# Prediction of cheese yield using near infrared spectroscopy

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## Introduction

Cheese yield is the amount of cheese manufactured from a given amount of milk<sup>1</sup> and is a major factor in the efficiency and profitability of cheese production.<sup>2</sup> Cheese yield is influenced by many factors, including i) milk composition,<sup>3,4</sup> especially protein (casein) and fat concentrations,<sup>5,6</sup> ii) milk pretreatments, iii) coagulant type, and iv) somatic cell counts.<sup>7</sup> Cheese yield is also influenced by the aptitude of milk to coagulate,<sup>8,9</sup> which is commonly defined on the basis of three parameters: a) milk coagulation time, b) curd firmness and c) speed of aggregation. Coagulation time and curd firmness have been studied extensively and are considered, together with fat and casein contents, in the development of cheese yield predictive formulas. The relationship between curd firmness and cheese yield is not linear; in particular, the influence of curd firmness on cheese yield is greater if the fat content is low.<sup>10</sup>

Selecting milk for cheese making based on quality and adequate coagulation properties could greatly assist cheese makers in obtaining greater and more consistent cheese yields.<sup>11</sup> It is therefore important to develop a rapid method for estimating final cheese yield based on raw material. This could allow cheese makers to have a constant check on the efficiency of the technological operations. At the same time, calculating the effects of individual milk components on cheese yield would permit a milk quality payment system that could remunerate each parameter for its actual value. The use of near infrared (NIR) spectroscopy coupled with chemometrics can provide continuous information about the composition of milk during cheese making and contribute to the control of the process: thus leading to an improvement of cheese quality.<sup>12</sup> The objective of this study was to investigate the utility of NIR spectroscopy for the development of an equation capable of predicting cheese yield during milk collection at the farm.

## **Materials and Methods**

#### Samples

Twenty nine raw milk samples were collected monthly (July to October 2008) from a dairy company Latteria Soresina (Soresina, Cremona, Italy). Each month, milk was taken from 2 tanks for creaming, before it was divided into several copper vats in which protected designation of origin (PDO) Grana Padano hard cooked cheese was made. Samples were representative of the milk collected daily for use in an Italian cheese making factory.

Fat and total casein percentage,  $\alpha_{s1}$ ,  $\beta$  and  $\kappa$ -casein amount,  $\alpha$ -lactoalbumin and  $\beta$ -lattoglobulin concentrations, and protein amount (as the sum of serum proteins and casein concentration) were determined by official methods of analysis.<sup>13</sup> The actual cheese yield was expressed as a percent ratio between cheese weight (CW) and milk weight (MW), and evaluated on the same day of the cheese making.

#### Near infrared spectroscopy

Before cheese making, milk samples were evaluated for the aptitude to coagulate by Optigraph instrument (Ysebaert, Frepillon, France),<sup>14</sup> based on NIR principles (fixed wavelength = 890 nm). Milk samples were then heated to the renneting temperature ( $35 \pm 2^{\circ}$ C). The rennet solution (chymosin 20% ± 5, REMCAT 130 IMCU.g<sup>-1</sup>, Naturen, CHR Hansen, Milan) was diluted (1.6 ml.100ml<sup>-1</sup>) in sodium acetate-3-hydrate (0.07M)/acetic acid (1M) buffer (pH 5.5), and 200 µl of the solution was added to 10 ml of milk. Milk samples were monitored for 60 min after rennet addition.

Three parameters were obtained from the observed curves: milk coagulation time (CT), indicated by the maximum of the first derivative of the signal; curd firmness (CF), determined 30 min after the addition of the rennet, and the aggregation rate (AR), calculated from the slope of the linear region of the curve.

### Data processing

Pearson correlation was used to determine the significance and direction of a possible correlation among variables that can influence cheese yield (p < 0.05).<sup>15</sup> Models for the prediction of cheese yield were developed using partial least squares (PLS) regression and validated by cross-validation. Prior to PLS regression, all data were standardised, i.e. first mean centered then scaled to unit variance. PLS regression was carried out using The Unscrambler 9.2 (Camo inc., Oslo, Norway).

#### **Results and Discussion**

The relationships between cheese yield and milk quality plus milk clotting ability, assessed using Pearson's correlation coefficients, are reported in Table 1.

Fat g.l⁻¹	Protein g.l <sup>-1</sup>	Casein g.l <sup>-1</sup>	Coagulation time min	Curd firmness mA	α-S1 casein g.l <sup>-1</sup>	β casein g.l <sup>-1</sup>	K casein g.l <sup>-1</sup>	Cheese yield
1								
0.82***	1							
0.97***	0.87***	1						
0.54**	0.62**	0.56**	1					
-0.23 <sup>ns</sup>	-0.25 <sup>ns</sup>	-0.19 <sup>ns</sup>	-0.30 <sup>ns</sup>	1				
0.79***	0.97***	0.81***	0.62**	-0.22 <sup>ns</sup>	1			
0.83*** 0.49** <b>0.98</b> ***	0.97*** 0.79*** <b>0.83</b> ***	0.88*** 0.51** <b>0.95</b> ***	0.60** 0.61** <b>0.59*</b> *	-0.27 <sup>ns</sup> -0.26 <sup>ns</sup> <b>-0.24<sup>ns</sup></b>	0.94 <sup>ns</sup> 0.81*** <b>0.81</b> ***	1 0.70*** <b>0.83</b> ***	1 <b>0.50</b> *	1
	Fat g.l <sup>-1</sup> 1 0.82*** 0.97*** 0.54** -0.23 <sup>ns</sup> 0.79*** 0.83*** 0.49** <b>0.98</b> ***	Fat g.l <sup>-1</sup> Protein g.l <sup>-1</sup> 1 0.82***   0.97*** 1   0.54** 0.62**   -0.23 <sup>ns</sup> -0.25 <sup>ns</sup> 0.79*** 0.97***   0.83*** 0.97***   0.49** 0.79***   0.98*** 0.83***	$\begin{array}{c} Fat\\ g.l^{-1} \\ \end{array} \begin{array}{c} Protein\\ g.l^{-1} \\ \end{array} \begin{array}{c} Casein\\ g.l^{-1} \\ \end{array} \begin{array}{c} Gasein\\ Gasei$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$

|--|

\*\*\* P < 0.001; \*\* P < 0.01; \* P < 0.5; ns = not significant

Cheese yield was more positively correlated with fat, protein and casein ( $R^2 = 0.92$ , p < 0.001) than with variables related to milk clotting ability (coagulation time,  $R^2 = 0.59$ , p < 0.05; curd firmness,  $R^2 = -0.24$ ). Concentrations of protein (casein) and fat in milk have previously been shown to be important influences on cheese yield.<sup>16</sup> Casein amount and quality are the most important factors in determining yield, because this protein aggregate forms the structural matrix (network of micelles) of cheese and can retain fat and moisture; casein is also highly hydrated and thus retains moisture during cheesemaking.<sup>17</sup> The obtained results were in agreement with the reported literature, showing a strong positive correlation between cheese yield and casein content (specially  $\alpha_{S1}$ - and  $\beta$ -casein; Table 1). Casein subunits play a very important role in the formation of casein micelles. Their rather constant relationship can be changed, and these variations significantly affect the degree of dispersion of the milk system and, consequently, the properties of the native casein complex, with significant influence on the milk coagulation, cheese yield and cheese quality.<sup>18</sup>

Previous studies have found that  $\alpha_{s1}$ -casein has a significant influence on protein content<sup>19</sup>, while  $\beta$ -casein genotypes were found to be associated with fat percentage, fat and protein yield<sup>20</sup> and curd firmness<sup>21</sup>. No correlations were found between curd firmness, fat and protein content; the amount of fat appears to have no influence the coagulum strength, nor was it able to be completely retained during the cheese making process. Protein content was influenced by the total casein content and its relative composition.

A prediction model of cheese yield, developed using PLS regression, was applied to the 29 milk samples. The accuracy of this model was assessed using the ratio of performance to deviation (RPD) and the correlation coefficient (r) for predicted vs measured data in cross-validation.

The correlation coefficients obtained for standardised variables that most contributed to the prediction of cheese yield were: fat, total protein, casein, and variables of milk clotting ability (coagulation time, curd firmness and speed of aggregation). The obtained PLS predictive equation was:

Cheese yield =  $9.726 + (0.581* \text{ fat}) + (0.345* \text{ casein}) + (0.017* \text{ total protein}) + (0.138* \text{ coagulation time}) + (-2.54e^{-02*} \text{ curd firmness}) + (-0.153* \text{ speed of aggregation})$ 

The predictive equations used 3 latent variables selected by cross-validation. The calibration and the validation curves of the PLS model are shown in Figure 1.

This model was characterised by r (correlation coefficient) = 0.98, which indicated a good provisional model, and RMSECV = 0.036 (Table 2).



**Figure 1.** Linear regression between the predicted values, obtained by cross-validation for the cheese yield of the 29 samples and the real cheese yield of the same samples.

Table 2. Statistical parameters associated with
calibration and prediction curves for cheese
yield.

Parameter	Sample set			
	Calibration	Validation		
Number of samples	29	29		
Range	7.96–8.61	7.93–8.61		
Mean	8.23	8.23		
Standard deviation (SD)	0.26	0.25		
r	0.98	0.98		
RMSEC	0.036			
RMSEP		0.53		
Slope	0.98	0.95		
Bias	-7.064x10 <sup>-7</sup>	-0.006		

The predicted cheese yield model (RPD = 4.82) was found to be of high practical utility in agreement with RPD classification reported by Williams.<sup>22</sup> The PLS model can be considered good, even if the real cheese yield was overestimated by an average of 1.36%.

## Conclusion

These preliminary results demonstrated the suitability of NIR spectroscopy for predicting cheese yield without excessive sample preparation and lengthy analyses. The suitability of NIR spectroscopy for the determination of fat, protein and casein contents had already been demonstrated in previous papers.<sup>23,24</sup> The collected information could be a basis for the implementation of NIR applicability during the cheese production process in order to promote frequent controls during the whole production chain, from milking to cheese ripening.

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