Agronomy Research **12**(3), 787–792, 2014

Effect of cooling rates and low crystallization temperatures on morphology of lactose crystals obtained from Ricotta cheese whey

A. Pisponen*, S. Pajumägi, H. Mootse, A. Sats, V. Poikalainen and A. Karus

Department of Food Science and Technology, Estonian University of Life Sciences, Kreutzwaldi 56/5, EE51014 Tartu, Estonia; *Correspondence: anna.pisponen@emu.ee

Abstract. In the traditional process of lactose production from cheese whey crystallization temperature is reached by slow cooling. Lactose morphology obtained in this way has been well investigated. The objective of this work was to study morphological parameters of lactose crystals during crystallization at low temperatures, using rapid and extra rapid cooling. Ricotta whey was chosen for analysis because this raw material has been poorly investigated as a source of lactose production. Three temperatures (2, 6 and 12°C) were used for crystallization of lactose. Rapid (0.5°C min^{-1}) and extra rapid cooling (3°C min^{-1}) rates were used to achieve these temperatures. Dimensions of crystals were examined with optical stereo-, and scanning electron microscopes. Pure lactose solution was used as a reference during the study. The results of the study showed the impact of some Ricotta whey components on the crystals morphology and size. These components reduce crystals size, and linked with low crystallization temperature, modify the shape of crystals faces. Extra rapid cooling of Ricotta whey contributes to the growth of needle-like crystals more than the rapid one. In a pure lactose solution no needle-shaped crystals were observed.

Key words: lactose, Ricotta cheese whey, crystallization, cooling rate.

INTRODUCTION

Industrial crystallization of lactose from cheese whey is a slow process based on cooling of deproteinated and concentrated whey. First of all, a concentrate is cooled rapidly to 30°C. After that it is seeded and cooled rather slowly, at about 1 to 3°C h^{-1}. Crystallization temperature of 20°C is normally recommended. (Tan, 2010) As early as in 1930s, Whritter & Could (1931) proposed such scheme for lactose production because they found that lactose crystallization was faster at 30°C than at any lower temperature. Crystallization of lactose at low temperatures has been well studied over many decades in connection with ice-cream investigations (Nickerson, 1954, 1956; Livney et al., 1995). The phenomenon of cooling rate in general has also been investigated and used for manufacturing of lactose (Paterson, 2010). No information was found in the literature concerning the effects of low crystallization temperatures and the cooling rate on the morphological parameters of lactose crystals. The objective of this work was to study morphological parameters of lactose crystals during crystallization at low temperatures, using rapid and extra rapid cooling.

MATERIALS AND METHODS

Ricotta cheese whey was used for this study because of additional deproteinization process carried out during Ricotta production. It was provided by a local cheese factory, OÜ Põltsamaa Juustutööstus, Estonia. The lactose, fat and protein contents were measured at the Milk Analysis Laboratory of the Estonian Animal Recording Centre using an automated infrared milk analyzer (CombiFoss FT+, Foss Electric, Denmark). The ash content was estimated using the International IDF Standard 42B : 1990; pH was measured by the METTLER TOLEDO pH-meter (Mettler-Toledo International Inc., Switzerland). The dry matter content was measured using METTLER TOLEDO RH83 moisture analyzer (Mettler-Toledo International Inc., Switzerland).

Ricotta whey was heated to 90°C and then 0.4% w/w of lactic acid (Sigma-Aldrich Chemie GmbH, Germany) was added for precipitation of protein residues, which were removed by filtering through common paper filter. Concentration of Ricotta cheese whey was carried out by water evaporation at low temperature (60°C) during approximately 3.5 h. Concentration factor of 10 was achieved by this treatment. It corresponds to total solids concentration of ca 50% w/w (Pisponen et al., 2013). Concentrated Ricotta whey was poured into test tubes, by 10 ml into each one. Crystallization was carried out at the temperatures of 2, 6 and 12°C during 24 hours without agitation in an incubator Panasonic MIR-154-PE (Japan). Rapid (0.5°C min^{-1}) and extra rapid cooling (3°C min^{-1}) rates were used to achieve these temperatures. Extra rapid cooling was carried out using ultra low temperature freezer Panasonic MDF-C8V1 (Japan). The pure lactose solution with concentration 50% w/w was used as reference solution and was subjected to the same stages, in order to study the impact of impurities on crystallization process. Thus, there were two investigated parameters for each sample: cooling rate and temperature of crystallization. Crystals were washed with 10% w/w spirit solution and distilled water to clean crystals from molasses.

The shape and the size of crystals were examined using a microscope Nikon SMZ 1000 (Nikon Corporation, Japan), equipped with the digital camera Nikon DS-U2/L2 USB (Nikon Corporation, Japan). The height of crystals was estimated using the software NIS-Elements D 3.1 (Nikon Corporation, Japan). Used measurement technique has been described in our earlier study in Pisponen et al. (2013). At least 40 crystals were examined for each data set. In order to avoid errors, only single, clearly distinguishable crystals were inspected. A scanning electron microscope Leo 1430VP (LEO Electron Microscopy Ltd, England) was used for studying the surface of crystals.

RESULTS AND DISCUSSION

The content of dry matter, fat, protein and lactose in Ricotta cheese whey are given in Table 1. Five measurements were considered for each data set. Data values obtained in this study were lower than those of the normal cheese whey (Jelen, 2003).

The average values of crystals heights are shown in Table 2. Crystals obtained from pure lactose solution in most cases were larger than crystals obtained from Ricotta cheese whey. This difference can be explained by presence of protein residues in Ricotta cheese whey. Modler & Lefkovitch (1986) and Miumoni et al. (2005)

demonstrated in their research with cheese whey that protein lowered the final size of crystals. Minerals, contained in Ricotta cheese whey, can also lower the final size of crystals. Guu & Zall (1991) found, that potassium, sodium and calcium can retard or inhibit lactose crystallization from cheese whey. General appearance of crystals is shown in Fig. 1.

Table 1. Content of dry matter, fat, protein, lactose, ash content (%w/w) and pH in native Ricotta cheese whey

	Dry matter	Fat	Protein	Lactose	pH	Ash content
Average	4.9	0.16	0.44	3.85	5.67	0.51
Standard deviation	0.23	0.01	0.02	0.07	0.08	0.08

Table 2. The average, ± standard deviation, minimum and maximum values (within brackets) of crystals height (μm) obtained from different solutions under different crystallization temperatures and cooling rates

Temperature,°C	2		6		12	
Rate of cooling	Rapid	Extra rapid	Rapid	Extra rapid	Rapid	Extra rapid
Ricotta cheese whey	155 ± 20 (128–196)	78 ± 26 (41–127)	182 ± 76 (93–388)	133 ± 73 (39–384)	179 ± 59 (80–351)	123 ± 53 (62–337)
Pure lactose solution	166 ± 77 (77–450)	128 ± 52 (39–288)	165 ± 66 (46–302)	192 ± 72 (94–385)	158 ± 68 (55–387)	220 ± 65 (111–372)

Rapid cooling does not affect the size of crystals obtained from pure lactose solution; they remained almost identical at different crystallization temperatures. When using extra rapid cooling, the difference in crystals sizes became obvious. The height of crystals, obtained from pure lactose solution, increased along with rising of crystallization temperatures. There was no such a clear dependence in size of crystals, obtained from Ricotta cheese whey; the shortest crystals appeared at 2°C and the highest at 6°C. Difference in sizes between crystals, obtained at 6 and 12°C, was considerably smaller than between 2 and 6°C. This indicates to a certain optimum between 2 and 12°C. A large amount of needle-shape crystals was obtained at 6 and 12°C during crystallization from Ricotta cheese whey at extra rapid cooling (Fig. 1). Needle-shaped crystals are formed when the super saturation of material in solution is very high (Hartel, 2001). Extra rapid cooling increases the content of crystalline material in solution and promotes the appearance of a host in crystallization centers (Paterson, 2010). In our experiment, extra rapid cooling could promote occurrence of even more crystallization centers, which lead to appearance of extra small crystals. There were no needle-shaped crystals observed in a pure lactose solution (Fig 1), which leads to the conclusion that impurities have bigger impact on the crystal form than crystallization conditions. Crystals obtained in the research were generally smaller than crystals obtained in our earlier study at the same concentration factor, but at a slower cooling rate (Pisponen et al., 2013).

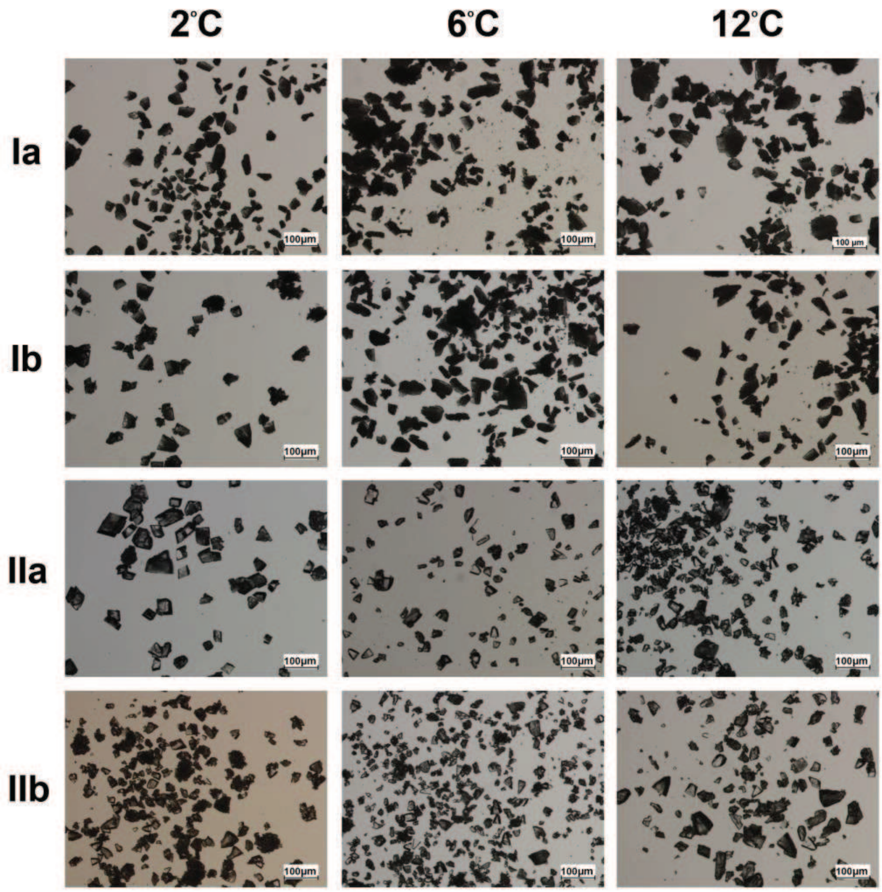

Figure 1. Lactose crystals forms depending on crystallization temperature and cooling rate: solution: I–Ricotta cheese whey II–pure lactose solution: a–rapid cooling and b–extra rapid cooling.

The SEM photos show that faces of some crystals, grown in Ricotta cheese whey, were modified (Fig. 2 b). According to Hartel`s (2001) theory crystals grow imperfectly, if their growth speed is too rapid. Molecules do not have time to attach the right place on the crystal face (in the crystal lattice). They rapidly incorporate at random position, making crystal surface rough. In our case crystals were growing very rapidly due to a sharp drop in temperature. But crystals, grown at the same conditions in pure lactose solution, had smooth faces (Fig. 2 a).

It can be supposed, that not only growth speed can affect the shape of crystals. Macromolecules from Ricotta cheese whey such as proteins may adsorb on the crystal surface and influence the manner by which molecules attach to the crystals lattice (Hartel & Shastry, 1991). Riboflavin can also adsorb on the growing crystal and modify its shape (Holsinger, 1988). Lifran et al. (2007) found, that riboflavin makes the tip of the crystal irregular, which can be observed on crystals, obtained from Ricotta cheese whey (Fig. 2 b).

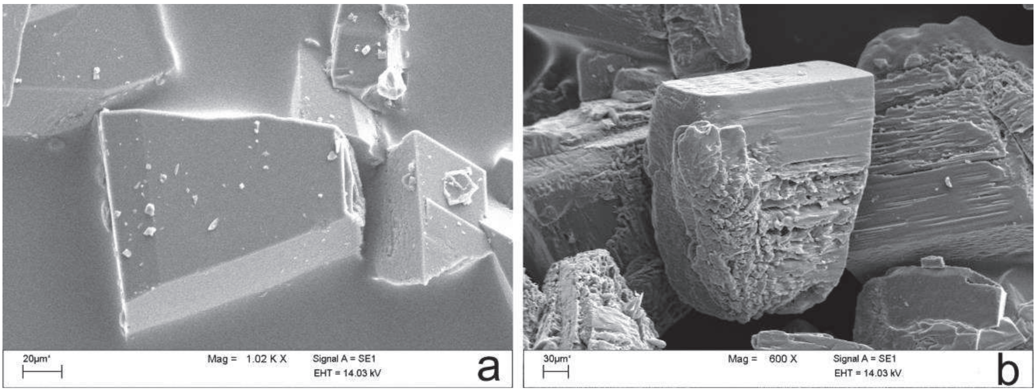

Figure 2. SEM images of lactose growing in a – pure supersaturated lactose solution, b – Ricotta cheese whey.

According to Cabrera-Vermilyea (1958) model, impurities adsorbed on the crystal face prevent the smooth growth of a new layer. Crystal of lactose grows in a spiral (Dincer et al., 2009); therefore layers grow one upon another. If the lower layer`s growth is stopped by immobile impurity, the upper layer grows to it, until it completely covers impurity and move farther (Hartel, 2001). Any kind of particles adsorbed on a crystal face, or rough and pitted surface, can potentially serve as nucleation centers. Furthermore, a rapid rate of cooling generally leads to nucleation at a lower temperature, rather than the slow cooling. (Hartel, 2001) In our case, these effects could trigger nucleation on the crystal surface and increase its roughness. However, it contradicts the theory that under conditions of rapid growth the additives cannot adsorb on the surface because of competition with the molecules of crystallizing material (Holsinger, 1988).

It remains unclear why (010) faces (base of crystal) were well-developed when other faces were rough (Fig. 2 b). Inasmuch as (010) face becomes the fastest growing face (Dincer et al., 2009), it can be assumed that it starts to form until the supersaturation becomes critical and impurities from solution accumulates in the boundary layer of the crystal later on (Hartel & Shastry, 1991).

CONCLUSIONS

This study presents influence of rapid and extra rapid cooling rate and crystallization at low temperatures on the morphological parameters of lactose crystals. Experiments show that temperature of crystallization and cooling rate impact mainly the size of lactose crystals. A strong dependence between crystallization temperature, cooling rate and crystals sizes was observed. Morphology of crystals was affected by impurities from solution more than crystallization conditions. There was some amount of irregular and needle-shaped crystals, obtained from Ricotta cheese whey. In a pure lactose solution no needle-shaped crystals were observed. The impact of impurities on certain faces of crystal requires an additional research.

REFERENCES

Cabrera, N. & Vermilyea, D.A. 1958. The Growth of Crystal from Solution. In Doremus, R.H., Roberts, B.W. & Turnbull, D. (eds.): *Growth and Perfection of Crystals.* Chapman & Hall, London, pp. 393–410.

Dincer, T.D., Ogden, M.I. & Parkinson, G.M. 2009. Crystal growth mechanisms of the (010) face of alpha-lactose monohydrate crystals. *J. Crys. Gro*. **311**, 2427–2432.

Guu, M.Y.K. & Zall, R.R. 1991. Lactose crystallization: Effects of minerals and seeding. *Process Biochem.* **26**, 167–172.

Hartel, R.W. 2001. *Crystallization in Foods*, Aspen Publishers, Inc, Gaithersburg, 325 pp.

Hartel, R.W. & Shastry, A.V. 1991. Sugar crystallization in food products. *Crit. Rev. Food Sci. & Nutr.* **1**(1), 49–112.

Holsinger, V.H. 1988. Lactose. In Jennes, R., Marth, E.H., Wong, N.P., Keeney, M. (eds): *Fundamentals of Dairy Chemistry, 3rd edn*, Springer, New York, pp. 279–343.

Jelen, P. 2003. Whey: Composition, properties, processing and uses. In Roginski, H. (ed): *Encyclopedia of Dairy Science.* Academic Press, Waltham, pp. 2653–2661.

Lifran, E.V., Vu, T.T.L., Durham, R.J., Hourigan, J.A. & Sleigh, R.W. 2007. Crystallization kinetics of lactose in the presence of lactose phosphate. *Powder Tech*. **179**, 43–54.

Livney, Y.D., Donhowe, D.P. & Hartel, R.W. 1995. Influence of temperature on crystallization of lactose in ice-cream. *Int. J. Food Sci. Tech.* 30, 311–320.

Miumoni, A., Schuck, P. & Bouhallab, S. 2005. Kinetics of lactose crystallization and crystal size as monitored by refractometry and laser light scattering: effect of proteins. *Dairy Sci. Tech* **85**, 253–260.

Modler, H.W. & Lefkovitch, L.P. 1986. Influence of pH, casein and whey protein denaturation on the composition, crystal size nad yield of lactose from condensed whey. *J. Dairy Sci.* **69**, 684–697.

Nickerson, T.A. 1956. Lactose crystallization in ice cream II. Factors affecting rate and quantity. *J. Dairy Sci.* **39**(10), 1342–1350.

Nickerson, T.A. 1954. Lactose crystallization in ice cream I. Control of crystal size by seeding. *J. Dairy Sci.* **37**(9), 1099–1105.

Paterson, AH.J. 2010. Production and Uses of Lactose. In McSweeney, P.L.H. & Fox, P.F. (eds): *Advanced in Dairy Chemistry Volume 3: Lactose, Water, Salts and Minor Constituents, 3rd edn,* Springer, New York, pp. 105–120.

Pisponen, A., Pajumägi, S., Mootse, H., Karus, A. & Poikalainen, V. 2013. The lactose from Ricotta cheese whey: the effect of pH and concentration on the size and morphology of lactose crystals. *Dairy Sci. & Technol.* **93**, 477–486.

Tan, R. 2010. Manufacture of Sweetened Condenced Milk and the Significance of Lactose Therein. In McSweeney, P.L.H., Fox, P.F. (eds): *Advanced in Dairy Chemistry Volume 3: Lactose, Water, Salts and Minor Constituents, 3rd edn,* Springer, New York, pp. 36–57.

Whritten, E.O. & Gould, S.P. 1931. Speed of crystallization of lactose, galactose, glucose and sucrose from pure solution. *Ind. Eng. Chem.* **23**(6), 670–673.