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Effect of pH and sodium chloride on the viscosity of skim milk retentates

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Summary

Skim milk retentates of different protein and NaCl contents were inoculated with 1-10 % of mixed starter of *L. bulgaricus* and *S. thermophilus* (1 : 1) and incubated at 37°C. The relative viscosities of these retentates were followed as a function of changes in pH and added NaCl.

The relative viscosity of retentate was affected by its protein content, NaCl added and pH. The relative viscosity of retentate increased with the increase in its protein content (4 to 17.3 %) but the relation was non-linear.

Addition of 1 % NaCl to retentate increased its relative viscosity, that decreased on standing or addition of higher percentage of NaCl (2 and 3 %). The relative viscosity of retentate gradually decreased with the decrease in pH, followed by reincrease at pH less than 6.00. The pH at which the relative viscosity began to reincrease was dependant on the percentage of added NaCl.

Key words : Relative viscosity - Retentate - Skim milk - Sodium chloride - Starter - pH - Protein content.

Résumé

Influence du pH et du chlorure de sodium sur la viscosité de rétentats de lait écrémé

Des rétentats de lait écrémé de différentes teneurs en protéines et NaCl ont été ensemencés avec 1-10 % de levains mixtes de *L. bulgaricus* et *S. thermophilus* (1 : 1) et incubés à 37 °C.

Les viscosités relatives de ces rétentats ont été suivies en fonction des variations du PH et du NaCl ajouté.

La viscosité relative du rétentat est influencée par sa teneur en protéines, l'addition de NaCl et le pH. Elle augmente avec l'augmentation de la teneur en protéines (4 à 17,3 %), mais la relation est non linéaire.

L'addition de 1 % de NaCl au rétentat augmente dans un premier temps sa viscosité relative, mais celle-ci diminue ensuite au cours de la conservation. L'ajout d'un pourcentage plus élevé de NaCl (2 et 3 %) décroît également la viscosité relative.

Cette caractéristique physique du rétentat diminue également graduellement avec la baisse du pH mais réaugmente à pH inférieur à 6,0. Le pH auquel la viscosité relative commence à croître est dépendant du pourcentage de NaCl ajouté.

Mots clés : Viscosité relative - Rétentat - Lait écrémé - Chlorure de sodium - Levain - pH - Teneur en protéines.

Introduction

Viscosity is one of the important characteristics of fluids which determine their mass and heat transfer properties. The viscosity of milk has received attention by several investigators and adequate information on this subject has been reviewed (JENNESS *et al.*, 1974).

The viscosity of concentrated milks and ultrafiltration retentates has received much less attention than milk. The fast increase in the use of ultrafiltration in dairy industry makes it necessary to study the properties of the resulting fluids particularly UF retentates. The viscosity of UF retentates and factors affecting it are of great importance as the viscosity of retentates is rapidly increased with the increase in protein content (CULIOLI *et al.*, 1974). Therefore, the viscosity of retentate partially determine the concentration factor that can be reached during ultrafiltration processing, and the behaviour of retentates during processing. The effect of protein content and temperature on the viscosity of retentates have been studied (CULIOLI *et al.*, 1974 ; PIERRE *et al.*, 1978).

Determination of viscosity of retentates of high protein content is tedious and need proper selection of methodology. However, the relative viscosity offers a more simple tool for comparative studies and changes in viscosity as affected by different factors.

The present paper deals with the relative viscosity of UF retentates as affected by protein content, addition of NaCl and changes in pH as a result of growth of starter bacteria.

I. Materials and methods

A. *Preparation of UF retentates*

Skim milk powder (Dairy Crest, UK) was reconstituted in deionized water at the rate of 10 % and ultrafiltered at 45 °C and 3.6 and 0.6 bar inlet and outlet pressure respectively using DDS Lab 20 UF unit and GR61P membrane. The protein content of the retentate was found to be 17.3 %. The retentate was diluted with suitable quantities of permeate to obtain retentates with 12, 8, and 4 % protein respectively. NaCl was added to the prepared retentates at the rate of 0, 0.5, 1.0, 2.0 and 3.0 % respectively directly before inoculation with starter.

B. *Inoculation with starter*

Pure strains of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* were obtained from Ch. Hansen A/S (Copenhagen, Denmark) and they were propagated in sterilized skim milk. Retentates were inoculated with a mixed starter of the two organisms (1 : 1) at the rate of 0, 1, 2, 3, 5 and 10 % respectively and incubated at 37 °C for 8 h or until curdled. The relative viscosity and pH of these retentates were followed at 2 h intervals.

C. *Determination of relative viscosity*

The viscosity of retentate was determined relative to that of the corresponding permeate using Hoppler viscosimeter. NaCl was added to permeate at the rate of 0, 0.5, 1, 2 and 3 % respectively. The time taken by the falling ball in retentate was divided by that taken in permeate containing the same NaCl content. Measurements were carried out at 37 °C. The pH was determined in the retentates using a pH meter with combined electrode.

II. Results and discussion

Figure 1 shows the changes in pH of skim milk retentates as a function of percentage of starter and NaCl added, and incubation time. These results indicate that the pH of retentate decreased with the increase in percentage of starter added. A linear relation was evident between the pH and the log percentage of starter

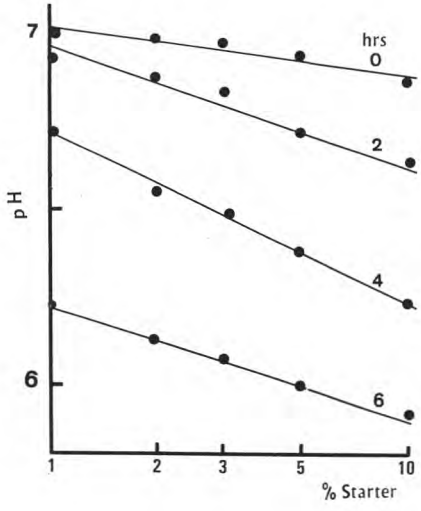

Fig. 1

pH changes as a function of the percentage of added starter and incubation time for retentate containing 12 % protein.

Changements de pH en fonction du pourcentage de levain ajouté et du temps d'incubation d'un rétentat à 12 % de protéines.

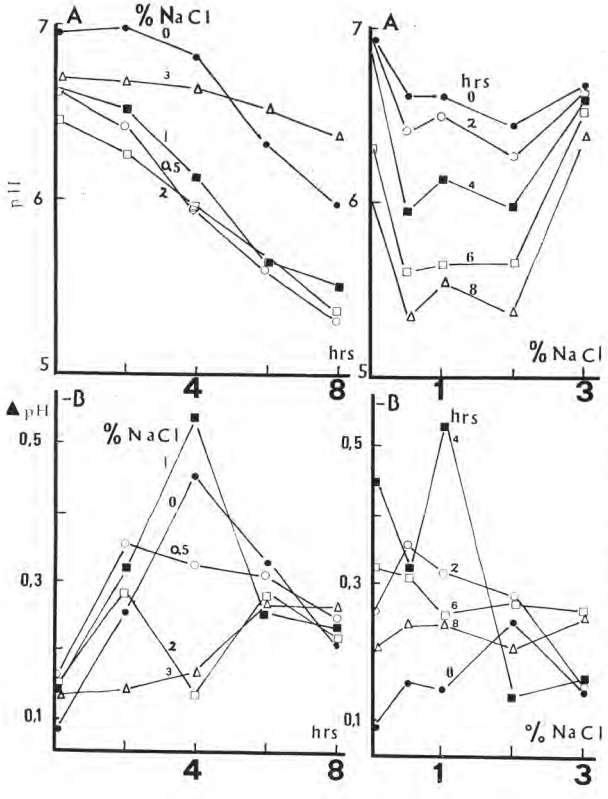

Fig. 2

Coefficient A and B from the equation (1) as a function of NaCl and incubation time for the retentates containing 17.3 % protein : ● 0 time, 0 NaCl ; ○ 2 hr, 0.5 % NaCl ; ■ 4 hr, 1 % NaCl ; □ 6 hr, 2 % NaCl ; △ 8 hr, 3 % NaCl.

Coefficients A et B de l'équation (1) en fonction du NaCl ajouté et du temps d'incubation pour des rétentats contenant 17,3 % de protéines : ● sans ajout de NaCl, sans incubation ; ○ incubation 2 h, 0,5 % NaCl ; ■ incubation 4 h, 1 % NaCl ; □ incubation 6 h, 2 % NaCl ; △ incubation 8 h, 3 % NaCl.

added to retentate containing 12 % protein (fig. 1). The relation between the pH and the log percentage of starter added was expressed using the equation

$$pH = A + B \log S \qquad (1)$$

where

A = pH value calculated by the least square method for 1 % starter,

B = pH decrease when % of starter added is increased 10 times,

S = percentage of starter added.

Figures 2 and 3 show the coefficients A and B for pH changes in retentates containing 12 and 17.3 % protein as a function of percentage of NaCl added and incubation time. These results indicate :

— Within the limits of standard deviation, there was no difference between the retentates containing 12 and 17.3 % protein as to their pH changes in relation to percentage of starter and NaCl added and incubation time. This suggest that the starter bacteria grow equally in 12 and 17.3 % protein retentates under the mentioned conditions.

— There is a steady decrease in the A value with the incubation time while the B value increased to a maximum at 4 h and slightly decreased thereafter.

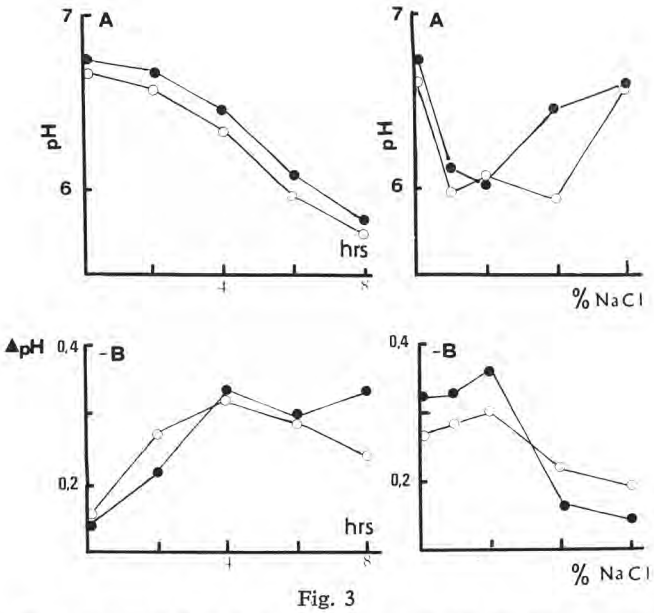

Fig. 3

Average values of coefficients A and B from equation (1) as function of percentage of NaCl and incubation time for retentates containing 12 (○) *and 17.3 %* (●) *protein.*

Valeurs moyennes des coefficients A et B de l'équation (1) en fonction du pourcentage de NaCl ajouté et du temps d'incubation pour des rétentats contenant 12 % (○) *et 17,3 %* (●) *de protéines.*

— The coefficient A decrease on addition of small percentage of NaCl and reincrease with further addition of NaCl. This decrease could be explained on exchange of Na^+ with H^+ in NH_3^+ group on the protein moiety setting free proton and decreasing pH (LING, 1963). Further increase in NaCl added would retard bacterial growth and acid production. Addition of NaCl up to 1 % had no significant effect on the coefficient B, but it decreased with further increase in NaCl added indicating retarded bacterial growth and acid development.

The changes in the viscosity of retentate as affected by addition of NaCl and growth of starter bacteria were followed by determining the relative viscosity to that of permeate. NaCl was added to permeate at the same percentage added to the tested retentate in order to correct for the direct contribution of added NaCl to viscosity. Addition of NaCl slightly increased the viscosity of permeate and linear relation was found between the amount of added NaCl and viscosity of permeate.

Results illustrated in figure 4 clearly indicate that the relative viscosity of retentate was greatly affected by their protein content and that this relation was not linear. The same was apparent between the relative viscosity of retentate containing 17.3 % protein and the percentage of starter added. Addition of 1 and 10 % starter (3.5 % protein) inoculum would lower the protein content of 17.3 % protein retentate by 0.14 and 1.4 % respectively and in turn its relative

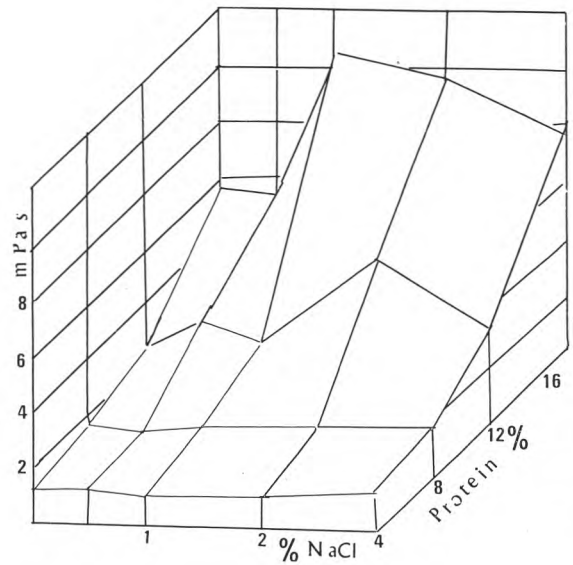

Fig. 4

Viscosity of milk retentates as a function of percentage of salt added and protein concentration.

Viscosité des rétentats de lait en fonction du pourcentage de NaCl ajouté et de la concentration en protéines.

viscosity. However, this relation was found linear on the basis of log viscosity and log starter percentage added (fig. 5). The same relation was also obtained in retentate with low protein content (12 %) (fig. 5). The relation between the relative viscosity and the percentage of stater added was expressed using the equation:

$$\text{Log relative viscosity} = A + B \log S \qquad (2)$$

where

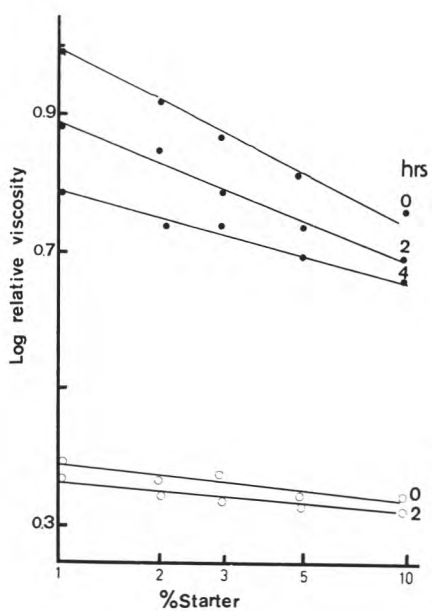

Fig. 5

Changes in relative viscosity of retentates containing 12 (○) and 17.3 % (●) protein as a function of added starter and incubation time.

Changements de viscosité relative des rétentats contenant 12 % (○) et 17,3 % (●) de protéines en fonction du levain ajouté et du temps d'incubation.

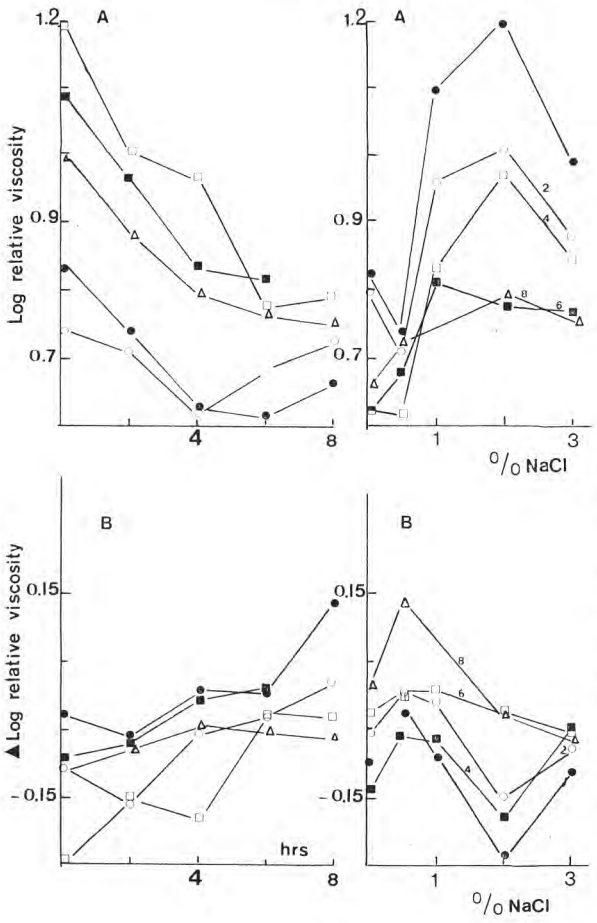

Fig. 6

Coefficients A and B from the equation (2) as a function of NaCl and incubation time for retentates containing 17.3 % protein : ● 0 time, 0 NaCl ; ○ 2 hr, 0.5 % NaCl ; ■ 4 hr, 1 % NaCl ; □ 6 hr, 2 % NaCl ; △ 8 hr, 3 % NaCl.

Coefficients A et B de l'équation (2) en fonction du pourcentage de NaCl ajouté et du temps d'incubation pour des rétentats contenant 17,3 % de protéines : ● sans incubation, sans ajout de NaCl ; ○ 2 h d'incubation, 0,5 % NaCl ; ■ 4 h d'incubation, 1 % NaCl ; □ 6 h d'incubation, 2 % NaCl ; △ 8 h d'incubation, 3 % NaCl.

A = log relative viscosity calculated by the least square method for 1 % starter added,

B = decrease in log relative viscosity with % starter added is increased 10 times,

S = percentage of starter added.

Figures 6 and 7 show changes in coefficient A and B for log relative viscosity in retentate containing 17.3 and 17,3 and 12 % protein respectively as a function of percentage of NaCl added and in incubation time. The results indicate :

— The changes in relative viscosity in retentates containing 12 and 17.3 % protein follow the same trend of variation as affected by NaCl added and incubation time. A steady decrease in the A value with incubation time to a minimum at 6 h and reincreased thereafter. The B value increased steadly during incubation.

— The coefficient A steadly increased on addition of increased quantities of NaCl in 12 % protein retentates, while in 17.3 % protein retentate, coeffi-

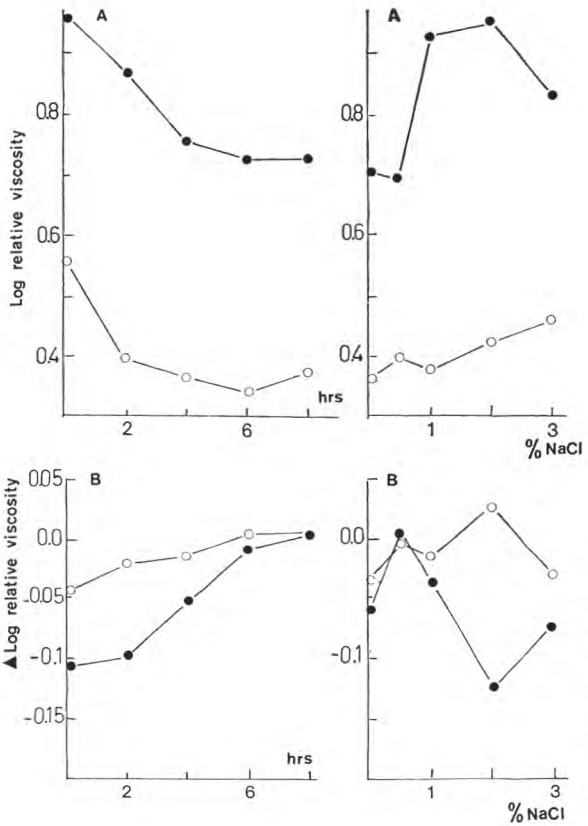

Fig. 7

Average values of coefficient A and B from equation (2) as functions of percentage of NaCl and incubation time for retentates containing 12 (○) and 17.3 % (●).

Valeurs moyennes des coefficients A et B de l'équation (2) en fonction du pourcentage de NaCl ajouté et du temps d'incubation pour des rétentats contenant 12 % (○) et 17,3 % (●) de protéines.

cient A increased with the addition of NaCl up to 2 % and then redecreased slightly. Addition of 0.5 % NaCl slightly increased coefficient B, followed by decrease on further addition of NaCl (2 % NaCl in 17.3 % protein retentate) and reincreased at 3 % NaCl. The changes in B value in 12 % protein retentates were slightly different from 17.3 % protein retentate.

The effect of NaCl on the relative viscosity of retentates can be explained on the basis that the action of NaCl on the colloidal system of milk is a slow reaction that need time to reach equilibrium. The obtained results suggest that NaCl may have the following effects on the colloidal system of milk :

1. Immediately after the addition of NaCl, Na^+ would exchange with the cations on the surface of the casein micelles without any disruption in the casein micelle structure. The formed Na-Ca caseinate would increase the hydration of the protein particles and the relative viscosity of retentate.

2. Further contact of NaCl with the colloidal system would exchange more Ca^{++} for Na^+. At this stage, removing Ca^{++} from casein micelles would decrease its micellar size as Ca^{++}acts as the interacting nodes in the colloidal system of milk. This would reduce the relative viscosity of retentate. Saito and Hirose (1972) found that the addition of 1 M NaCl to casein micelles, release

part of the Ca^{++} and solubilize part of the protein which support the present finding.

The relative viscosity of retentate with added NaCl was probably controlled by the two mentioned reactions and the relative viscosity would depend on the ratio of added salt/protein in retentates and the time lapsed from the addition of NaCl.

The growth of starter bacteria in the retentate of different protein contents was accompanied with a decrease in pH and relative viscosity. This can be attributed to reduction in hydration of casein micelles (Sood *et al.*, 1979). However, this does not exclude the possible proteolysis in casein by starter enzymes (Lawreance and Thomas, 1979) reducing the molecular size of casein micelles and consequently reducing their relative viscosities. At pH less than 6.00, the relative viscosity of retentates reincreased again due to possible aggregation of altered casein through hydrophobic interactions forming large aggregates. Albertsen *et al.* (1983) showed that the viscosity of UF retentate reach a minimum at pH 5.2 and then reincreased. In the present study retentates were prepared from reconstituted skim milk. The presence of partially denatured whey proteins would modify the functional properties of retentates rendering them more sensitive to changes in pH. The pH at which the relative viscosity started to reincrease was also affected by the percentage of added NaCl. The higher the NaCl added, the lower the pH at which the relative viscosity of retentate began to reincrease. This can be explained on the basis that the increased ionic strength of the medium with added NaCl would increase the repulsive forces and delay aggregation of the altered casein micelles.

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