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Effectiveness of mid-infrared spectroscopy to predict the color of bovine milk and the relationship between milk color and traditional milk quality traits

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ABSTRACT

The color of milk affects the subsequent color features of the resulting dairy products; milk color is also related to milk fat concentration. The objective of the present study was to quantify the ability of mid-infrared spectroscopy (MIRS) to predict color-related traits in milk samples and to estimate the correlations between these color-related characteristics and traditional milk quality traits. Mid-infrared spectral data were available on 601 milk samples from 529 cows, all of which had corresponding gold standard milk color measures determined using a Chroma Meter; milk color was expressed using the CIELAB uniform color space. Separate prediction equations were developed for each of the 3 color parameters (L^* = lightness, a^* = greenness, b^* = yellowness) using partial least squares regression. Accuracy of prediction was determined using both cross validation on a calibration data set ($n = 422$ to 457 samples) and external validation on a data set of 144 to 152 samples. Moderate accuracy of prediction was achieved for the b^* index (coefficient of correlation for external validation = 0.72), although poor predictive ability was obtained for both a^* and L^* indices (coefficient of correlation for external validation of 0.30 and 0.55, respectively). The linear regression coefficient of the gold standard values on the respective MIRS-predicted values of a^* , L^* , and b^* was 0.81, 0.88, and 0.96, respectively; only the regression coefficient on L^* was different from 1. The mean bias of prediction (i.e., the average difference between the MIRS-predicted values and gold standard values in external validation) was not different from zero for any of 3 parameters evaluated. A moderate correlation (0.56) existed between the MIRS-predicted L^* and b^* indices, both of which were weakly correlated with the a^* index. Milk fat, protein, and casein were moderately correlated with

both the gold standard and MIRS-predicted values for b^* . Results from the present study indicate that MIRS data provides an efficient, low-cost screening method to determine the b^* color of milk at a population level. **Key words:** cow milk, yellowness, Fourier transform infrared, grazing system

INTRODUCTION

Product color is one of the primary factors considered by consumers when making purchasing decisions, as it is often an indicator of ripeness, freshness, food safety, and attractiveness in the food industry (Hutchings, 1994). It is well known that milk color influences the color features of the subsequent dairy products, while also being related to the fat content of the milk (Winkelmann et al., 1999). Differences in milk color can also be related to the presence of abnormalities in milk; for example, mastitis attributable to *Streptococcus esculin* infection causes milk to have a more reddish/yellowish color, whereas mastitis due to *Streptococcus dysgalactiae* also leads to a change in milk color (Espada and Vijverberg, 2002).

The white color of milk is a function of the milk's physical structure; the dispersion of both casein micelles and fat globules in the milk is responsible for the diffusion of incident light and is related to lightness (L^* ; Raty and Peiponen, 1999). The natural pigmentations from carotenoids, protein, and riboflavin are also associated with the white color of milk. Milk with a low carotenoid content, high protein, and high riboflavin tends to be whiter (Solah et al., 2007), or in other words have a greater L^* index value.

The yellow color (yellowness index; b^*) of bovine milk is closely related to the level of β -carotene and fat content; a greater milk fat and β -carotene content results in an incremental increase to the b^* index of milk, hence the milk will have a more yellow color. Feeding and selective breeding of cows may be used to alter the carotenoid level and thus color of dairy products (Norieze et al., 2006b). Cows fed grass silage

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tend to produce milk with yellower fat and greater β -carotene content than milk produced by cows on a hay diet (Nozière et al., 2006a; Calderon et al., 2007). Breeds of cows, such as Jerseys, that produce milk with a greater carotenoid and fat content, produce more yellow color milk than breeds such as Holstein-Friesians (Winkelman et al., 1999). Loss of carotenoids from milk is minimal when transferred into butter and cheese, therefore also contributing to the yellow coloration of these dairy products.

Yellower dairy products may be considered favorable or unfavorable depending on the target market. For example, yellower products are considered an unfavorable attribute in Middle Eastern dairy markets (Keen and Wilson, 1992). However, in Europe, a yellower color is favorable in high-fat dairy products such as butter and full-fat cheeses (Casalis et al., 1972; Hutchings, 1994).

Because the gold standard methods for the determination of milk color [i.e., Chroma Meter (Minolta, Osaka, Japan) or a NH310 Color Meter Milk (Shenzhen 3NH Technology Co. Ltd., Shenzhen, China)] or for the determination of milk carotenoid content, can be relatively costly and also require sub-sampling of milk for analysis, the use of an analytical system already in place (e.g., mid-infrared spectroscopy; **MIRS**) to determine milk color may be more logical. Mid-infrared spectroscopy is currently used by milk recording organizations worldwide to predict milk fat, protein, casein, and lactose concentration and has recently been used to predict more detailed milk composition traits (De Marchi et al., 2014) or animal traits (McParland et al., 2014). The use of MIRS to predict novel milk quality traits is therefore appealing because the mid-infrared spectrum is available at a negligible additional cost and may be undertaken as part of the routine quantification of other components in milk. Nevertheless, to our knowledge, no study has attempted to evaluate the potential of MIRS to predict milk color traits.

The aim of the present study was to evaluate the ability of MIRS to predict milk color-related traits and to estimate the correlations between these milk color traits and a selection of traditional milk quality traits.

MATERIALS AND METHODS

Milk Sample Collection

Between August 2013 and August 2014, inclusive, 730 milk samples from 621 cows were obtained from 7 research farms operated by the Teagasc Animal and Grassland Research and Innovation Centre (Moorepark, Fermoy, Co. Cork, Ireland). Milk composition was recorded weekly using a MilkoScan FT6000 (Foss Electronic A/S, Hillerød, Denmark), and the resulting

spectrum, containing 1,060 transmittance data in the mid-infrared region between 900 and 5,000 cm^{-1} , was stored. Following MIRS analysis, the milk samples were stored at 4°C for further analysis. Samples were selected to maximize diversity of breed [Holstein-Friesian ($n = 454$), Jersey ($n = 117$), Norwegian Red ($n = 15$), and crossbreds ($n = 144$)], stage of lactation (5 to 375 d in milk), milking time (i.e., AM or PM milking), and parity (1 to 11). Samples with preservative added ($n = 129$) were not considered in the present study for the determination of milk color. The final data set used in the present study comprised 601 milk samples, 461 of which were from spring-calving cows fed a predominantly grazed grass-based diet and the remaining 140 samples were from autumn-calving cows fed a TMR diet in the early stages of lactation.

Gold Standard Milk Color Determination

Milk color was measured using a Chroma Meter CR400 (Konica Minolta Sensing Europe, Nieuwegein, the Netherlands) with a closed cone, set on the $L^* a^* b^*$ system, and the Chroma meter was calibrated on a white tile. A 10-mL sub-sample of each milk sample was measured in a cuvette and expressed using the CIE- $L^* a^* b^*$ uniform color space (CIELAB, 1976). The CIE- $L^* a^* b^*$ plots the color coordinates in a uniform color space, which has an L^* , a^* , and b^* axis, with L^* (lightness; on a scale from 0 to 100, where 0 = black and 100 = white), a^* (where $-a^*$ has a green color and $+a^*$ has a red color), and b^* (where $-b^*$ has a blue color and $+b^*$ has a yellow color). The more different from zero or the greater the absolute value is, the stronger the color (i.e., a sample with an absolute value close to zero has a lighter color than a sample with an absolute value close to 100).

Data Analysis

Outlier samples were considered to be samples with a gold standard value >3 from the mean. No L^* or b^* indices outliers were removed, but 28 outliers were removed based on the a^* index. All 3 milk color traits were normally distributed. Descriptive statistics were calculated within the Holstein-Friesian and Jersey breeds separately, as well as across all breeds combined and within season. The differences between the means of Holstein Friesian and Jersey cows and between the means of autumn and spring cows were derived using ANOVA in Microsoft Excel (Microsoft Corp., Redmond, WA). Spectral data were transformed from transmittance to linear absorbance using a logarithmic transformation of the reciprocal of the wavelength values (Soyeurt et al., 2011). Prediction models were developed using un-

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