Lait 80 (2000) 555–571

Original article

Residual amount of water in a draining curd of Camembert cheese and physicochemical characteristics of the drained curd as modified by the pH at renneting, the casein concentration and the ionic strength of milk

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(Received 14 March 2000; accepted 6 July 2000)

Abstract — Camembert soft cheeses were prepared from cheese milk modified using membrane technologies to obtain different casein contents (27–37 $g \cdot kg^{-1}$) and different ionic strengths estimated from the dilution factor of milk with a lactose solution (0.6 compared to 1.0 in normal milk). Renneting was performed at pH 6.0 or 6.4. The amount of water remaining in the draining curds, expressed as the water to casein ratio, R ($g \cdot g^{-1}$), was calculated from the whey drainage kinetics data. A kinetics of variation of R during drainage was obtained in each of the experimental conditions and was characterised by fitting with a descriptive mathematical model involving 2 relaxation processes and 5 parameters: R (g water$\cdot g^{-1}$ casein) = R1 exp(–t/τ1) + R2 exp(–t/τ2) + R3. The R values at the end of drainage, obtained from the model for t = 1 000 min, fitted well with the water amounts determined in the drained curds by chemical analysis. The values of the equation parameters, obtained by fitting the equation to experimental data, were correlated to the levels of factors which were varied in the experiments. This allowed a mean value for each parameter to be obtained, and values for the effects of factors. The amount of water remaining in drained curd at the end of drainage was dependent on the milk casein content and pH at renneting factors through a negative effect. Prediction of final values of R by calculation from this data was possible. However, the correlation with experimental values was low. In addition, the drained curd composition and rheological properties were determined and the effects of the factors on these curd characteristics were quantified. A positive effect of the pH at renneting on the dry matter and ash contents of drained curd was observed. Incidentally, these factors also modified its rheological properties.

cheese curd / water content / drainage / moisture / rheological property / composition

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Résumé — Étude de l'évolution de la teneur en eau du caillé au cours de l'égouttage, de la composition et des propriétés rhéologiques du caillé égoutté, en relation avec le pH à l'emprésurage, la teneur en caséine du lait et sa force ionique. Des fromages à pâte molle de type Camembert ont été préparés à partir de laits modifiés par des technologies à membranes pour obtenir différentes teneurs en caséine (27–37 $g \cdot kg^{-1}$) et différentes forces ioniques estimées d'après le taux de dilution par une solution de lactose (0,6 par comparaison à 1,0 dans le lait non modifié). L'emprésurage était réalisé à pH 6,0 ou 6,4. La quantité d'eau résiduelle dans le caillé en cours d'égouttage, exprimée par le rapport de la quantité d'eau du caillé à la quantité de caséine du lait initial, R ($g \cdot g^{-1}$) était calculée à partir des données de l'égouttage. Une cinétique de R était obtenue pour chaque expérience d'égouttage. Un ajustement de ces cinétiques à un modèle mathématique a été réalisé en utilisant une équation à 3 étapes, R (g eau$\cdot g^{-1}$ caséine) = $R1 \exp(-t/\tau 1) + R2 \exp(-t/\tau 2) + R3$. R3 était une mesure de l'humidité du caillé en fin d'égouttage. Une valeur pour chacun des 5 paramètres de l'équation a ainsi été obtenue pour chaque expérience. Ces valeurs ont été corrélées avec le niveau des facteurs dans les expériences à l'aide d'un calcul de régression linéaire, ce qui a permis d'obtenir une valeur moyenne pour chaque paramètre et un intervalle de variation de cette moyenne, quantifiant l'effet de chaque facteur. Ces données ont été introduites dans l'équation, qui permettait alors de calculer Rcalc., une valeur prédictive pour R en fonction des niveaux choisis pour les facteurs. Pour les fromages en fin d'égouttage, la valeur de Rcalc. a été calculée (t = 1 000 min) et comparée à la teneur en eau du caillé déterminée expérimentalement par l'analyse. L'effet des facteurs sur la composition du caillé et sur ses propriétés rhéologiques a également été étudié.

caillé / teneur en eau / égouttage / propriété rhéologique / composition

1. INTRODUCTION

Curd formation is an essential step in cheese manufacture because it determines the composition and structure of cheese, although considerable changes in curd structure occur later, during pressing, salting and ripening. The formation of curd is achieved in cheese manufacture by the coagulation of casein through rennet addition followed by curd syneresis and drainage of whey. Many biochemical changes occur in the protein network during curd formation and drainage, such as the differential concentration of casein and fat in curd, the reduction of casein micelle voluminosity, the decrease of water content [43] and the solubilisation of calcium phosphate from the casein micelle due to milk acidification by lactic bacteria. The development of these phenomena depends on the rates of whey expulsion, and concurrently on the acidification process, but also on the composition and treatments applied to milk before and during coagulation. Water removal depends on the endogenous syneresis behaviour, which is closely related to the dynamic character of the casein [41]. Also, any change in character of coagulum as affected by milk composition and by external conditions, like cutting, salting, effective pressure on the curd and the possibility of whey flowing out through pores between curd grains, would modify on the one side, the part of water retained by the curd and, on the other side, its specific properties [38, 40, 43].

The drastic changes in the ability of casein to retain water which occur during the rennet coagulation process, resulting from the release of the hydrophilic caseinomacropeptide from the casein micelle, from the decrease in the native hydration of casein during milk acidification and from the rearrangement of the network of casein particles in curd, have been reported in the literature [32, 42]. The evolution of the amount of water remaining in curd ($g \cdot g^{-1}$ casein) during curd drainage has been also studied and reported, for example by Casiraghi et al. [3]. This parameter can be calculated from the water balance, by determination of the amount of water drained off in whey. This requires a very accurate determination of the weight of whey during drainage, which was generally not achieved, because loss due to evaporation is difficult to reduce. Calculation of this parameter may allow a better understanding of the effect of milk composition on whey expulsion, independently of coagulum composition.

While investigations on drained curd properties are available for several cheese varieties such as Cheddar, Cheshire or Emmental [4, 5, 11, 15, 48], and also soft cheeses [18, 36], no systematic investigation have been done on Camembert drained curd and its composition and rheological properties in relation with the composition of milk or the technological parameters.

Thus, the objective of this work was to characterise the drainage process and the properties of the drained curd during Camembert cheese making. In first part of the work, the water amount remaining in the curd on drainage was calculated from the drainage data already described [8] and the kinetics versus time were drawn. Fitting of the experimental curves to a two relaxation process equation was performed to obtain a descriptive model. The effects of variation in the conditions during coagulation were investigated. Three factors were tested at two levels each: the pH at renneting, the casein level in milk and the ionic strength. In a second part of the work, the drained curd obtained at the end of the drainage process was characterised, its chemical composition and rheological properties were determined and the effect of the factors on these parameters were quantified.

2. MATERIALS AND METHODS

Experimental conditions for curd manufacture and experimentation were described extensively in an earlier report [8]. The general outline was as follows.

2.1. Milk

Milk samples of different casein level and ionic strength were prepared from ultrafiltered skim milk by recombination of retentate, permeate and a 50 $g \cdot L^{-1}$ lactose/water solution. Cream was then added in sufficient amounts to obtain a fat/casein ratio equal to 1 (w/w). Milk samples were heat treated (72 °C, 20 s), using an Actijoule equipment, model 1959 (Actini, Evian-les-Bains, France).

2.2. Curd preparation

Curd was prepared from 2 kg of milk. Milk was inoculated with 1.54 g per 100 L of a mesophilic lactic starter (MM 100 from Texel, Dangé Saint Romain, France), which corresponded to 7×10^6 $CFU \cdot mL^{-1}$ and was maintained at 12 °C during 18 h for prematuration. Milk was then warmed and incubated at 33 °C. When the pH of milk reached 6.0 or 6.4, rennet was added (7.4 mg chymosin per 100 L, from a liquid rennet solution containing 520 $mg \cdot L^{-1}$ chymosin – SBI Gand Gassiot, France). The coagulum (maintained at 33 °C) was cut into 2 cm side cubes, 40 min after rennet addition. Moulding occurred 40 min later and was performed into the drainage pilot device [37]. The shape of the mould was cylindrical (11 cm in diameter, 16.5 cm in height) with a perforated bottom. At this time, measurements of the whey drainage kinetics began, as weights and pH of whey and temperature as a function of time over 18 h. A temperature gradient was applied in the drainage room, from 28 °C to 18 °C over 18 h. The curd was turned over after 1 h and 4 h of drainage.

The yields of drained curd were calculated as the weight of drained curd obtained per kg of milk (no correction was made for the different water contents).

2.3. Analyses

Biochemical analyses were performed on milk (m), whey (w) and drained curd (c). Fractionation of total nitrogen matter in milk was according to the Rowland procedure [35] to separate non-casein nitrogen (NCN) and non-protein nitrogen fractions (NPN). Total nitrogen matter (TN) in milk and nitrogen matter in the NCN and NPN fractions were obtained from N analysis using the Kjeldahl method and the converting factors 6.38, 6.25 and 6.19 respectively for TN, NCN and NPN (values which take into account the corrections for precipitate volume during fractionation, if any). Dry matter (DM) of milk, whey and drained curd was obtained by weighing before and after drying in an oven during 7 h at 102 (±2) °C. Calcium and potassium contents were determined using atomic absorption spectrophotometry [1]. The casein content of milk (CN) was calculated as CN = TNm – NCNm.

The water to casein ratio in the drained curd, Wc/CNc, was determined from chemical analyses, as Wc/CNc ($g \cdot g^{-1}$) = (1 000 – DMc) / (0.989 × TNc). The coefficient 0.989 arose from the following: it was considered that TNc contained 95% of paracasein and that paracasein corresponded to 96% of the casein (CNc) before renneting, due to the caseino-macropeptide release. The water content in the non-fat part of drained curd was calculated as: W/nFc = (1 000 – DMc) / (1 000 – Fatc).

The ionic strength (IS) in the non-modified milk at its native pH was kept as a reference (IS = 1.0). In modified milks, the IS variation (at the native pH) was estimated as proportional to the dilution performed in milk through the addition of the lactose solution. Dilution of milk with lactose solution does not proportionally lower the ionic strength since some salts in the micelle can pass into the soluble phase. This slight modification of ionic strength was neglected with regard to the dilution factor. Moreover, ionic strength pertains to its initial value, i.e. at the initial pH value, as the reduction of the pH from 6.4 to 6.0 led to the dissolution of colloidal salts. The dilution was quantified from the total K contents of the milk. The dilution factor of the modified milk was then calculated as the ratio of total K in modified milk compared to that in the non-modified one.

2.4. Calculation of R

Total water present in milk and the residual water present in the draining curd were calculated from the water contents in milk and in drained whey, as R, expressed in g water$\cdot g^{-1}$ casein of the milk. In milk, it was Rm = (1 000 – DMm) / CN. In the draining curd at the time t, it was Rexp.(t) obtained as the difference between Rm and the amount of water contained in the drained whey at time t : Rexp.(t) = Rm – ((1 000 – DMw) × Weight of whey (t)) / CN. The value DMw, used for this determination, was the average value of DM determined on the total whey obtained, assuming a constant composition of whey during the whole process.

2.5. Characterisation of R kinetics

The experimental kinetic curves of Rexp. were described by two relaxation processes of widely different relaxation times (τ1, τ2), as shown in the following equation derived from the model used in the corresponding whey drainage kinetics [8]:

$$R\ (\text{g water}\cdot \text{g}^{-1}\ \text{CN}) = R1 \times \exp(-t/\tau 1) + R2 \times \exp(-t/\tau 2) + R3 \quad (1)$$

with R, the water to casein ratio in the draining curd at time t. The equation (1) involved 5 parameters: R1, R2 and R3, the water to casein ratio, respectively during the relaxation processes 1 and 2, and at the end of curd drainage, τ1 and τ2, the relaxation times during processes 1 and 2. The initial R in milk (Rm) could be calculated as

Rm = R1 + R2 + R3. During the rapid relaxation process, R tended towards R2 + R3. At the end of the slow relaxation process, i.e. at t >> τ2, R tended towards R3. Thus, R3 represented the fraction of R remaining in curd at the end of drainage and consequently, the final value of R in the drained curd.

2.6. Rheological measurements

Rheological properties of the drained curd were performed with an Instron Universal testing machine (model 4501) using the series IX software (Instron). A compression test using a 100 N load cell and a plate of 60 mm in diameter was performed. Sampling from the drained curds maintained at 12 °C involved the cutting of 8 cylinders (20 mm in diameter and 20 mm height), 5 near the ring and 3 at the centre of each piece of curd. They were equilibrated at this temperature for 1 h prior to compression. Cheese specimens were compressed at 30 mm·min^{-1} until 85% compression was reached. The rheological parameters were the fracture stress, the fracture (Hencky) strain, the work at fracture and Young's modulus. The fracture stress data were corrected for the increase of the cylinder surface during compression assuming a constant volume of the sample.

2.7. Experimental design

Three factors were studied, pH at renneting (ren.pH), casein level in milk (CN) and ionic strength (IS), using an experimental design at two levels: ren.pH = 6.0 and 6.4; CN = 27 g·kg^{-1} (CN 27) and 37 g·kg^{-1} (CN 37); IS = 0.6 (IS 0.6) and 1.0 (IS 1.0). Eight experiments were made to test the factors, four at the lower level and four at the upper level. A 3-time repetition of one experimentation was performed to test the repeatability. Two experiments were repeated for confirmation. The fat/CN ratio in milk was maintained constant, so that the fat level in the milks varied in the experiments. However the effect of the fat level was not tested separately and remained included in the CN effect. The effects of factors on the parameters of equation (1) were derived by means of a linear regression model. First, a linear regression analysis including the overall factors, i.e. factor effects and first order interactions, was performed on each parameter. The effect quantifies the variation of the mean value of the parameter as induced by the variation of a factor level of one coded unit, meaning one half of the total variation of the factor level in the design. Interactions between factors measured the difference in a factor effect according to the factor being tested at the high or low level of a second factor. Second, data were reanalysed with factors at a significance level $P \leq 0.05$. This allowed an average value for each parameter to be obtained, and values for the "effect" corresponding to each significant factor and the determination coefficient of this reduced model.

3. RESULTS AND DISCUSSION

3.1. Residual water during drainage

3.1.1. Kinetics of Rexp. from drainage data

3.1.1.1. Rexp. as a function of time

An example of Rexp. variation as a function of the draining time is given in Figure 1 for four experiments involving different levels of factors. At the time t = 0, the initial value of Rexp. equalled Rm, the value existing in milk. During the first 10 min of the early drainage, Rexp. drastically decreased (Fig. 1a). During further draining, up to 400 min, Rexp. followed an exponential curve. In a late drainage (400–1 000 min), no change in Rexp. was observed. Roughly, a same course of change of Rexp. during drainage was obtained, whatever the

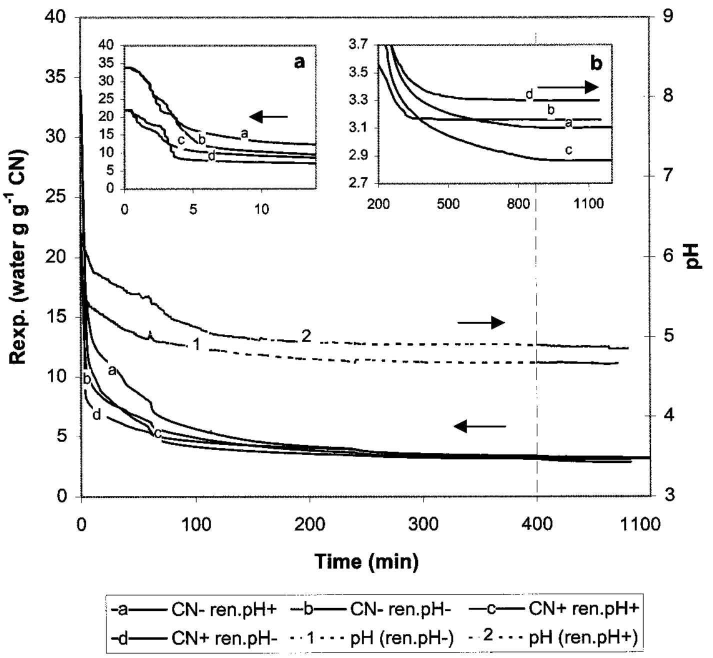

Figure 1. Water content of the draining curd, Rexp. (g·g^{-1} casein), during the course of whey drainage experiments on 4 different milk samples: ren.pH = 6.0 or 6.4 (ren.pH–/+), CN = 27 or 37 g·kg^{-1} (CN–/+), IS = 1.0. Expanded scales in the inset. The arrows indicate the reference axes.

Figure 1. Évolution de la teneur en eau du caillé, Rexp. (g·g^{-1} caséine), pendant l'égouttage de 4 laits dans différentes conditions expérimentales : ren.pH = 6.0 ou 6.4 (ren.pH–/+), CN = 27 ou 37 g·kg^{-1} (CN–/+), IS = 1.0. Expansion d'échelle dans le médaillon. La flèche indique l'axe des ordonnées.

experiments; however, specific differences in relation with the factor levels were apparent (Figs. 1a and 1b). The effects of CN and ren.pH factors are apparent at the different stages. At t = 0, Rm varied according to the CN level of the milk, from 33 g·g^{-1} CN, at the lower CN level (CN = 27), meaning the non-modified milk, to 22 g·g^{-1} CN (CN = 37), as shown in Figure 1a. Moreover, a lower ren.pH led to both a lower Rexp. during the second drainage stage and a higher final Rexp. (Fig. 1b).

3.1.1.2. Rexp. as a function of pH

The values of Rexp. obtained between 10 min to 800 min of drainage in Figure 1, were plotted against the value of the pH of whey at the same time. The relation is shown in Figure 2. Rexp. decreased in a quite linear way as the pH of whey decreased, which seems to suggest that the residual water amount in curd is determined by the value of the pH in the draining curd [3]. An effect of CN was also observed in the 6.0 to 5.5 pH range, the higher CN, the lower Rexp. This CN effect decreased and finally disappeared as the pH decreased from 5.5 to 5.0.

One milk, corresponding to the following factor levels (CN = 27, ren.pH = 6.0, IS = 0.6), behaved very differently from the others, since Rexp. remained much higher at the end of the first draining stage, leading to

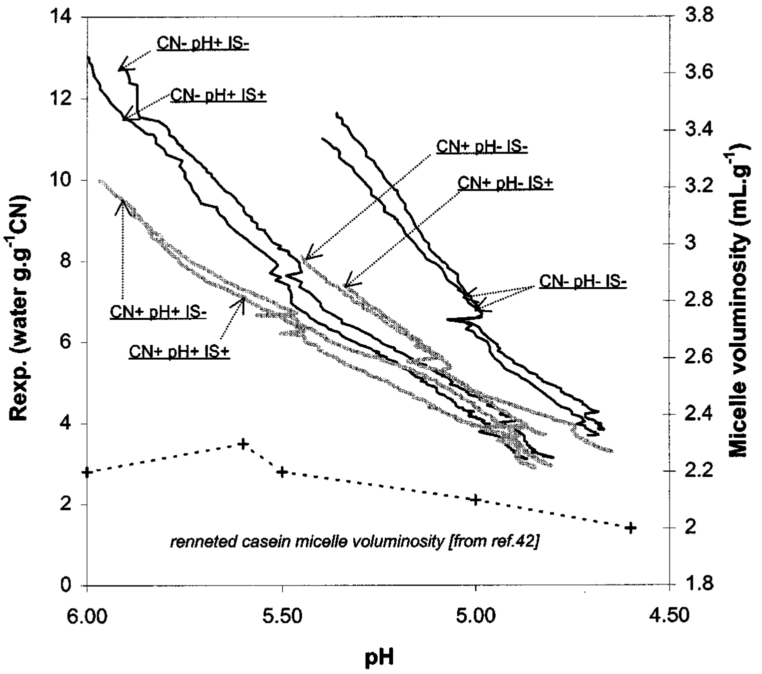

Figure 2. Relation between the water content of the draining curd, Rexp., and the pH of the whey at each time of drainage (from 10 to 800 min) for the 13 assays, ren.pH = 6.0 or 6.4 (pH–/+), CN = 27 or 37 $g \cdot kg^{-1}$ (CN–/+), IS = 0.6 or 1.0 (IS–/+).

Figure 2. Relation entre la teneur en eau du caillé, Rexp. et le pH du lactosérum au cours de l'égouttage (de 10 min à 800 min) pour les 13 essais, ren.pH = 6,0 ou 6,4 (pH–/+), CN = 27 ou 37 $g \cdot kg^{-1}$ (CN–/+), IS = 0,6 ou 1,0 (IS–/+).

a Rexp. value in the 12 $g \cdot g^{-1}$ range at pH 5.5, which compared to a value in the 8 $g \cdot g^{-1}$ range for other milks. The following decrease of the Rexp. value as the pH decreased from 5.5 to 5.0, showed a linear slope at higher values than the other samples. This result was confirmed when repeating the experiment. This highlights the importance of the factor levels and of their interactions on the residual water content of curd during drainage.

3.1.2. Fitting of Rexp. kinetics and effect of factors

Equation (1), involving two exponential relaxation processes and a constant term, was chosen as it led to a better fitting of experimental data when compared to the same without a constant or to a single relaxation process equation.

A fitting of Rexp. experimental kinetic curves to equation (1) was done, which allowed for each experiment individual values to be obtained for the parameters and a corresponding Rfit. equation. An example of such a fitting is given in Figure 3 for the experimental conditions: CN = 27, ren.pH = 6.4, IS = 1.0. Differences between adjusted and experimental data appeared mainly in the early drainage, presumably due to the turning over of the curd mass. The correlation coefficient between Rexp. and Rfit. in this experiment was $r = 0.987$. The average correlation coefficient obtained for the

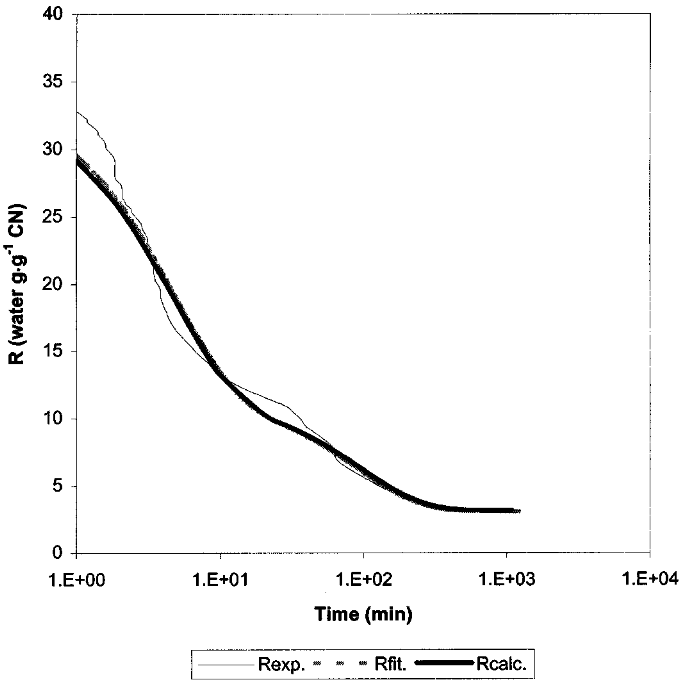

Figure 3. Comparison of R values obtained from experimental data (Rexp.) of a whey drainage kinetics (ren.pH = 6.4, milk CN = 27, IS = 1.0) with: (Rfit.) the fitting to the equation (1), R = R1 exp(–t/τ1) + R2 exp(–t/τ2) + R3 obtained by the least square method, and with (Rcalc.) the curve calculated from equation (2) keeping into account the average effects of factors CN, IS and ren.pH.

Figure 3. Comparaison des valeurs de R obtenues à partir des données expérimentales de l'égouttage (Rexp.) pour la cinétique (ren.pH = 6,4, milk CN = 27, IS = 1,0), avec (Rfit.) l'ajustement par la méthode des moindres carrés à l'aide de l'équation (1), R = R1 exp(–t/τ1) + R2 exp(–t/τ2) + R3, et avec (Rcalc.) les valeurs calculées à l'aide de l'équation (2) prenant en compte les niveaux des facteurs ren.pH, CN et IS.

whole set of experiments was $r = 0.990$ (SD = 0.006).

The effects of the factors on the kinetics parameters were calculated and are given in Table I, as well as the average values of the parameters. The value of Rm, the water in milk, was modified by a negative effect of CN, –6.08 $g \cdot g^{-1}$, which obviously resulted from the ultrafiltration treatment of milk. Significant effects of CN and ren.pH were observed on R1, R2 and R3. Consequently, in equation (1), the parameters had to be replaced by their average value and the effect values from Table I according to the factor levels. For instance, R1 will be calculated as: R1 = 19.00 – 4.68 × CN – 1.31 × ren.pH.

A higher CN level in milk led to a decrease mainly of R1 but also of R2, which was in accordance with the lower Rm value in milk. The incidence on R3 was lower, the –0.13 $g \cdot g^{-1}$ effect value was observed, corresponding to a relative variation of 4% only of the residual water content in the drained curd. The pH at renneting showed two opposite effects on R according to the stage of curd drainage. The effect of ren.pH on R1 was negative, which could be related

Table I. Effect of the renneting pH (ren.pH), the casein level in milk (CN) and the ionic strength (IS) on the water to casein ratio in initial milk (Rm) and on the parameters of the equation (1) giving the residual water in curd during drainage:
Rexp. = R1 × EXP (–t/τ1) + R2 × EXP (–t/τ2) + R3.
Rm, R1, R2, R3 in $g \cdot g^{-1}$ casein, τ1, τ2 in min. (n = 13).

Tableau I. Effet du pH à l'emprésurage (ren.pH), de la teneur en caséine du lait (CN) et de sa force ionique (IS) sur le rapport de la teneur en eau et en caséine dans le lait (Rm) et sur les paramètres de l'équation (1) donnant la teneur résiduelle en eau du caillé en cours d'égouttage :
Rexp. = R1 × exp (–t/τ1) + R2 × exp (–t/τ2) + R3.
Rm, R1, R2, R3 en $g \cdot g^{-1}$ caséine, τ1, τ2 en min. (n = 13).

	Effect of factors				
	ren.pH	CN	IS	Average value	r^2
Rm	ns	–6.08	ns	27.86	0.999
R1	–1.31	–4.68	ns	19.00	0.979
τ1	ns	0.6	–0.8	4.8[(a)]	0.754
R2	1.42	–1.28	ns	5.67	0.924
τ2	–6.6	12.5	ns	112.9[(b)]	0.951
R3	–0.17	–0.15	ns	3.24	0.660

(a) Significant interaction (P = 0.05): CN × IS = –1.1.
(b) Significant interaction (P = 0.05): CN × IS = –6.7; CN × ren.pH = –4.3; ren.pH × IS = –5.4.

(a) Interaction significative (p = 0,05) : CN × IS = –1,1.
(b) Interaction significative (p = 0,05) : CN × IS = –6,7 ; CN × ren.pH = –4,3 ; ren.pH × IS = –5,4.

to the lower syneresis of the coagulum when renneted at a higher pH [46]. The effect on R2 was positive, which could be related to the larger extent of variation of the pH values during the slow drainage relaxation process for the high ren.pH curds (Fig. 2). The effect on R3 was negative, and this could be related to the curd structure itself as formed during the early rennet coagulation of milk which conditioned the casein matrix structure of the curd [23]. The effect of pH observed on R was related to the changes induced by pH on the endogenous syneresis rate, reported as having a maximum in the 5.5–5.2 pH range [33]. No significant effect of milk ionic strength was reported on the residual water content of milk during curd drainage.

The average values of τ1 and τ2 were close to those obtained studying the whey drainage [8], which stands to reason as they relate to the same drainage experiments. The times τ1 and τ2 were both modified by a positive effect of CN, meaning a lower rate of drainage during the whole process at the higher CN level. The ren.pH factor showed a negative effect on τ2 only. IS had an effect of small amplitude on τ1. It was the only effect of IS on the equation parameters, apart from some significant interactions, which could mean that a significant effect of IS of low amplitude could be suspected and could possibly arise and be quantified from a complementary study.

Finally, taking into account the average values of the constants and the effects of factors reported in Table I, the equation allowing the calculation of the water in curd (Rcalc.) will be:

Rcalc. (g water$\cdot g^{-1}$ CN) = (19.00 – 4.68 × CN – 1.31 × ren.pH) × exp(–t/(4.8 + 0.6 × CN – 0.8 × IS – 1.1 × CN × IS)) + (5.67 – 1.28 × CN + 1.42 × ren.pH) × exp(–t/(112.9 + 12.5 × CN – 6.6 × ren.pH – 6.7 × CN × IS – 4.3 CN × ren.pH – 5.4 × IS × ren.pH)) + (3.24 – 0.15 × CN – 0.17 × ren.pH). (2)

These results agree well with those obtained previously on the whey drainage kinetics [8] and are complementary. They are in accordance with the results reported in the literature on the dependence on pH of the solvation and voluminosity of native or renneted casein micelles [7, 42, 44]. Otherwise, the comparison of our results with those of the literature is not easy, as the methods applied are different leading to the determination of different characteristics of casein. For example, the values of solvation and voluminosity of native or renneted micelles, as determined by viscometry, quasi-elastic light scattering or ultracentrifugation techniques could not be compared with the water to casein ratio calculated from curd drainage in the present study. Moreover, in the previous investigations referenced, authors experimented on milk and ren.pH adjustment by addition of an acid solution.

3.1.3. Forecasting of variation in R

The amount of water remaining in a curd at any moment of the drainage could be calculated from equation (2). As an example, Rcalc. was determined at the times t = 20 min and t = 1 000 min (meaning the end of drainage) for the higher and lower values of CN and of ren.pH (Fig. 4). This allowed to visualise the inverse effect of the renneting pH factor on the water amount remaining in curd at two different stages of drainage. The values of Rcalc. for t = 1 000 min, thus corresponding to the final drained curds, allowed a forecast to be made of the water content of the drained curd in relation with the CN and ren.pH levels.

3.1.4. Comparison of experimental and calculated water contents in curd

The water contents of the drained curds could be determined in three different ways: Rexp. (t = 1 000 min), from drainage data; Rcalc. (t = 1 000 min), from calculation using equation (2); or from chemical analysis of the drained curd. Chemical analysis of the drained curd allowed determination of the effective water amount recovered in the curd and, from this data, the water to casein ratio in drained curd (Wc/CNc) was calculated as described in Materials and

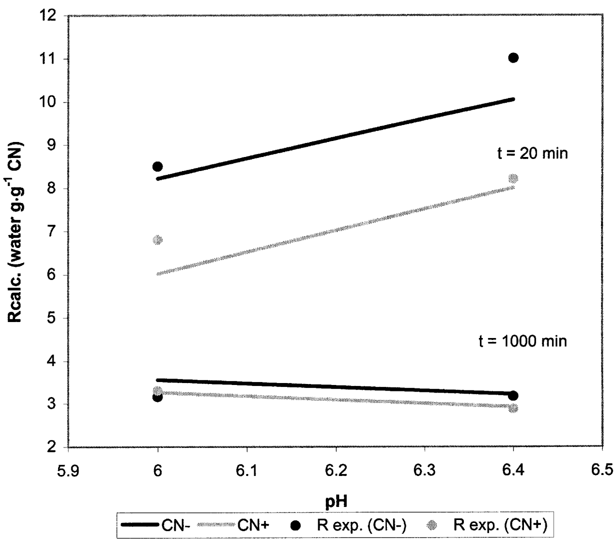

Figure 4. Rcalc. values calculated from equation (2) for the drainage times t = 20 min and t = 1 000 min and the factor levels: CN = 27 or 37 $g \cdot kg^{-1}$ (CN–/+), IS = 1.0, as a function of the renneting pH (pH).

Figure 4. Valeurs de Rcalc. obtenues à partir de l'équation (2) aux temps t = 20 min et t = 1 000 min, pour les niveaux des facteurs CN = 27 ou 37 $g \cdot kg^{-1}$ (CN–/+), IS = 1,0, en fonction du pH à l'emprésurage (pH).

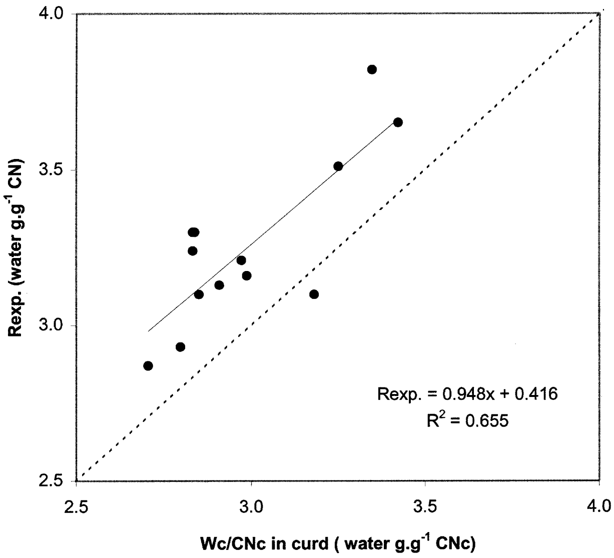

Figure 5. Comparison of the amounts of water in drained curd, (Rexp., t = 1 000 min), obtained from drainage data and (Wc/CNc), determined from chemical analysis of curds.

Figure 5. Comparaison des teneurs en eau du caillé égoutté obtenues d'après les résultats de l'égouttage (Rexp. à t = 1 000 min) et par l'analyse chimique des caillés égouttés (Wc/CNc).

Methods. The values of Wc/CNc were plotted against the values of Rexp. (t = 1 000 min). The relation is shown in Figure 5. The results showed that Wc/CNc was related to Rexp. by a correlation coefficient r = 0.809. However, it was observed that the Rexp. values obtained from drainage data were higher than the Wc/CNc ones by about 10%. The occurrence of some water evaporation from whey during the process is probable. As a matter of fact, the mass balance of water in the experiments showed a water recovery of about 98.83% (SD = 0.38) [8], meaning 11.7 g water loss per kg of milk. This value, related to the casein level in milk, led to a decrease of the final water amount of R of 0.43 to 0.31 g water·g^{-1} casein, according to the casein level in milk being 27 or 37 g·kg^{-1}, respectively. However, the results reported in Figure 6 showed that, apart from the difference between the absolute values themselves, the calculation of Rexp. from the whey drainage data allowed a prediction of the variation in the residual water content that will occur in drained curd as a result of modification of the cheesemilk and/or the coagulation conditions.

To test the validity of the mathematical model of equation (2) in predicting the water remaining in curd, the comparison of Rcalc. (t = 1 000 min) values with the Rexp. (t = 1 000 min) ones was performed (Fig. 6). The correlation obtained was low, r = 0.760, meaning that the predictive values of Rcalc. calculated using equation (2) did not describe accurately the experimental values of Rexp. As a matter of fact, some of the regression coefficients obtained from experiments, reported in Table I, are low (τ1, R3), which indicates that the model in equation (2) can be improved. It is likely that an effect of IS has to be taken into account, as it appeared in some data of the present experiment, but at a significance level slightly lower than was chosen.

3.2. Drained curd characterisation

3.2.1. Yields

Curd yield was 163.1 g·kg^{-1} as an average for the whole experimentation (Tab. II). Significant effects of the CN and ren.pH factors were observed. No significant effect of IS was observed. The CN effect was positive and amounted 28.0 g·kg^{-1} for a variation range of 5 g of casein in milk (according to the experimental design), representing a yield increase of about 5.6 g·kg^{-1} per g·kg^{-1} CN increase in milk. This increase could be roughly distributed as follows: 0.96 g

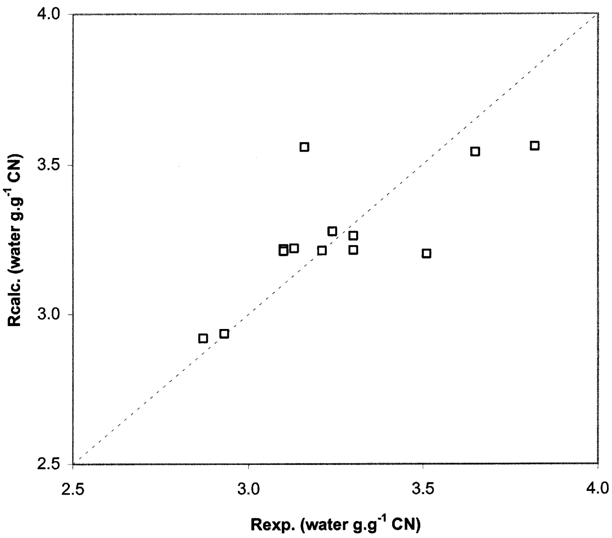

Figure 6. Comparison of the amounts of water in drained curd (Rcalc.), calculated according to equation (2), and (Rexp.), obtained from drainage data. t = 1 000 min.

Figure 6. Comparaison des teneurs en eau du caillé égoutté au temps t = 1 000 min ; (Rexp.), valeurs obtenues à partir des données de l'égouttage ; (Rcalc.), valeurs calculées à l'aide de l'équation (2).

Table II. Effect of the renneting pH (ren.pH), of the casein level in milk (CN) and of the ionic strength (IS) on the composition of drained curd (g·kg^{-1} curd). P = 0.05. DM, dry matter; TNM, total nitrogen matter; total Ca, total calcium; W/nFc: water in non-fat cheese.

Tableau II. Effet du pH à l'emprésurage (ren.pH), de la teneur en caséine du lait (CN) et de sa force ionique (IS) sur la composition du caillé égoutté (g·kg^{-1} caillé). p = 0,05. DM, matière sèche ; TNM, matière azotée totale ; total Ca, calcium total ; W/nFc : eau dans le fromage dégraissé.

	Effect of factors				
	ren.pH	CN	IS	Average value	r^2
Yield of curd g·kg^{-1} milk	–3.2	28.0	ns	163.1	0.977
DM	7.8	ns[(a)]	ns	429.7	0.580
TNM	ns	8.7	ns[(b)]	198.7	0.741
Fat	ns	ns	ns	192.7	–
Ash	1.19	1.08	0.85	14.30	0.928
Total Ca	0.60	ns	0.47	3.00	0.841
pH	0.08	ns	ns	4.64	0.660
Ca/TNM	0.30	ns	0.20	1.53	0.812
Fat/DM	ns	–0.013	ns	0.446	0.417
TNM/DM	ns	0.013	ns	0.463	0.697
W/nFc	ns[(c)]	–0.011	ns[(d)]	0.706	0.704

[(a)] Significant at P = 0.08, CN = 7.4; [(b)] significant at P = 0.08, IS = 3.7; [(c)] significant at P = 0.07, ren.pH = –0.6; [(d)] significant at P = 0.07, IS = –0.6.

[(a)] Effet significatif à p = 0,08, CN = 7,4 ; [(b)] effet significatif à p = 0,08, IS = 3,7 ; [(c)] effet significatif à p = 0,07, ren.pH = –0,6 ; [(d)] effet significatif à p = 0,07, IS = –0,6.

of paracasein, 1 g of fat and about 3.09 g of water (from the R3 value in Tab. I), with the remaining 0.55 g. According to Kosikowski [20], Lawrence et al. [21] and Mistry and Maubois [29], the yield increase using protein standardisation by ultrafiltration is due to reduced losses of fat and caseins in whey and to a better retention of whey proteins into curd. These 0.55 g of yield increase could be ascribed to a better retention of whey proteins. The positive correlation between the yield of curd and the casein plus fat content has often been reported in the literature [6, 11, 21] and various yield formulas have been proposed in relation to the milk protein and fat contents [12].

The effect of ren.pH on the yield was negative and was mainly due to a higher retention of water in curd at low ren.pH, as shown by the positive effect of ren.pH on DM. These data were in accordance with the negative effect of ren.pH observed on R3 (Tab. I). No significant effect of ionic strength on the yield of curd was observed.

3.2.2. Composition

The composition of drained curd was modified by the three factors (Tab. II). CN had a positive effect on the TNM and on the ash contents. A positive effect was observed on DM at $P = 0.08$ only. A higher retention of milk TNM occurred in the curd, as demonstrated by the positive effect of CN on TNM/DM. It was in relation with the higher incorporation in curd of whey proteins, of proteose peptone (for example the 1-25 βCN peptide) and of caseinomacropeptide as a result of milk ultrafiltration [3, 10, 24, 34]. A lower retention of water, thus giving a lower moisture in curd had been already reported in relation to the concentration of milk by ultrafiltration [2, 13, 14, 16]. It has been explained by the formation of a coarser gel network from milk having a higher protein level [13, 14]; it is expected that the coarser gels have a larger permeability and hence undergo a more rapid syneresis during cheese making [41]. The results in Table II showed a positive effect of CN on ash. It could be explained by a renneting step performed on micelles of a higher mineral content. Although the same ren.pH was used, the soluble-colloidal mineral equilibria in milk at a given pH were different in milk concentrated by ultrafiltration, due to the increased concentration of casein [1]. The coagulum obtained had a lower ability to early syneresis, however, after acidification, this coagulum seemed to acquire a higher permeability, as it led to a final drained curd having a higher DM content [28]. No effect of CN was observed on the total Ca content of the drained curd. This may be explained by the large pH decrease occurring during the drainage of the Camembert coagulum, down to an average pH value of 4.64 in the drained curd. Thus, even if more Ca was initially retained in the coagulum, the subsequent acidification led to its loss during drainage. The low amount of Ca remaining in the drained curd did not allow differences to be observed.

A higher pH at renneting led to a drier drained curd with a higher mineral content. The pH and the buffering ability (Ca/TNM) of curd were also increased. Similar results have been reported in the literature [21, 49]. The decrease in DM content with the reduction of ren.pH was mainly due to the increase in the residual water content of curd as judged by the increase in the values of R3 and W/nFc (Tabs. I and II). The demineralisation of curd in relation to the decrease in ren.pH would be due to a conversion of calcium and phosphate from colloidal into dissolved forms, which were thus removed from the curd network during whey drainage [17, 19, 22]. According to Lucey and Fox [26], the pH effect on the demineralisation of cheese will be greater when acid is produced prior to whey drainage to obtain a lower pH at renneting (as in the case for Camembert and Cheddar cheeses), than when acid is produced mainly after moulding, because then a smaller proportion of whey is removed after the initial drainage. Despite these differences in curd composition in relation to the change in ren.pH, no significant effects were detected in total nitrogen fraction and fat recovery of the curd. This points to the effect of ren.pH on curd composition being due to differences in the drainage process. At the higher ren.pH, less water was retained on the curd due to a greater drainage, as reported in the previous paper [8].

The variation of milk ionic strength had no significant effect on the yield and on the water retention of curd (R3). However, a positive effect was observed on ash, total calcium and protein recovery. It was related to the dilution of milk by addition of the lactose solution that decreased the non-protein content of milk. A negative effect of IS was observed on the water content in the non-fat curd. This effect was not significant for DM, presumably because the variations of fat masked the phenomenon. These results are in accordance with those obtained previously [9], as the effect of ionic strength observed on the whey drainage kinetics was so tiny that it was no more significant after modelling, in spite of small significant effects observed on the experimental data themselves [8].

The fat content of drained curds, 192.7 $g \cdot kg^{-1}$ as an average, was modified by none of the three factors and fat recovery was 96.9% whatever the conditions. The fat/DM ratio, amounting to 0.446, was modified only by a negative effect of CN, presumably in relation with the concomitant positive CN effect observed on TNM [25, 30]. Thus, the addition of fat, made at a constant fat/CN ratio = 1, allowed drained curds to be obtained having quite the same composition regarding fat, and this allowed the effects observed for the CN factor, which, in the design, include both casein and fat variations, to be attributed mainly to the casein content in milk, apart from the yield of curd.

To resume, the results showed that the composition of the drained curd varied according to the factor levels. A drier curd with a higher mineral content was obtained from a milk having a higher casein and fat content and renneted at a higher pH.

3.2.3. Rheological properties

The rheological properties of drained curd varied according to the changes in milk and curd composition (Tab. III). The consequences of varying process parameters such as casein concentration or pH at renneting on rheological properties of curd are probably mainly due to the effects of these parameters on curd composition or pH in curd. Due to the low number of experiments, it is anyway impossible to test the effect of each parameter through a model including curd composition and pH.

Table III. Effect of the renneting pH (ren.pH), of the casein level in milk (CN) and of the ionic strength (IS) on the rheological properties of the drained curd. $P = 0.05$.

Tableau III. Effet du pH à l'emprésurage (ren.pH), de la teneur en caséine du lait (CN) et de sa force ionique (IS) sur les propriétés rhéologiques du caillé égoutté. $p = 0,05$.

	Effect of factors						
	ren.pH	CN	IS	CN × ren.pH	CN ×IS	Average value	r^2
Young modulus (kPa)	ns	30.5	ns[(a)]	ns	–10.5	152.7	0.874
Fracture strain	0.018	–0.025	ns	0.021	ns	0.379	0.586
Fracture stress (kPa)	2.55	4.76	ns	1.96	ns	33.80	0.824

[(a)] Significant at $P = 0.07$, IS = –8.3.

[(a)] Effet significatif à $p = 0,07$, IS = –8,3.

An increase in CN led to an increase in curd rigidity (Young modulus) and in curd firmness (fracture stress). The fracture strain was lowered at higher CN, meaning the formation of a shorter curd. Similar results were already reported in relation with the increase in casein and fat contents of milk [5, 27]. The increase in the solid character of curd at higher CN could be due, according to Prentice et al. [31], to the higher total solid (DM) and the higher total protein (TNM) contents of curd (Tab. II). The proteins and particularly casein are effectively recognised to give a solid appearance to curd as they promote a continuous elastic framework [46, 47]. The decrease both in the residual water content of curd, R3 (Tab. I), and in the water in non-fat curd, W/nFc (Tab. II), could be also implied. As a matter of fact, it is reported in the literature that water could act as a lubricant between casein particles and fat [27, 36], thus a decrease in the water content of curd could reduce the ability of caseins to move between casein micelles and fat, so that a firmer curd which is more resistant to any deformation was produced [22, 46].

The ren.pH had an incidence on most of the rheological properties of curd. This was presumably in relation with the mineral content of the micelle at renneting. Colloidal calcium and phosphate form crosslinks within the casein network at coagulation [22]. In this way, minerals play an important role in curd rigidity and firmness, which depends on the number and type of bonds in the protein network [47]. It may be assumed that a coarser curd will be produced, having a more cross-linked structure [17] and thicker strands [13, 14] within the network, which are more resistant to fracture. The decrease in curd firmness and curd longness with decreasing pH at renneting was probably due to the mineral solubilisation of curd, leading to a reduction of cross-links and also to the swelling of casein particles [39], which increased the water content of curd at lower pH at renneting.

The positive interaction between ren.pH and CN on the firmness and longness of curd outlined the importance of curd minerals and hence, of curd structure during the drainage process. A higher mineral content of casein particles would lead to the establishment of a higher number of high strength interactions between caseins.

No significant effect of ionic strength was observed on the rheological properties of drained curd in the present study. This could be related to the lack of effect of IS on curd composition.

ACKNOWLEDGEMENTS

We are indebted to J. Fauquant, N. Hamard-Leconte and J.M. Samson for performing milk preparation in the laboratory pilot hall. The study has been carried out with financial support from the Commission of the European Communities, Agriculture and Fisheries (FAIR) specific RTD programme, CT 96-1056, Cheese Texture Improvement through Rationalisation and Optimisation of Structure. It does not necessarily reflect its views and in no way anticipates the Commission’s future policy in this area.

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