



Influence of curd cutting programme and stirring speed on the prediction of syneresis indices in cheese-making using NIR light backscatter

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ABSTRACT

An NIR reflectance sensor, with a large field of view and a fibre-optic connection to a spectrometer for measuring light backscatter at 980 nm, was used to monitor the syneresis process online during cheese-making with the goal of predicting syneresis indices (curd moisture content, yield of whey and fat losses to whey) over a range of curd cutting programmes and stirring speeds. A series of trials were carried out in an 11 L cheese vat using recombined whole milk. A factorial experimental design consisting of three curd stirring speeds and three cutting programmes, was undertaken. Milk was coagulated under constant conditions and the casein gel was cut when the elastic modulus reached 35 Pa. Among the syneresis indices investigated, the most accurate and most parsimonious multivariate model developed was for predicting yield of whey involving three terms, namely light backscatter, milk fat content and cutting intensity ($R^2 = 0.83$, $SEy = 6.13$ g/100 g), while the best simple model also predicted this syneresis index using the light backscatter alone ($R^2 = 0.80$, $SEy = 6.53$ g/100 g). In this model the main predictor was the light backscatter response from the NIR light back scatter sensor. The sensor also predicted curd moisture with a similar accuracy.

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1. Introduction

The rate and extent of syneresis are critical in determining the composition, yield and quality attributes of cheese (Lawrence & Gilles, 1980; Pearse & Mackinlay, 1989; Daviau et al., 2000). Syneresis is empirically controlled by temperature, pH, enzyme concentration and processing time. A better control of syneresis would improve the consistency of curd moisture and help to keep it within the legal limits. 'Weak body', texture defects and a propensity to develop off-flavours in commercial cheddar cheese were found to be associated with high moisture (Fox, 1975). It is proposed that online control of syneresis could decrease the production of downgraded cheese and improve cheese quality.

Due to the increasing scale of manufacturing, a more automated system for controlling cheese manufacture would be desirable and this may include the monitoring of milk coagulation and/or syneresis. Castillo, Payne, Hicks, and Lopez (2000) found that

a fibre-optic light backscatter sensor (CoAguLite, CL) can be used to measure changes in diffuse reflectance of milk and is an objective approach for monitoring the progress of milk coagulation. O'Callaghan, Mulholland, Duffy, O'Donnell, and Payne (2001) and O'Callaghan, O'Donnell, and Payne (2002) evaluated a range of online sensor technologies (near infrared light backscatter and thermal hot-wire sensors) to determine the cutting time of milk gels and found that a near infrared sensor gave the best prediction of curd formation in situations where the protein level varied. Castillo, Lucey, Wang, and Payne (2006) suggested that it may be possible to develop a light backscatter sensor capable of monitoring both coagulation and syneresis over a range of coagulation temperature and inoculum concentration conditions, which could lead to better control of moisture content and an improvement in the final properties of cheese such as homogeneity and quality.

Other technologies are also being investigated for monitoring syneresis. Everard et al. (2007) studied the influence of milk pH and stirring speed on cheese curd syneresis by computer vision and colour measurement techniques. These techniques showed potential for monitoring syneresis in cheese-making, although the study had the limitation that optical measurements were taken at the surface, and it was found that low stirring speeds were not

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effective in re-suspending sinking curd and this confounded the prediction of curd moisture.

Taifi et al. (2006) used an ultrasonic technique to monitor coagulation and syneresis under quiescent conditions and, as a result, the proposed technique is not applicable while cutting and stirring the milk gel.

In previous studies a large field of view sensor (LFV) for monitoring both milk coagulation and curd syneresis was designed by Castillo, Payne, and Shea (2005) and evaluated over a range of cutting times, temperatures and calcium chloride levels (Fagan et al., 2007a). Fagan et al., 2007a and Fagan et al., 2007b also investigated if the proposed combined sensor technology for coagulation and syneresis monitoring could be used to improve curd moisture content control. They showed that curd moisture and yield could potentially be predicted using an LFV sensor and that the sensor response was most sensitive to coagulation and syneresis processes at 980 nm.

The objective of this study was to investigate the prediction of syneresis indices (curd moisture content, yield of whey and fat losses to whey) during cheese-making using an LFV light backscatter sensor at 980 nm over a range of curd cutting programmes and stirring speeds.

2. Materials and methods

2.1. Milk preparation

Whole milk was reconstituted to a total solids level of 12% with target fat and protein levels of 3.5% and 3.3% respectively, in an 11 L cheese vat (Type CAL 10L, Pierre Guerin Technologies, Mauze, France) from skim milk powder (Irish Dairy Board, Dublin), distilled water and cream (Dairygold, Cork, Ireland) at 42 °C while being stirred at 44 rpm. This procedure ensured a low standard deviation in rheologically determined ($G' > 0.5$ Pa) gel times (SD = 1.3 min), under constant experimental conditions. Calcium chloride ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$) was added at 2.04 mmol/L to the milk, which was then cooled to 8 °C and held overnight under gentle agitation conditions (10 rpm).

2.2. Milk coagulation

On the subsequent day the milk was heated to 32 °C. Temperature was controlled (32 ± 0.1 °C) using a water bath (Grant Y28, Grant Instruments Ltd., UK) and a heating jacket on the vat. The milk temperature in the vat was also verified using a temperature probe (Tloop Thermometer, Sensor Tech Ltd., Louth, Ireland). Milk pH was adjusted to 6.5 at 32 °C using a 1 M HCl solution. Approximately 30 mL of milk was removed for compositional analysis

using the Milko Scan 605 (Foss Electric, Denmark) to determine fat, protein and lactose contents. Rennet (CHY-MAX extra, EC 3.4.23.4, isozyme B, 600 IMCU/mL, Chr Hansen Ireland Ltd., Ireland) was added to the milk (0.18 g of chymosin/kg of milk) in the cheese vat while being stirred constantly at 31 rpm. Stirring was stopped after 3 min and the stirrers were replaced with twin cutting blades in readiness for cutting (Fig. 1b).

2.3. Determination of cutting time and gel cutting procedure

Small amplitude oscillatory rheometry was performed to determine the gel cutting time (t_{cut}) using a Bohlin CVO Rheometer (Bohlin, Cirencester, UK). A concentric cylinder (C25) measurement system was used with a 1.25 mm gap between bob and cup. A 13 mL sample of milk was removed from the cheese vat at 3 min after rennet addition and immediately transferred to the rheometer cup which was pre-warmed to the assay temperature. The rheometer was working at a frequency of 1 Hz and a strain of 0.01 in amplitude, which is within the linear viscoelastic region. After temperature equilibration of the milk sample and the bob at 32 °C, measurements were taken every 40 s.

A full factorial experimental design with three gel cutting intensities (CI) and three curd stirring speeds (SS) was used and undertaken in three replicates ($n = 27$). When the elastic modulus, G' , reached 35 Pa, cutting of the gel was initiated ($t = 0$). Three cutting programmes, a, b and c, were used. In each case, cutting was carried out in three cycles over a total duration of 3 min with short rest periods between cycles and an increase in speed over the first two cycles as outlined by Everard et al. (2008). Different speed settings, set using a variable speed drive, were used for each programme, giving a variation in total revolutions of the cutting blades, namely 4.2, 8.3 and 12.5 for programmes a, b and c, respectively). After gel cutting, the cutting blades were replaced with the twin stirrers in order to agitate the curd/whey mixture. At $t = 4$ min stirring was initiated at the appropriate speed according to the experimental design (10, 16 or 22 rpm).

2.4. Sampling procedure for the curd/whey mixture

Curd and whey samples were removed using a specially designed online sampler, manufactured by the University of Kentucky in collaboration with Teagasc and University College Dublin (Fig. 1a). The online sampler allowed sampling to take place without interrupting stirring and without light interference in the sensor. Curd and whey samples were removed for compositional analysis at $t = 5$ min and every 10 min thereafter up to $t = 75$ min (i.e. 8 samples). The sample volume was alternated between 180 and 270 mL of curd/whey mixture. The larger volume enabled

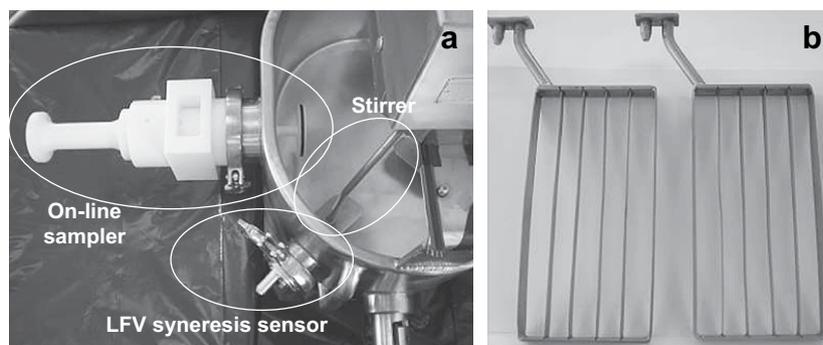


Fig. 1. (a) Cheese vat showing the online sampler for collecting curd/whey mixture from the vat during stirring, the online LFV sensor and stirrer, and (b) the twin cutting blades used to cut the milk gel.

analysis of fat in whey to be carried out. The curd/whey mixture was immediately separated using a stainless steel sieve and pan (Endecotts Ltd., London, UK) with a 75 μm absolute pore size. Whey and curd were weighed (HC32000 Reflex EE balance, Avery Weigh-Tronix, Dublin, Ireland) for determination of yields.

2.5. Measurements of syneresis indices

Yield of whey (Y_w) and curd on a wet basis (Y_c) were calculated as the percentage of the whey or curd, respectively, in each sample that was collected. Curd moisture content (M_c) was determined by oven drying (102 °C overnight) as described in Everard et al. (2008). Y_w and M_c were determined at 10 min intervals during syneresis. Whey fat (F_w) was measured by the Rose–Gottlieb method (IDF, 1987) and determined at 15 min after gel cutting and at 20 min intervals thereafter up to 75 min.

2.6. Syneresis optical measurements

In this study an upgraded NIR sensor with a large field of view (Castillo, Fagan, Payne, O'Donnell, & O'Callaghan, 2007), which measures light backscatter, was installed in the cheese vat wall for monitoring syneresis (Fig. 1a). The sensor was modified from the previous LFV sensor designed by Castillo et al. (2005) to ensure it would operate at a higher emitting light intensity and respond with greater sensitivity to the changes occurring in the cheese vat during coagulation and syneresis.

The sensor employed had a 20 mm diameter glass window (Melles Griot Inc., Rochester, NY, USA) for collecting an optical signal from the curd/whey mixture. Near infrared light from a 6 W tungsten halogen light source (model LS1B, Ocean Optics, Inc., Dunedin, FL, USA) was transmitted to the mix of curd and whey through a fixed large diameter optical fibre (5 mm diameter) (Fiberoptics Technology, Inc., Pomfret, CT, USA), a vertical polarizer (Edmund Optics, Inc., Barrington, NJ, USA) and the glass window until it reached the sample. Backscattered light was collected over a large area through the glass window. A horizontal polarizing plate ensured that any light reflected by the window was eliminated. Reflected light was then transmitted through a second fibre (5 mm diameter) and a collimating lens (Edmund Optics Inc.) that focussed the scattered light onto a $\sim 800 \mu\text{m}$ diameter fibre optic cable (Spectran Specialty Optics, Avon, CN,

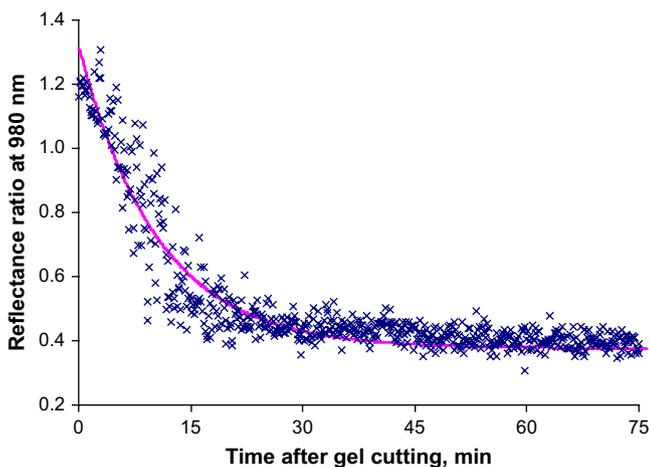


Fig. 2. Typical LFV sensor response, R_s (\times) during syneresis in this study ($T = 32 \text{ }^\circ\text{C}$; $\text{CaCl}_2 \cdot 2\text{H}_2\text{O} = 2.04 \text{ mmol/L}$) at 980 nm and fit to the first-order equation (Eq. (1)), R_t (solid line). $SS = 16 \text{ rpm}$; $CI = 4.2$ total revolutions. The 95% confidence band on the fit is ± 0.0093 .

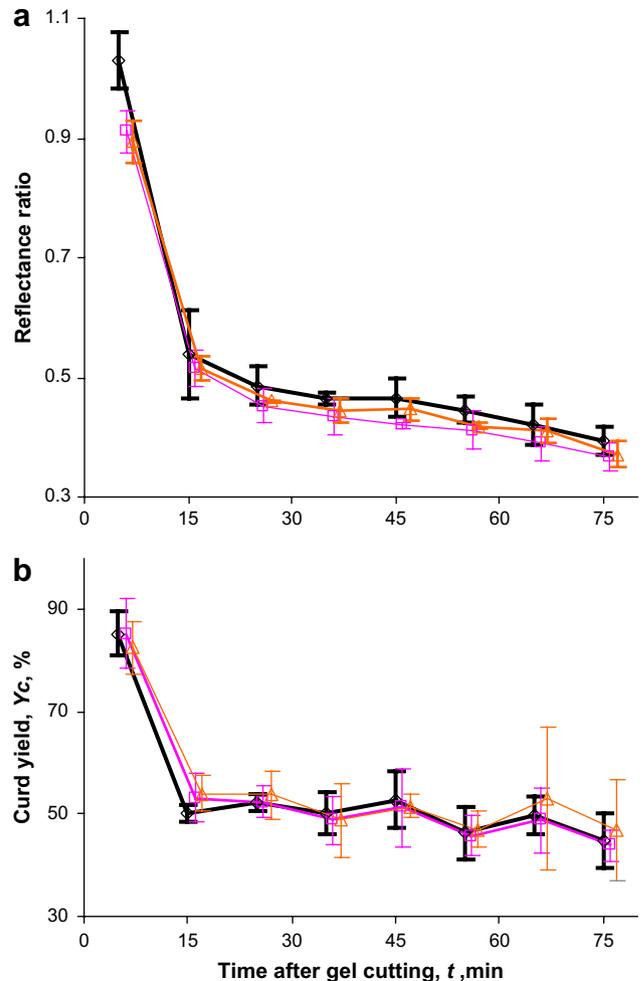


Fig. 3. (a) Light backscatter (reflectance ratio, R_t) and (b) curd yield (Y_c) vs. time after gel cutting (t), over three gel cutting programmes (\blacklozenge , a; \square , b; \triangle , c) at one curd stirring speed (22 rpm). The error bars show mean \pm SD for three replicates. While the graphs for (b) and (c) are shifted in time by 1 or 2 min, respectively, for visual clarity, it should be noted that all measurements were carried out synchronously according to the sampling times. Details of cutting programmes are given in Section 2.

USA) to the master unit of a miniature fibre optic spectrometer (HR2000CG-UV-NIR, Ocean Optics BV, Duiven, Netherlands), which was used as a light detector. The detected light was averaged and recorded at intervals of 7 s (SpectraSuite software v. 5.1, Ocean Optics BV). The data were read using The Unscrambler

Table 1

Effect of cutting intensity (CI) and time after gel cutting (t) on light backscatter (reflectance ratio) and curd yield.

Source	DF	F value ($N = 63$)	
		Y_c	R_t
CI	2	0.24 ns	2.29 ns
t	6	13.1***	810***
CI \times t	12	0.49 ns	3.10**
Residual	42		
Total	62		

Effects were determined by mixed model analysis (PROC MIXED). Y_c , curd yield; R_t , the light backscatter (reflectance ratio) from the LFV sensor at 980 nm; t , time after gel cutting. Significance: *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$; ns, not significant. Seven time-points, excluding $t = 5$ min after cutting, in each trial were included in this analysis. The table shows the variation for one stirring speed ($SS = 16 \text{ rpm}$). Similar effects were found at all curd stirring speeds.

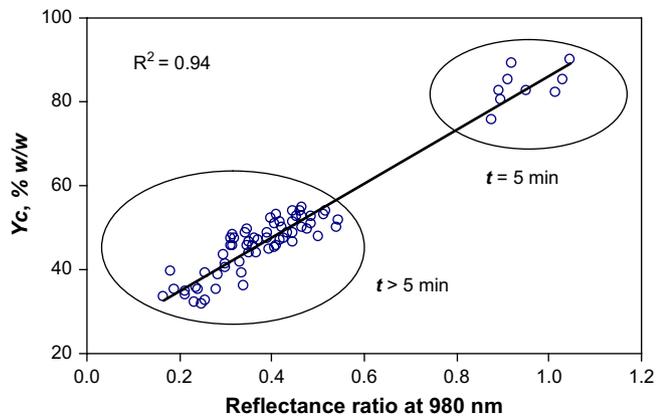


Fig. 4. Regression of curd yield (Y_c) vs. light backscatter (reflectance ratio). Each point represents the average of 3 replicates at one sampling time, i.e. 5, 15, 25, 35, 45, 55, 65 or 75 min ($P \leq 0.001$, $N = 72$).

software (v9.2, Camo Process AS, Oslo, Norway). The dark spectrum was subtracted at the start of each trial.

2.7. Calculation of the light backscatter ratio

The intensity of backscattered light at 980 nm was measured as voltage, V , and the light backscatter ratio (R_s), was calculated as V/V_0 , where V_0 was the average of voltage (sensor output) at 980 nm over the first minute after rennet addition. To eliminate scatter in the data, due to the inhomogeneous nature of curd particles, the light backscatter ratio (R_s) was then fitted to the following first-order equation (Fagan, Castillo, et al., 2007):

$$R_t = R_\infty + (R_0 - R_\infty)e^{-k_{LFV} t} \quad (1)$$

where R_t is the modelled light backscatter ratio during syneresis at time t (min), R_∞ is the light backscatter ratio at an infinite time, R_0 represents the light backscatter ratio immediately after gel cutting, and k_{LFV} is the kinetic rate constant (per minute) for the LfV sensor response during syneresis (Fig. 2). The parameters R_∞ , R_0 and k_{LFV} , were estimated using least squares optimisation with the Solver tool in Microsoft Excel.

Table 2

Multivariate linear models for prediction of whey fat loss, curd moisture content and yield of whey using optical reflectance ratio in combination with independent variables (stirring speed and cutting intensity) and co-variables (milk fat content and time after gel cutting).

Dependent variable ^a	Multivariate model						DF	SEy (g/100 g) ^d	R^2	Simple model R^2
	Significant factors, ^b their levels of significance, standardised coefficients, SC, and Student's t values, t_{St} ^c									
F_w	SS***	R_t^*	t^*	CI**	F_m^*		94	0.04	0.59	0.42 ^e
	t_{St}	5.01	2.33	-2.10	-2.93	-2.23				
	SC	0.44	0.28	-0.21	-0.21	-0.15				
M_c	R_t^*	t^{***}	SS***	F_m^{***}	CI**		194	1.10	0.82	0.65
	t_{St}	11.91	-8.42	-5.33	-5.15	3.00				
	SC	0.58	-0.39	-0.18	-0.16	0.09				
Y_w	R_t^*	F_m^{***}	CI***				196	6.13	0.83	0.80
	t_{St}	-30.2	-4.54	-4.04						
	SC	-0.91	-0.14	-0.12						

Coefficients of determination for simple models with R_t alone are shown for comparison.

^a F_w , whey fat; M_c , curd moisture content; Y_w , yield of whey.

^b R_t , the light backscatter (reflectance ratio) from the LfV sensor at 980 nm; SS, stirring speed; CI, cutting mode; t , time after gel cutting; F_m , milk fat content. In each model the factors are arranged in order of their effect as indicated by their standardised coefficients.

^c Level of significance of each factor is indicated as follows: *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$. t_{St} , Student's t value; SC, standardised coefficients.

^d SEy, standard error of estimate.

^e The simple model for F_w required a transformation in R_t , i.e. $F_w = a(1/R_t) + b$.

2.8. Statistical analysis

Multiple linear regression was used in this study to determine the significant factors to include in linear models for predicting syneresis-related parameters, using SigmaStat 3.1 software (Systat Software UK Ltd., London, UK). The regression models were tested for normality and equality of variance. The most useful factors in the models were determined by backward stepwise regression. Analysis of variance on repeated measures data (i.e. effect of time) was done by mixed model analysis, using PROC MIXED in the SAS statistical package (version 9.1.3., SAS Institute, Cary, NC, USA).

3. Results and discussion

3.1. LfV sensor response during syneresis

A typical LfV sensor response following gel cutting at 35 Pa is shown in Fig. 2, in which the decrease in R_s up to 75 min after gel cutting is observed. A rapid decrease is observed from 1.3 to 0.5 over the first 15–20 min and after 20 min R_s decreases gradually from 0.5 to 0.4. Both observations are consistent with a first order reaction, as reported by Fagan, Castillo, et al. (2007). The scatter observed in R_s in Fig. 2 is due to curd particles passing close to the sensor window.

3.2. Effect of cutting intensity and stirring speed on reflectance ratio and curd yield on wet basis

Fig. 3 shows the trends with time of the sensor response (reflectance ratio, R_t , averaged at each sampling time using Eq. (1)), and curd yield (Y_c), for all three gel cutting programmes, a, b and c (4.2, 8.3 and 12.5 total revolutions) respectively, and one curd stirring speed (22 rpm), averaged across three replicates. Mixed model analysis shows that cutting intensity (CI) did not significantly influence Y_c in this study and but it did have an interactive effect with time on the optical signal response from the LfV sensor (Table 1). It can be inferred from the error bars in Fig. 3 that the interactive effect of CI and time on R_t occurs at early times after gel cutting, i.e. around 15 min. While Fig. 3 involves one stirring speed, i.e. 22 rpm, the same effects were observed across all curd stirring speeds (Table 1).

3.3. Prediction of curd yield using NIR light backscatter

Regression of Y_c against R_t shows a linear relationship with standard error of prediction (SEy) of 0.05% w/w (Fig. 4). Thus, 94% of the variation in Y_c in this figure is explained by the sensor response, R_t . The data points are in two clusters, according to sampling times, $t = 5$ min and $t > 5$ min, respectively, reflecting the rapid changes taking place in the first 15–20 min of syneresis.

Summarising Sections 3.1–3.3, most of the variation in Y_c and R_t is an effect of time after gel cutting and most of this variation occurred in the first 20 min after gel cutting, but the variation in Y_c can be largely explained using R_t and the effect of time is accounted for.

3.4. Prediction of syneresis indices in cheese-making using NIR light backscatter

Multivariate linear models were developed using LFV sensor response and other variables, which could be used to predict the most useful measures of syneresis. For comparison, simple linear regression was used to predict the same syneresis indices using the light backscatter sensor response alone. Regression was used to obtain linear models for predicting F_w , M_c and Y_w as a function of time during syneresis using independent variables (stirring speed and cutting intensity), co-variables (milk fat content, F_m and time after gel cutting) and dependent variable (optical reflectance ratio, R_t) derived from the LFV sensor response during syneresis.

The significant factors for predicting F_w , M_c , and Y_w are shown in Table 2. The linear model predicted F_w with a SEy of 0.04 g/100 g and R^2 of 0.59 (Fig. 5a). The following five parameters listed in order of decreasing effect were used in the prediction model, i.e. SS, R_t , t , CI and F_m . This model is not likely to be suitable for prediction of whey fat loss in commercial practice, but it shows some of the main factors which contribute to loss of fat in cheese-making (such as stirring speed, time after gel cutting and cutting intensity). Thus, it was found that the light backscatter ratio, in conjunction with known technology parameters, was significantly related to changes in the fat content of the whey in the cheese vat and also that time affected the whey fat losses (Table 2). This was in accordance with the results obtained by Fagan, Castillo, O'Donnell, O'Callaghan, and Payne (2008), who came to a similar conclusion using variables in their study which covered a range of temperature, cutting time and calcium levels. The fact that light backscatter (R_t) does not have the most significant effect in the prediction model is supported by the poor R^2 of 0.42 when R_t alone is used to predict F_w in a simple model.

M_c and Y_w were predicted with SEy of 1.10 and 6.13 g/100 g, respectively, and the models fitted with R^2 of 0.82 and 0.83, respectively (Fig. 5b and c). As in Fig. 4, the data points in Fig. 5b,c fall into two clusters according to time of sampling, i.e. when $t = 5$ min and the other is when $t > 5$ min, due to the syneresis rate diminishing when the contraction of the casein matrix starts levelling out around 15–20 min after gel cutting. Five distinct terms were used to predict the linear model of M_c , i.e. R_t , t , SS, F_m and CI, in that order of significance. The primary significance of R_t as a factor reflects the fact that R_t alone explains 65% of the variation in M_c (Table 2). The significance of time after gel cutting and of fat in milk confirm the findings of Everard et al. (2008) vis-à-vis the same work. In agreement with the findings of Fagan et al. (2008), the results showed that light backscatter ratio was a good predictor in the M_c model in conjunction with milk composition and cheese technology parameters. The different studies, involving different sensors and cheese vats, produced different models, although broadly similar in principle. This relates to (i) different experimental variables used, (ii) differences in cheese-making

technology, e.g. gel cutting systems, curd/whey sampling systems, and (iii) differences in the milk used.

The last linear model was developed to predict Y_w using three factors, i.e. R_t , F_m and CI, in that order of significance. This model (along with the model for M_c) gave the best overall prediction in terms of $R^2 = 0.83$, i.e. proportion of explained variation. It appears that with known milk fat and under given cutting conditions, 83% of the variation in yield of whey (and curd) can be accounted for. R_t was very a significant term in the prediction Y_w model over the course of syneresis, giving an R^2 of 0.80 when R_t is used alone in

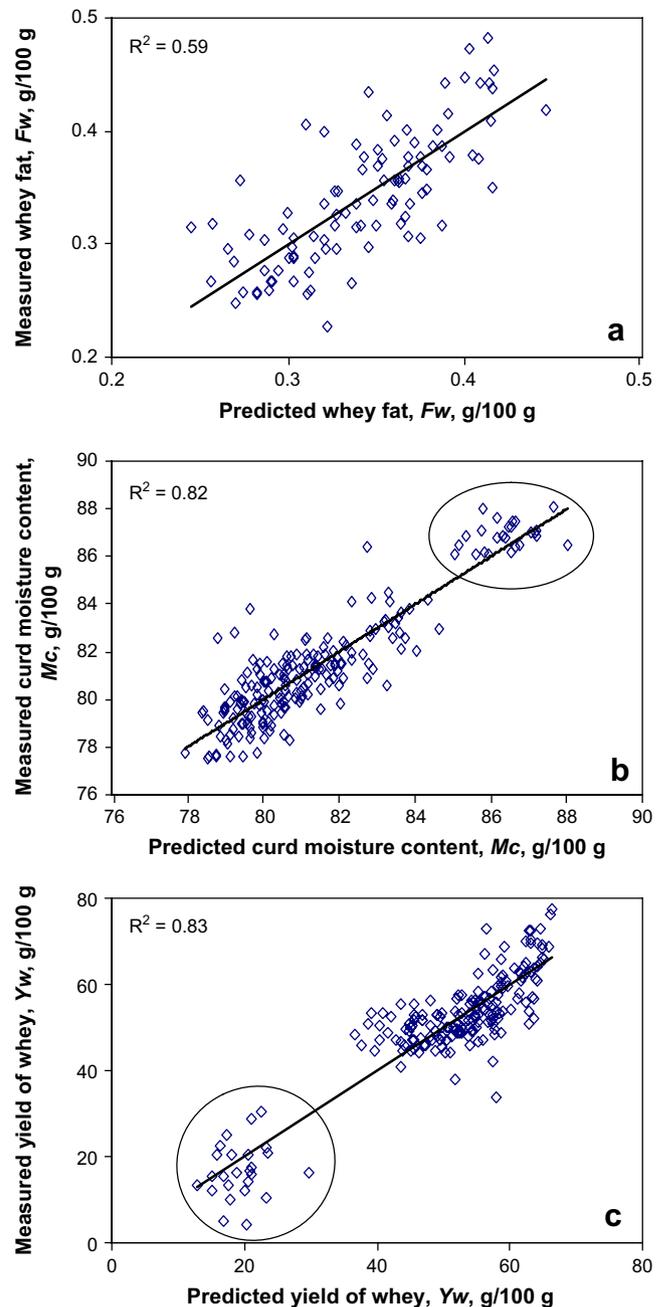


Fig. 5. Measured vs. predicted syneresis indices using linear models (cf. Table 2): (a) whey fat model, F_w (SS, R_t , t , CI, F_m), $N = 100$, (b) curd moisture content model, M_c (R_t , t , SS, F_m , CI), $N = 200$, and (c) yield of whey model, Y_w (R_t , F_m , CI), $N = 200$. F_w was determined at 20 min intervals between $t = 15$ and $t = 75$ min after gel cutting; M_c and Y_w were determined at 10 min intervals from $t = 5$ –75 min after gel cutting. The ellipses enclose data points corresponding to $t = 5$ min.

predicting yield of whey (Table 2). Combining the findings of Everard et al. (2008), which showed that yield of whey is largely a function of time and is not influenced by the cutting programme, with our finding that Y_w can be predicted from R_t , F_m and CI, it can be concluded that R_t accounts for the effects of time after gel cutting, t , and that CI appears in the prediction model to compensate for the influence of other parameters on R_t , which are influenced by CI. This hypothesis explains why time is needed in a model for predicting Y_w when the optical sensor is not available but is omitted when the optical sensor is used, and also why CI is included in the latter situation although it has no significant influence in the former situation. Marshall (1982) found that increased fat concentration in milk caused a large reduction in the rate of syneresis. Our study shows that F_m was a factor, though of lesser significance than R_t , in the last two models, showing that when milk composition varies (in this case fat and water content), this needs to be explicitly accounted for in a prediction model involving R_t .

4. Conclusions

This study demonstrated the potential of an LFV NIR sensor to predict key syneresis indices in cheese-making (mainly curd moisture content and yield of whey) over a range of curd cutting programmes and stirring speeds. This study found that light backscatter at 980 nm detected with a large field of view sensor has the biggest effect in linear prediction models developed for curd moisture content and yield of whey. The most accurate and parsimonious models were for predicting the yield of whey, which is in effect a definition of syneresis. Whey production during syneresis could be predicted using light backscatter alone, or using a multivariate model involving fat in milk, implying that online use of NIR sensors on a cheese vat for syneresis prediction may work well in conjunction with online measurement of milk fat. In respect of the experimental variables, this work indicated that the gel cutting programme did not have a clear effect on curd yield.

The significance of this study is that it adds to the various studies which have been undertaken using NIR light backscatter for online monitoring in cheese-making. Taken together, these studies provide information on the most sensitive co-variables which should be included in prediction models. This study also confirms that technology factors, specific to each factory, need to be adequately considered. Thus, the application of these findings in cheese factories involves co-variables which were found to be significant in this and other studies, judiciously selected according to the extent of their variability in the context of each specific factory.

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