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Instructions for use

Near-infrared spectroscopic sensing system for online milk quality assessment in a milking robot

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#### **Abstract**

A near-infrared (NIR) spectroscopic sensing system was constructed on an experimental basis. This system enabled NIR spectra of raw milk to be obtained in an automatic milking system (milking robot system) over a wavelength range of 600 nm to 1050 nm. Calibration models for determining three major milk constituents (fat, protein and lactose), somatic cell count (SCC) and milk urea nitrogen (MUN) of unhomogenized milk were developed, and the precision and accuracy of the models were validated. The coefficient of determination (r<sup>2</sup>) and standard error of prediction (SEP) of the validation set for fat were 0.95 and 0.25%, respectively. The values of r<sup>2</sup> and SEP for lactose were 0.83 and 0.26%, those for protein were 0.72 and 0.15%, those for SCC were 0.68 and 0.28 log SCC/mL, and those for MUN were 0.53 and 1.50 mg/dL, respectively. These results indicate that the NIR spectroscopic system can be used to assess milk quality in real time in an automatic milking system. The system can provide dairy farmers with information on milk quality and physiological condition of an individual cow and, therefore, give them feedback control for optimizing dairy farm management. By using the system, dairy farmers will be able to produce high-quality milk and precision dairy farming will be realized.

Keywords: Quality control, Spectroscopy, Management system, Diagnosis, Monitoring

## 1. Introduction

Dairy farming is labor-intensive and involves many tasks such as feeding, milking, livestock management, feed crop production and manure treatment. Large-scale dairy farmers manage their livestock in groups, a system known as herd management. However, monitoring the milk quality of each cow and managing each cow according to milk quality and physiological condition, a system known as individual cow management, is also essential for optimum production of high-quality milk. Milk quality is greatly affected by the physiological condition of cows. Therefore, assessment of milk quality of each cow is necessary for individual cow management (Svennersten-Sjaunja *et al.*, 1997). Recently, there has been a need for an automatic on-line method that will enable dairy farmers to assess milk quality of an individual cow during milking.

Near-infrared spectroscopy (NIRS) is a nondestructive method for quality evaluation. Advantages of NIRS include, but are not limited to, the fact that on-line measurement can be performed rapidly, pollution-free and without pre-treatment. NIRS has been widely used in quality evaluation of foods and agricultural commodities including rice (Kawamura *et al.*, 2002; Kawamura *et al.*, 2003; Natsuga *et al.*, 2006), wheat (Natsuga *et al.*, 2001) and satsuma mandarins (Miyamoto *et al.*, 1998). NIRS has also been used to assess milk quality (Sato *et al.*, 1987; Tsenkova *et al.*, 1999; Tsenkova *et al.*, 2001; Natsuga *et al.*, 2002), but it has been difficult to apply NIRS to real-time on-line monitoring of milk quality of an individual cow during milking.

An experimental, on-line, near-infrared (NIR) spectroscopic sensing system has been constructed to assess milk quality. Kawamura *et al.* (2007) reported that the NIR spectroscopic sensing system can be used for real-time assessment of milk quality during milking with sufficient precision and accuracy. Based on these results, the NIR spectroscopic sensing system was installed in an automatic milking system (a milking robot system). A milking robot is a system that performs voluntary milking for cows (i.e., each cow deciding milking time and milking interval) at any time during the day without the requirement of human labor. Milking robot systems have been available commercially since 1990s, and the use of these systems has improved herd management. However, in the milking robot system, a sensing system to examine milk quality and physiological condition of the individual cow is needed.

In this study, the precision and accuracy of the NIR spectroscopic sensing system for assessing milk quality during milking by a milking robot system were validated.

#### 2. Materials and methods

### 2.1 Near-infrared spectroscopic sensing system

An experimental, on-line, NIR spectroscopic sensing system for assessing milk quality of an individual cow during milking was constructed. The system consisted of an NIR spectroscopic instrument, a milk flow meter, a milk sampler and a laptop computer (Fig. 1). The system was installed in a milking robot system (Astronaut, Lely Industries NV, Maasland, Holland) with the milk being bypassed from the teat cups to a milk jar (Fig. 2). Raw milk from the milking robot continuously flowed into the milk chamber of the spectrum sensor and flowed out through an outlet pipe for surplus milk to the milk flow meter. The volume of milk sample in the chamber was about 230 mL. The optical axes of a halogen lamp and an optical fiber were set at right angles to each other at the same levels (Fig. 3). The spectrum sensor acquired spectra of diffusion transmittance (interactance) through the milk. The diffusion transmittance spectra were recorded in the wavelength range of 600 to 1050 nm at 1-nm intervals every 10 seconds during milking (Table 1). Six continual spectra were averaged to obtain a spectrum for one minute.

# 2.2 Cows and milk samples

Seventeen Holstein cows in the stage of early lactation to late lactation were used in the experiment (Table 2). The experiment was conducted all day and night on October 20th and 21st, 2003. Milking was automatically started whenever a cow walked into the milking robot. Milk samples were collected from the milk sampler every minute during

milking. The experiment was conducted to cover variations in milk spectra caused by cow individuality, calving times, lactation stage, milking time and environmental temperature.

## 2.3 Reference analyses

Three major milk constituents (fat, protein and lactose), somatic cell count (SCC) and milk urea nitrogen (MUN) of raw milk were measured as indices of milk quality in this study. The milk constituents and MUN were determined using a Milkoscan 4000 (Foss Electric, Hillerod, Denmark), and SCC was determined using a Fossomatic 5000 (Foss Electric). The total number of samples used for reference analyses was 216 for milk constituents and SCC and 210 for MUN. SCC was converted into common logarithms.

### 2.4 Chemometric analyses

Chemometric analyses were carried out to develop calibration models for each milk quality item and to validate the precision and accuracy of the models. Spectral data analyses software (The Unscrambler ver. 9.6, Camo AS, Trondheim, Norway) was used for the analyses. The reference samples were randomly divided into two sample sets: a calibration subset containing two-thirds of all samples and a validation subset containing the remaining samples (one-third). The statistical method of partial least squares (PLS) was used to develop calibration models from the transmittance spectra and reference data. Pretreatment of the spectra such as smoothing or second derivatives was not performed.

#### 3. Results and discussion

### 3.1 NIR spectra

Figure 4 shows an example of original NIR spectra of raw milk from cow number 304 during milking on October 21, 2003. Optical density of the transmittance spectra exceeds 100% in the wavelength range of 600 to 1050 except 950 to 1020 nm. A reference spectrum was measured when there was no milk in the milk chamber, i.e., when there was only air in the milk chamber. Transmittance spectra of raw milk were measured when the chamber was full with milk. The optical density exceeded 100% because of scattering of light by fat globules in raw milk. The deep valley of the spectra in the wavelength range of 970 to 990 nm in Fig. 4 indicates second-overtone absorption by water molecules. The two valleys in the spectra around 740 nm and 840 nm indicate overtone absorption by C-H strings and C-C strings that are associated with fat (triacylglycerol).

## 3.2 Precision and accuracy of calibration models

The validation statistics of the NIR sensing system for determination of milk quality are summarized in Table 3. Correlations between reference and NIRS-predicted values of fat, lactose, protein, SCC and MUN are shown in Figures 5 to 9, respectively.

The three major milk constituents are the main factors determining the quality of milk. The milk constituents are affected by the physiological condition of each cow and feedstuff. Monitoring of milk constituents during milking every day can be used for management of each cow and her feed. The coefficient of determination (r²), standard error of prediction (SEP) and bias of the validation set for fat were 0.95, 0.25% and -0.06%, respectively. The values of r², SEP and bias for lactose were 0.83, 0.26% and 0.00%, respectively, and the values of r², SEP and bias for protein were 0.72, 0.15% and 0.00%, respectively. Sufficient levels of precision and accuracy for predicting the three major milk constituents were indicated by the high values of r² and the small values of SEP compared with the range of each constituent and by the negligible values of bias (almost zero). The performance of the calibration model for fat was excellent. The reason for the high performance of the calibration model for fat was that milk spectra had much information on fat content from scattering of light by fat globules and absorption by C-H strings and C-C strings of triacylglycerol. The results indicated that the NIR spectroscopic sensing system constructed in this study can be used for real-time on-line assessment of milk constituents during milking by a milking robot.

SCC has been accepted as the world standard for mastitis diagnosis and it is an important indicator of milk quality. A cow that produces milk containing less than 100,000 somatic cells per mL (i.e., 4 log SCC/mL) is healthy, while a cow that produces milk containing more than 200,000 somatic cells per mL (i.e., 5.3 log SCC/mL) may have subclinical mastitis (Satu, 2003). The values of r<sup>2</sup> and SEP for SCC prediction were 0.68 and 0.28 log SCC/mL, respectively. Using the calibration model for SCC to classify milk samples into two qualitative groups (milk samples from healthy cows and milk samples from other cows) gave a probability for classifying them correctly of 82% (Shenk *et al.*, 1993). Thus, the calibration model could also be used for diagnosis of subclinical mastitis.

MUN is an indicator of protein feeding efficiency in dairy cows (Godden *et al.*, 2001; Nousiainen *et al.*, 2004). When MUN is very low, milk production becomes poor. On the other hand, when MUN is very high, environmental nitrogen emission is increased by urine and fecal output from the cow (Frank *et al.*, 2002) and infertility of the cow increases (Rajala-Schultz *et al.*, 2001). The values of  $r^2$  and SEP for MUN prediction were 0.53 and 1.50 mg/dL, respectively. The performance of the calibration model for MUN was lower than the performance of the calibration models for other milk quality items. However, a calibration model with these levels of accuracy and precision could be used for monitoring the nutritional status of individual cows.

The results of validation of the calibration models developed in this study indicated that the NIR spectroscopic sensing system could be used to assess milk quality during milking. The samples for the calibration subset and validation subset were taken within the same time period in this study. In the practical use of the NIR sensing system for real-time on-line monitoring of milk quality, calibration models must be used for unknown samples, i.e., calibration models developed from samples taken in one time period must be used to assess milk quality in a later time period. To maintain the precision and accuracy of the calibration models in practical use, it is necessary to update the calibration sample set periodically and develop updated calibration models. Future work is needed for practical use of the NIR sensing system to monitor milk quality during milking.

### 4. Conclusion

The on-line NIR spectroscopic sensing system developed in this study can be used for

real-time on-line monitoring of fat, protein, lactose, SCC and MUN during milking by a milking robot with sufficient precision and accuracy. The system can provide dairy farmers with information on milk quality and physiological condition of an individual cow and therefore give them feedback control for optimizing dairy farm management. By using the system, dairy farmers will be able to produce high-quality milk and precision dairy farming will be realized.

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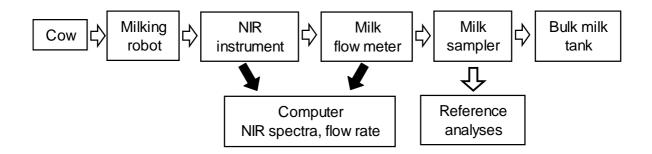


Fig. 1. Flow chart of an on-line near-infrared spectroscopic sensing system for assessing milk quality in an automatic milking system.

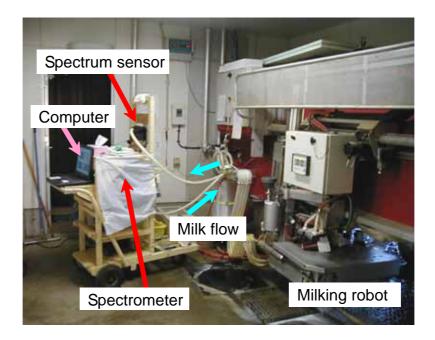


Fig. 2. On-line near-infrared spectroscopic sensing system installed in an automatic milking system.

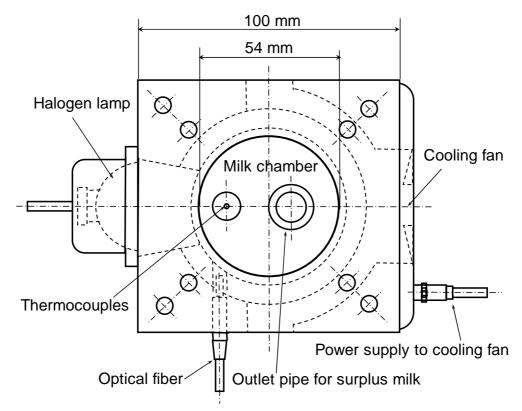


Fig. 3. Plane view of the near-infrared spectrum sensor.

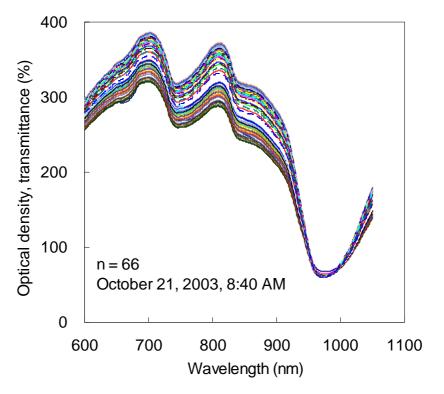


Fig. 4. Original spectra of unhomogenized milk from cow number 304 during milking on October 21, 2003.

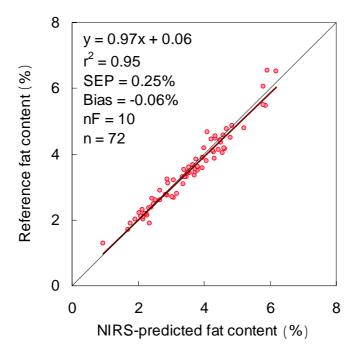


Fig. 5. Correlation between reference fat content and NIRS-predicted fat content.

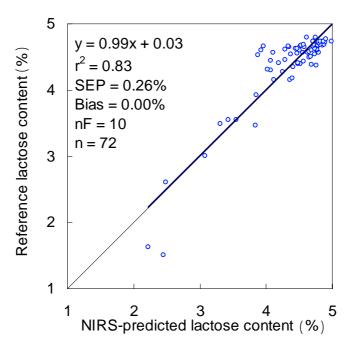


Fig. 6. Correlation between reference lactose content and NIRS-predicted lactose content.

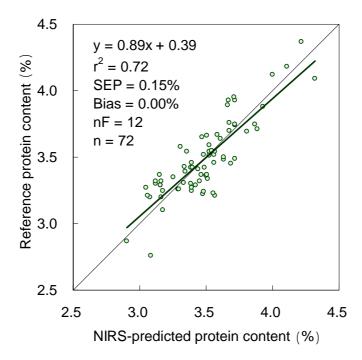


Fig. 7. Correlation between reference protein content and NIRS-predicted protein content.

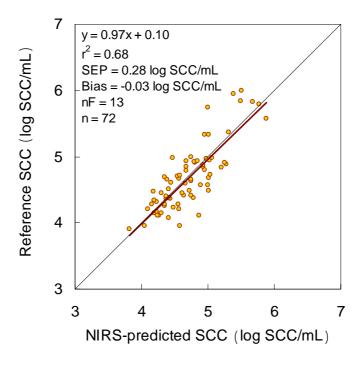


Fig. 8. Correlation between reference SCC and NIRS-predicted SCC.

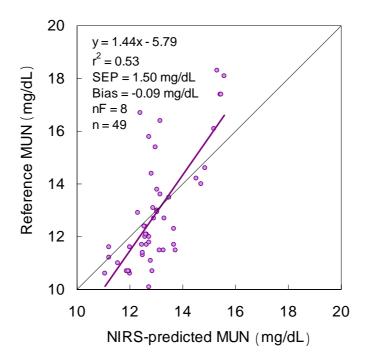


Fig. 8. Correlation between reference MUN and NIRS-predicted MUN.

Table 1. Specifications of the near-infrared spectroscopic instrument

|  | 0 '(' ('   |  |  |  |  |
|--|--|--|--|--|--|
| Devices  | Specifications   |  |  |  |  |
| Spectrum sensor                                  | Diffusion transmittance  |  |  |  |  |
| Spectrum sensor                                  | spectrum sensor  |  |  |  |  |
| Light source                                     | Halogen lamp   |  |  |  |  |
| Ontical fibor                                    | Silica glass fiber,  |  |  |  |  |
| Optical fiber                                    | 0.6-mm in diameter   |  |  |  |  |
| Milk chamber surface                             | Glass  |  |  |  |  |
| Volume of milk sample                            | Approx 230 mL  |  |  |  |  |
| Distance between                                 | 93 mm  |  |  |  |  |
| optical axis and milk level                      | 90 Hilli   |  |  |  |  |
| Spectrometer                                     | Diffraction grating spectrometer                                       |  |  |  |  |
| Optical density                                  | Transmittance  |  |  |  |  |
| Wavelength range                                 | 000 4050 4 !.(   |  |  |  |  |
| vvavololigili laligo                             | 600 - 1050 nm , 1-nm intervals   |  |  |  |  |
| Wavelength resolution                            | Approx 5 nm  |  |  |  |  |
| 0  | •  |  |  |  |  |
| Wavelength resolution                            | Approx 5 nm  |  |  |  |  |
| Wavelength resolution Photocell                  | Approx 5 nm<br>Linear array CCD, 2048 pixels                           |  |  |  |  |
| Wavelength resolution Photocell Thermocontroller | Approx 5 nm<br>Linear array CCD, 2048 pixels<br>Peltier cooling system |  |  |  |  |

Table 2. Cows used in the experiment

| Cow<br>number | Date of birth | Date of latest calving | Calving times | Experimental date and time in 2003   |                                | Number of reference samples |
|---------------|---------------|------------------------|---------------|--------------------------------------|--------------------------------|-----------------------------|
| 263           | Oct 01, 1998  | Jan 05, 2003           | 3             | Oct 20<br>Oct 20<br>Oct 21           | 20:22<br>22:40<br>7:11         | 9                           |
| 281           | Apr 28, 1997  | Oct 20, 2002           | 4             | Oct 20<br>Oct 21                     | 19:00<br>3:49                  | 9                           |
| 297           | Jan 01, 1999  | Aug 09, 2003           | 3             | Oct 20<br>Oct 21                     | 15:59<br>5:11                  | 11                          |
| 300           | Mar 14, 1999  | Mar 08, 2003           | 3             | Oct 20<br>Oct 21                     | 20:08<br>6:41                  | 17                          |
| 301           | Mar 18, 1999  | Apr 09, 2003           | 3             | Oct 20<br>Oct 21                     | 21:32<br>8:05                  | 12                          |
| 304           | Apr 27, 1999  | Apr 16, 2003           | 3             | Oct 21<br>Oct 21                     | 1:12<br>8:40                   | 15                          |
| 310           | Jun 25, 1999  | Jul 08, 2003           | 3             | Oct 20<br>Oct 20                     | 15:38<br>23:06                 | 9                           |
| 311           | Apr 10, 1999  | Oct 08, 2003           | 3             | Oct 21                               | 5:39                           | 5                           |
| 312           | May 18, 1999  | Sep 07, 2003           | 3             | Oct 20<br>Oct 20<br>Oct 21<br>Oct 21 | 14:45<br>22:24<br>6:10<br>8:16 | 16                          |
| 320           | Sep 02, 1999  | Dec 18, 2002           | 2             | Oct 20                               | 19:11                          | 7                           |
| 322           | Nov 04, 1999  | May 24, 2003           | 2             | Oct 20<br>Oct 21                     | 20:46<br>7:02                  | 13                          |
| 325           | Feb 19, 2000  | Mar 03, 2003           | 2             | Oct 20<br>Oct 21<br>Oct 21           | 20:29<br>5:50<br>8:46          | 9                           |
| 326           | Oct 29, 1999  | Aug 07, 2003           | 2             | Oct 20<br>Oct 21                     | 19:47<br>6:00                  | 20                          |
| 331           | Mar 03, 2000  | Mar 03, 2003           | 2             | Oct 21                               | 6:52                           | 8                           |
| 333           | Mar 23, 2000  | Sep 02, 2003           | 2             | Oct 20<br>Oct 21                     | 20:57<br>4:58                  | 19                          |
| 334           | Mar 27, 2000  | May 26, 2003           | 2             | Oct 21                               | 1:35                           | 7                           |
| 344           | Jul 25, 2000  | Sep 13, 2003           | 2             | Oct 20<br>Oct 20<br>Oct 21           | 15:08<br>22:08<br>4:09         | 30                          |

 $Table\ 3.\ Validation\ statistics\ of\ the\ near-infrared\ spectroscopic\ sensing\ system\ for\ determination\ of\ milk\ quality$ 

|                    | Calibration |    |               |                | Validation |    |               |                |      |       |                   |
|--------------------|-------------|----|---------------|----------------|------------|----|---------------|----------------|------|-------|-------------------|
| Milk quality items | n1          | nF | Range         | r <sup>2</sup> | SEC        | n2 | Range         | r <sup>2</sup> | SEP  | Bias  | Regression line   |
| Fat(%)             | 144         | 10 | 1.01 - 7.39   | 0.96           | 0.24       | 72 | 0.94 - 6.19   | 0.95           | 0.25 | -0.06 | y = 0.97 x + 0.06 |
| Lactose(%)         | 144         | 10 | 2.06 - 5.06   | 0.82           | 0.28       | 72 | 2.22 - 4.99   | 0.83           | 0.26 | 0.00  | y = 0.99 x + 0.03 |
| Protein(%)         | 144         | 12 | 2.77 - 4.38   | 0.78           | 0.15       | 72 | 2.91 - 4.32   | 0.72           | 0.15 | 0.00  | y = 0.89 x + 0.39 |
| SCC<br>(logSCC/mL) | 144         | 13 | 3.74 - 5.84   | 0.64           | 0.33       | 72 | 3.82 - 5.88   | 0.68           | 0.28 | -0.03 | y = 0.97 x + 0.10 |
| MUN(mg/dL)         | 98          | 8  | 10.41 - 15.73 | 0.31           | 1.68       | 49 | 11.07 - 15.58 | 0.53           | 1.50 | -0.09 | y = 1.44 x - 5.79 |

n1: total number of calibration samples. n2: number of validation samples. nF: number of factor  $r^2$ : coefficient of determination. SEC: standard error of calibration SEP: standard error of prediction. Regression line: regression line from predicted value (x) to reference value (y).