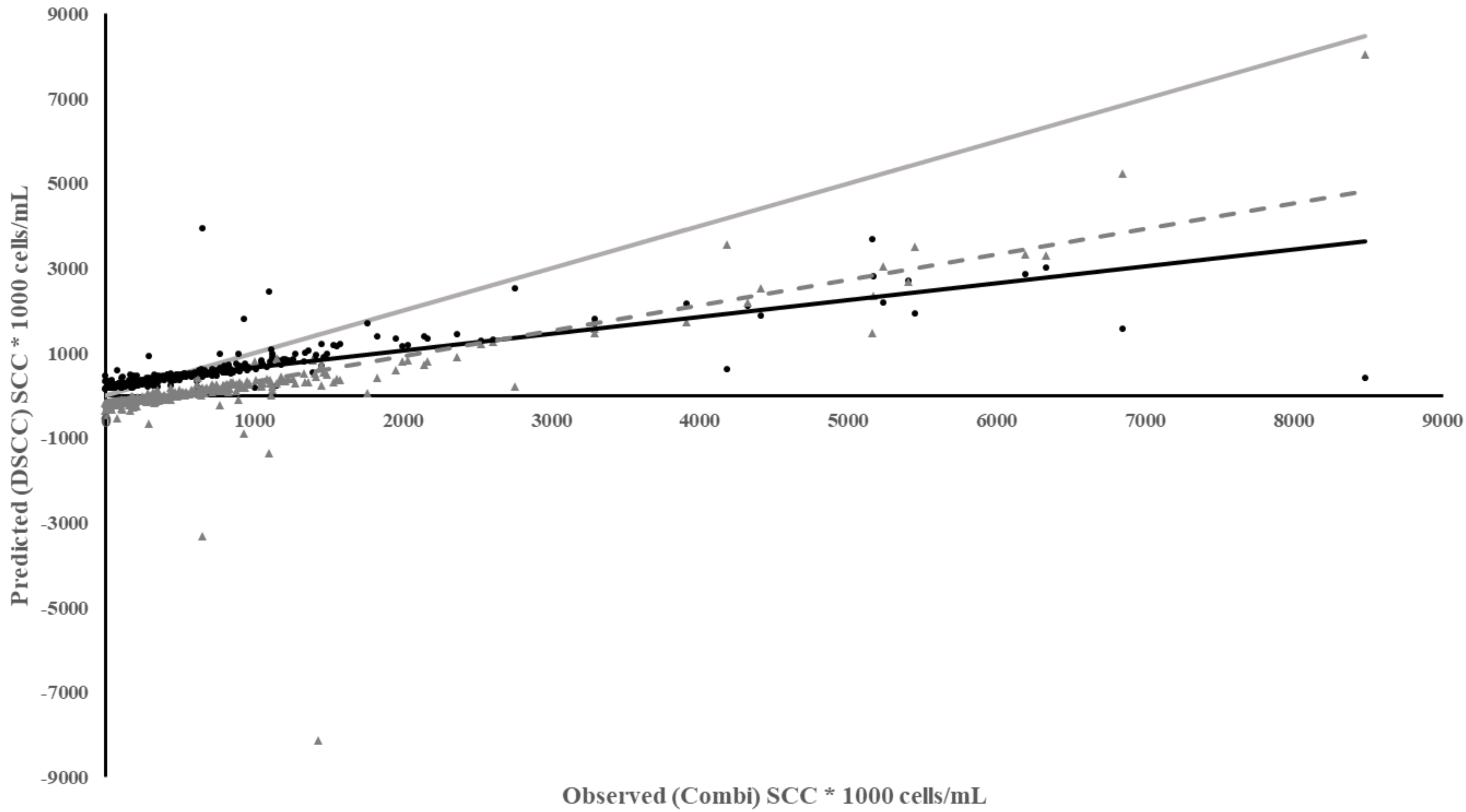
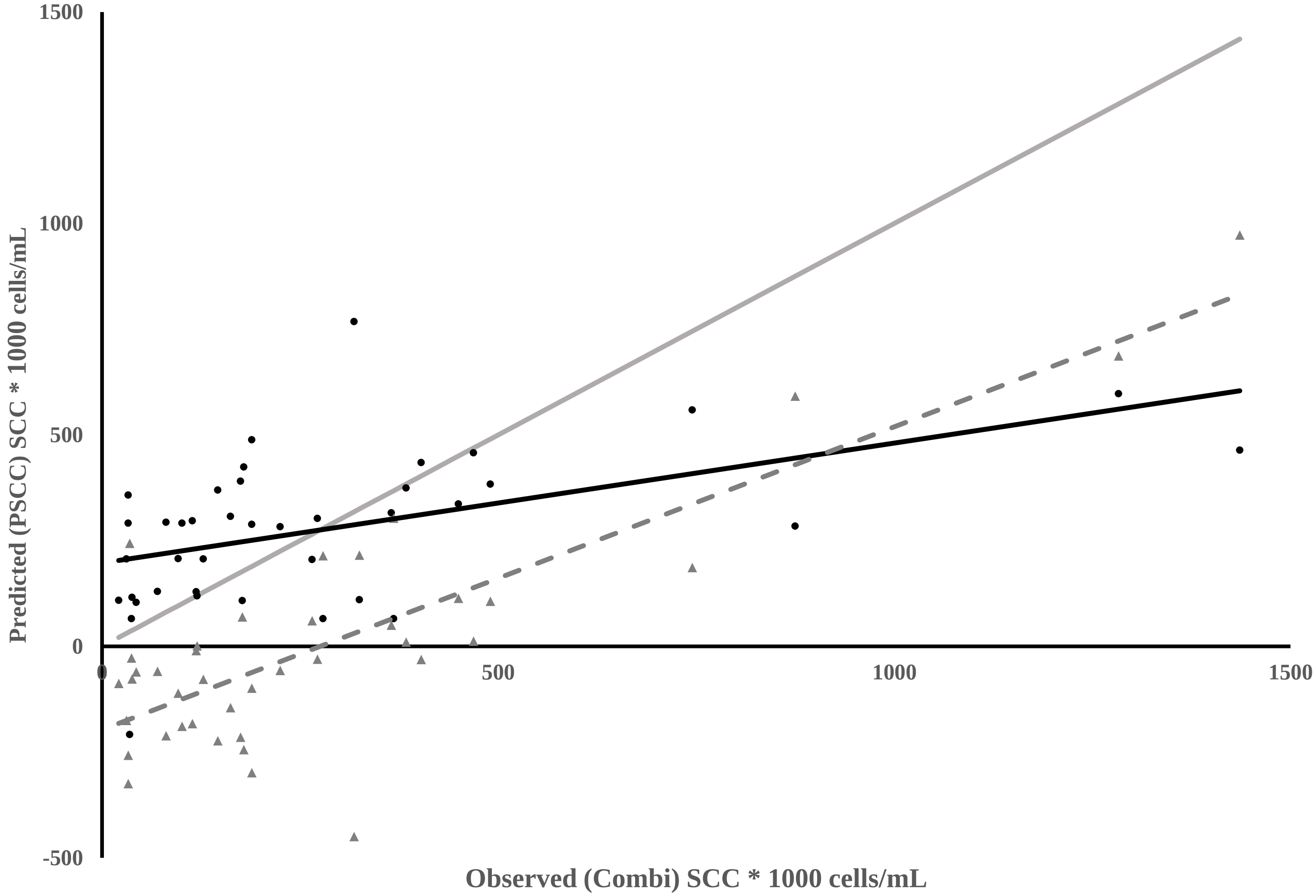


Figure 3. Plot of Predicted SCC using previous test day SCC (PSCC) vs. Observed SCC from Combi for individual milk samples. Combi SCC from each quarter were averaged per cow to compare to each test day in Dairy 1.

Meter Data (•), Residuals (▲), Regression (▬), Reference Regression line where slope = 1, intercept = 0 (▬), Regression of Residuals on Observed SCC using Combi (– – – –) $P < 0.01$



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