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## Research paper

# Optimizing NaCl uptake in experimental Emmentaler cheese: Effects of brining concentration, temperature, and duration

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#### ABSTRACT

Unidirectional salt transfer and moisture loss were investigated in experimental Emmentaler cheese cuboids after the following brining treatments: concentration (16 and 23 % w/w), temperature (11 and 16 °C), and time (24 and 72 h) over a ripening period of 0.5–3 months. Cheese manufacture and ripening conditions followed specifications for Emmentaler AOP cheeses. The results showed that the influence of 72 h brining time had a significant effect (P = 0.017) on salt uptake during the brining of experimental Emmentaler cheese. The temperature and concentration of the brine had no significant influence. By contrast, the higher temperature of 16 °C inhibited salt uptake and water loss, whereas the lower brine concentration of 16 % (w/w) did not reduce salt uptake but reduced water loss when brined equally at 11 °C for 72 h. These latter conditions are proposed to improve salt uptake and minimise moisture loss in Emmentaler AOP hard cheeses. The experimental data were modelled by Fick's second law over all ripening times, resulting in an overall diffusion coefficient of 2.21  $10^{-10}$  m<sup>2</sup> s<sup>-1</sup>.

## 1. Introduction

In the traditional cheese-making process, only milk, starters, nonstarters, and rennet are involved. Additionally, NaCl is very important and is added mostly either to the curd or through diffusion during brining by surface dry-salting and/or smear washing with brine (Bisig et al., 2025; Fröhlich-Wyder, 2012). During the last few decades, the NaCl content in Swiss Emmentaler AOP cheese has decreased from around 1.5 % to around 0.35 % (w/w) (Fröhlich-Wyder et al., 2022, 2025). Two major changes in the last century led to a lasting reduction of the NaCl content in Emmentaler AOP: the labour-intensive dry salting was abandoned from the 1970s onwards and, later on, the too eager salt reduction measures regarding the health aspects of the NaCl content in Emmentaler. Surprisingly, this reduction occurred despite Emmentaler salt content already being low compared to other cheese types at that time because of the unique large size of the wheels, amounting to approximately 100 kg. The resulting advantages for the texture properties of the cheese-smoother and more supple, enabling proper eye formation—were decisive in the salt reduction decisions in practice. Furthermore, NaCl was no longer necessary to prevent secondary fermentation, given the introduction of the facultative heterofermentative lactic acid bacteria in the 1980s (Fröhlich-Wyder et al., 2022) and the slowly fermenting propionic acid bacteria culture Prop 96 a decade later (Bachmann, 1998). However, flavour development was

neglected, and the cheeses tended to taste bland (Fröhlich-Wyder et al., 2025). Salt is necessary to enhance desired sweet and fruity notes and to avoid metallic notes and bitterness (Bisig et al., 2025). In fact, several studies have shown that a minimal salt content of around 0.8 % in cheese is necessary to avoid a bland taste (Bisig et al., 2025). An increase in the salt content of Emmentaler AOP is necessary to improve taste, but this is a challenging task to accomplish due to the size of the cheese loafs.

Mass transfer of salt in cheese (or in model systems) is complex, and different approaches have been studied over the last 100 years (see e.g. McDowell & Whelan, 1932; Floury et al., 2010; Pajonk, Saurel and Andrieu, 2003). Numerous studies have described the transfer of salt in cheese during brining and diffusion during succeeding ripening. When a cheese is in contact with brine, the difference in salt concentration between the cheese aqueous phase and the brine leads to diffusion of salt from the brine into the cheese, and the simultaneous loss of cheese moisture to rebuild the osmotic pressure equilibrium (Bisig et al., 2025; Giroux et al., 2022).

Factors that influence the salt uptake and movement from the exterior surface to the centre of the cheese are porosity (influenced by moisture content) of the cheese, tortuosity of the channels of water within the cheese matrix, proportion of water that is bound in cheese, viscosity of the free water, and interaction of NaCl with the protein matrix (Melilli, Barbano, Licitra, Portelli, et al., 2003). NaCl cannot travel through the protein matrix or the fat phase of cheese. Thus, cheese

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with a higher moisture content has a more porous structure but less tortuous water channels, and NaCl will penetrate the cheese more rapidly (Melilli, Barbano, Licitra, Portelli, et al., 2003).

Regardless of the type of cheese and its composition, the effective diffusion coefficients of salt have been reported in a review by Floury et al. (2010) to be between  $1\times 10^{-10}$  and  $5.3\times 10^{-10}\,\mathrm{m^2\,s^{-1}}$  at  $5\text{--}10\,^\circ\mathrm{C}$ , and Alehosseini et al. (2023) reported diffusion coefficients between  $1.4\times 10^{-10}$  and  $8.7\times 10^{-10}\,\mathrm{m^2\,s^{-1}}$  for rennet-induced micellar casein concentrate model systems with different temperatures, protein and Ca contents, and pH levels. These reported "effective diffusion coefficient values" have mostly been obtained by macroscopic and destructive analytical techniques and by adapted complex mathematical approaches. There are many experimental studies on the diffusion of salt and of CO2 for eye formation in Swiss-type cheese (Bisig et al., 2019; Fröhlich-Wyder et al., 2023; Lamichhane et al., 2021), whereas the diffusion of sugars, organic acids and peptides has been less extensively studied

Salt diffusion is one of the most important parameters during the ripening of cheese, as salt affects the water activity  $(a_W)$  of the cheese matrix and therefore the growth and survival of bacteria and the corresponding activity of enzymes, which influence the resultant aroma and flavour of the cheese, as well as the texture and colour of the cheese and the rind (Bisig et al., 2025). Additionally, in most cases, a large gradient of salt between the border zone and the core must be balanced during the ripening period. Equilibrium times range from 2 weeks in soft cheese to about 10 months in extra-hard cheese (e.g. Parmesan-type cheese).

Cheese ripening involves different mechanisms in parallel, such as bacterial growth, protein degradation, multi-component diffusion of solutes, migration of water, dehydration, temperature and pH changes, solubilisation of  ${\rm Ca}^{2+}$ , and many more.

The diffusion of salt in cheese is, by definition, the mass transfer caused by a random molecular motion in a region with a salt gradient. Molecular diffusion coefficients at a constant temperature can be predicted in very diluted solutions using the Stokes–Einstein equation. Diffusion in a heterogeneous system—such as cheese, a multi-phase and multi-component system—is more complex, as various hydrophobic and electrostatic interactions of salt within the porous cheese system and a complex protein network with incorporated fat globules may take place. The relationship between the concentration  $C(x, t) = C_{\text{salt}}$  of a diffusing substance, such as salt, and its displacement with time t in one direction x at a constant temperature is represented by Fick's second law, considering the diffusion coefficient D as constant (Floury et al., 2010; Gros & Rüegg, 1987):

$$\frac{\partial C_{salt}}{\partial t} = D \frac{\partial^2 C_{salt}}{\partial x^2}$$
 (Formula 1)

Gros et al. (1987) and Floury et al. (2010) reviewed different experimental techniques for the diffusion of salt and proposed possible mathematical solutions for obtaining effective diffusion coefficients ( $D_{\rm eff}$ ) in cheese. However, some drawbacks of the Fickian approach have been reported in food and summarised by Doulia et al. (2000).  $D_{\rm eff}$  may not be constant but dependent on salt concentration and sample temperature. Additionally, volume changes, especially dehydration and pH changes, are often ignored, as are the complex interactions between salt and the cheese matrix.

The aim of this study was to determine the importance of the three factors, starting from standard conditions for experimental Emmental cheese—brine concentration, temperature, and brining time (16 % salt, 11 °C and 24 h, respectively)—and their interactions on the salt uptake and diffusion during the ripening of Emmentaler cheese cuboids over a ripening time of 3 months under conditions as close as possible to practice (size of cheese cuboids, ripening time, ripening temperature profile, and brine treatment). Cheese moisture content was kept as constant as possible by wrapping the cuboids with a plastic bag, and ripening was performed according to the specifications of a standard Emmentaler AOP cheese-making procedure (10 days: 11 °C, 60 days:

22  $^{\circ}$ C, after day 70: 12–13  $^{\circ}$ C) to mimic conditions in practice. The experimental data were modelled using Fick's second law. The findings suggest recommendations for increasing the salt content of Emmentaler AOP in the future.

#### 2. Materials and methods

#### 2.1. Preparation of cheese cuboids (experimental set-up)

Model Emmentaler cheese was produced according to Guggisberg et al. (2015) to obtain a model cheese that was as similar to the original Emmentaler AOP as possible. The coagulated milk was filled in squared moulds instead of round moulds and pressed for 24 h. The cheese samples were cut in cuboid (rectangular parallelepiped-shaped) pieces of 7 cm  $\times$  7.5 cm  $\times$  30 cm before brining (volume: 1575 cm<sup>3</sup>, ( $\sim$ 25 °C), density:  $1.075 \text{ g cm}^{-3}$ , mass: 1.7 kg), as shown in Fig. 1a. In this study, Propionibacterium freudenreichii (PAB, Prop 96) was not added to the untreated cheese milk, as eye formation was supposed to disturb NaCl diffusion. Brining experiments were carried out, as illustrated in Fig. 1. for 24 h or 72 h. The brining of the 7 cm  $\times$  7.5 cm surface guaranteed a uniaxial mass transfer of NaCl. After the brine treatment (Table 1), all samples were placed in transparent vacuum pouches (vacuum bag WAW 90 µm, VC999 Verpackungssysteme AG, CH-9100 Herisau, Switzerland, with permeability parameters:  $O_2$ :  $<80 \text{ cm}^3 \text{ m}^{-2} 24 \text{ h}^{-1} \text{ bar}^{-1}$ ;  $H_2O$ : <6g m<sup>-2</sup> bar<sup>-1</sup> 24 h<sup>-1</sup>;  $CO_2$ : <174 cm<sup>3</sup> m<sup>-2</sup> bar<sup>-1</sup> 24 h<sup>-1</sup>) to prevent oxidation, dehydration and mould growth, after a drying step of 24 h. All cheese samples were produced from the same milk to avoid potential differences in the composition of the final model cheese.

#### 2.2. Study design and salt brine

The cuboid cheese pieces were brought in contact with the salt brine adjusted in Baumé (Bé) on two levels:  $15^\circ$  Bé [0.16 kg<sub>NaCl</sub> kg<sup>-1</sup><sub>water+NaCl</sub> = 16% (w/w)] or  $21^\circ$  Bé [0.23 kg<sub>NaCl</sub> kg<sup>-1</sup><sub>water+NaCl</sub> = 23% (w/w)], (Weast & Astle, 1979, p. 60), for either 24 h or 72 h at  $11^\circ$ C or  $16^\circ$ C (Floury et al., 2010; Pajonk et al., 2003) (Fig. 1b and Table 1). The brine solutions were adjusted with lactic acid (80%) to pH 5.4. The brine solutions used in this study were prepared from an initial brine solution used for several years in the Agroscope research cheese dairy. Old saturated brine was used because it contained "usual Ca content" and pH and was close to practical conditions (Jakob et al., 2005).

After brining, the cuboid cheese samples were air dried for 24 h and packed in a vacuum bag. These samples were stored for 9 days at 11  $^{\circ}\text{C}$  (cold-room storage) and for 60 days at 22  $^{\circ}\text{C}$  (warm-room storage) to simulate practical conditions. After a total of 70 days, the samples were stored at 12–13  $^{\circ}\text{C}$ .

Each experimental variant consisted of three cuboid cheese samples, which were analysed at distinct timepoints of the ripening process, after 0.5 months, 1.5 months, and 3 months. At each time point, a sample was sliced into 10 pieces with increasing distance from the contact surface ( $x = 0.0, ..., 0.3 \, m$ ), and their salt and moisture contents were analysed.

## 2.3. Analysis of NaCl and moisture content

The analysis of NaCl was calculated after the analysis of Cl by argentometric titration according to ISO 9543:2006 (IDF 88:2006). The moisture content of cheese was analysed with a drying cabinet method by following ISO 5534:2004 (IDF 4:2004).

## 2.4. Calculation of equilibrium NaCl and moisture values

Calculation of equilibrium NaCl (g kg $^{-1}$ ) values (for t $\rightarrow \infty$ ) was performed by integrating the NaCl content along the distance from 1.5 to 27.5 cm. The experimental NaCl profiles were loaded from an Excel file into the TableCurve2D software (Inpixon, Düsseldorf, Germany) and were integrated from 1.5 to 27.5 cm, as the values were not normally



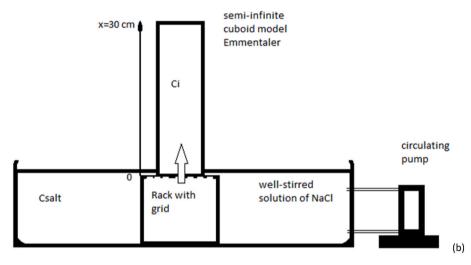


Fig. 1. Creation of cuboid cheese samples (a) and (b) diagram of the semi-infinite cuboid model Emmentaler standing (on a rack with a grid) vertically in the well-stirred solution containing NaCl (either for 24 h or for 72 h). Additionally, the cheese sample was stabilised by fixation (not shown in the diagram). The maximum length of the model Emmentaler was x=30 cm, and the height and width were 7 cm and 7.5 cm, respectively. Salt concentration in the cheese sample (Ci) was analysed after 0.5, 1.5, and 3 months by slicing the cheese sample. The concentration of salt in the brine was Csalt (adjusted in Baumé (Bé) on two levels: 16% (w/w) or 23% (w/w) ( $15^{\circ}$  Bé or  $21^{\circ}$  Bé)). The temperature of the whole system was controlled by the temperature control of the climatized room (on two levels:  $11^{\circ}$ C and  $16^{\circ}$ C). The arrow points in the direction of NaCl diffusion.

Table 1
Experimental design: The salt brine properties were on two levels for each of the three factors: temperatures 11 and 16 °C; brining time of 24 h and 72 h, and NaCl concentration of 16 % (w/w) and 23 % (w/w) (15° and 21° Bé).

Variant	Temperature (T): [°C]	Duration (D): [h]	Concentration (C): (w/w)	Concentration (C): [kg <sub>NaCl</sub> kg <sup>-1</sup> <sub>water+NaCl</sub> ]	Concentration (C): [° Bé]
1	11	24	16	0.16	15° Bé
2	11	24	23	0.23	21° Bé
3	11	72	16	0.16	15° Bé
4	11	72	23	0.23	21° Bé
5	16	24	16	0.16	15° Bé
6	16	24	23	0.23	21° Bé
7	16	72	16	0.16	15° Bé
8	16	72	23	0.23	21° Bé

distributed. Calculated equilibrium values (for  $t\to\infty)$  for NaCl [g kg $^{-1}$ ] resulted by division with 26 cm to achieve a calculated uniform distribution. For moisture values, the average was calculated, as the values were normally distributed.

2.5. Determination of the mean diffusion coefficients after the second law of fick

Fick's second law (formula 1) has many well-known analytical and numerical solutions, depending on the specific initial- and/or boundary

conditions. Typically, these solutions are obtained assuming a constant diffusion coefficient, D. Under this assumption, the following initial boundary value problem, based on Fick's second law in one dimension, is used to determine the mean diffusion coefficient:

$$\frac{\partial c_{salt}(x,t)}{\partial t} = D \frac{\partial^2 (c_{salt}(x,t))}{\partial x^2} \quad x \in [0,\infty], t \in [0,\infty]$$

$$c_{salt}(x,0) = M \cdot \delta(x) + c_1$$

$$\frac{\partial c_{salt}(0,t)}{\partial x} = 0$$

Here,  $c_1$  [kg<sub>NaCl</sub> kg<sub>cheese</sub>] is the initial background NaCl concentration of the cheese sample prior to brining, and M [kg<sub>NaCl</sub> kg<sub>cheese</sub> × m] is the additional NaCl amount taken up by the sample during brining. These two contributions define the total amount of NaCl contained in the cheese sample for any t > 0.

From a physics point of view, our initial condition at t=0 describes a cheese sample with an original background concentration of  $c_I$  where an additional finite amount of salt M is located with very high density at one end of the cheese sample at x=0. Hence, contact with the salt brine takes place at some time t<0 and is not captured by our model. However, the effect of salt brine is included by the contribution from the strongly simplified Dirac distribution at boundary x=0 (Crank, 1975, p. 12). Since the cheese is fully disconnected from the salt solution for t>0, a Neumann boundary condition is assumed (isolated end), where salt fluxes are held fixed at  $dc_{salt}/dx=0$ . The influence of the second boundary was fully omitted due to its location far away from the initial distribution.

Under these assumptions, an adapted version of Green's function of the 1-D heat equation (heat kernel) solves our model system (Atkins, 2001):

$$c_{salt}(x,t) = c1 + \frac{M}{\sqrt{\pi^* D^* t}} exp \frac{-x^2}{4^* D^* t}$$
 (Formula 2)

where  $c_I$  is the mean NaCl content of the cheese samples without brining, and M represents the concentration of NaCl deposited at time t=0 in the plane x=0, or a factor that includes the surface of the cuboid cheese sample that was in contact with the brine  $(7 \text{ cm} \times 7.5 \text{ cm} = 52.5 \text{ cm}^2)$  and the amount of NaCl that was taken up during the brining time (24 h or 72 h). D is the mean diffusion coefficient that is modelled by the non-linear least-squares method, assuming that D is constant. Setting  $c_I = 0$  revokes the solution proposed by Crank (1975) for a semi-infinite sample using reflection at the boundary (see Equation 2.7 on p. 13).

## 2.6. Statistical analysis

Fitting and modelling the experimental data were performed using R (www.r-project.org) and the algorithm/method of "non-lineal least-squares" in the library (nls). Three-way and two-way ANOVA was applied using the library rstatix and the function "anova\_test" for the three fixed factors (brine concentration, brining time, and brine temperature) without interactions. Rstatix provides a pipe-friendly framework for performing different types of ANOVA tests.

#### 3. Results and discussion

Table 1 presents the study design. With 8  $\times$  3 variants of model Emmentaler cheese, two different salt brine concentrations (16 % (w/w) = 15° Bé and 23 % (w/w) = 21° Bé), two brining times (24 h, 72 h), and two levels of the brine temperature (11 °C, 16 °C) were studied. Each experimental variant consisted of three cuboid cheese samples, which were analysed at distinct timepoints of the ripening process, after 0.5 months, 1.5 months, and 3 months. At each time point, a sample was sliced into 10 pieces with increasing distance from the contact surface (x = 0.0, ..., 0.3 m), and their salt and moisture contents were analysed.

#### 3.1. Analysis of moisture loss of model emmentaler cheese cuboids

The moisture content and the related porosity of cheese are known to influence the absorption of NaCl (Melilli, Barbano, Licitra, Portelli, et al., 2003). For this reason, the cheese cuboids were taken and prepared from the same cheese production. The results for the mean water content of all slices are presented in Table 2. The mean water contents were comparable among the variants and the ripening steps (no significant effect p>0.05). The plastic film around the cheese cuboids during ripening prevented the drying out of the cheeses and mould formation. The standard deviation, however, showed that the moisture content was not homogeneous between the 10 slices of each cuboid block (Fig. 2).

Fig. 2 shows either linear or wavy lines. We observed the influence of brine temperature: the water distribution was more homogeneous in the cuboids treated at  $16\,^{\circ}$ C than at  $11\,^{\circ}$ C. A reason for this could be the interface of the cheese cuboids, which was immersed in the brine: the first  $10\,^{\circ}$ C of the  $16\,^{\circ}$ C-treated cuboids compared with the  $11\,^{\circ}$ C-treated cuboids had a substantially lower moisture loss if the brine concentration amounted to  $23\,^{\circ}$  (w/w). The higher temperature, which obviously led to a sealing of the surface, whether it is due to liquid fat (Kimber et al., 1974) and/or to the lower porosity of the interface, thus not only slowed down salt absorption but possibly also water loss, especially at a NaCl concentration of  $23\,^{\circ}$  (w/w) (Fig. 2).

Furthermore, a brine treatment at a lower concentration of 16% (w/w) was responsible for a more homogeneous moisture distribution from the beginning of the ripening time, especially for the 24 h duration (Fig. 2). This is somewhat surprising because the salt uptake was comparable to that of the higher brine concentration (Fig. 3). Thus, the water loss was smaller for the 16% (w/w) treatments. This suggests an interesting interaction, which might be important for an application in the production of Emmentaler AOP with its large size: in the 30 cm long cheese cuboids, a maximal salt uptake with minimal water loss was possible with a brine treatment of 16% (w/w) at 11 °C for 72 h (Fig. 4). The same phenomenon was also observed by Fröhlich-Wyder et al. (2025) in ripening experimental Emmentaler cheeses with a diameter of

Table 2 Mean moisture contents [g kg $^{-1}$ ] of the cuboid cheese samples. The values in the table represent the mean values of 10 slices (n = 10) at each time point (0.5, 1.5, and 3 months). The standard deviation (SD) is also provided (Experimental design according to Table 1.).

Variant	Moisture content [g kg <sup>-1</sup> ] 0.5 month	SD	Moisture content [g kg <sup>-1</sup> ] 1.5 months	SD	Moisture content [g kg <sup>-1</sup> ] 3 months	SD
1 (T11; D24; C16)	383.2	5.6	381.2	9.7	379.7	5.1
2 (T11; D24; C23)	376.9	17.4	381.4	13.1	379.4	5.4
3 (T11; D72; C16)	377.1	7.2	381.3	7.7	378.6	9.7
4 (T11; D72; C23)	368.2	9.1	380	18.0	378.4	8.0
5 (T16; D24; C16)	380	3.9	383.9	2.8	383.1	2.5
6 (T16; D24; C23)	383.7	11.4	385.8	8.8	390.2	5.0
7 (T16; D72; C16)	375.9	7.4	381.4	2. 7	381.4	4.7
8 (T16; D72; C23)	385.2	8.2	379.8	7.1	376.9	5.4
ANOVA						
Brine concentration	n.s.		n.s.		n.s.	
Brining time	n.s.		n.s.		n.s.	
Brine temperature	n.s.		n.s.		n.s.	

n.s.: not significant.

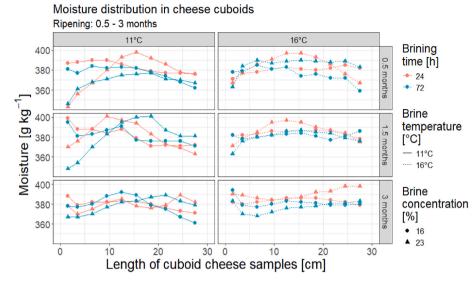


Fig. 2. Moisture distribution after 0.5, 1.5, and 3 months for 11 °C (left) and 16 °C (right) and 16 % (w/w) and 23 % NaCl. The 6 figures show the brining duration of 24 h and 72 h.

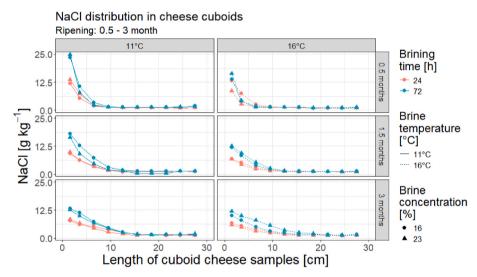


Fig. 3. NaCl distribution after 0.5, 1.5, and 3 months for 11 °C (left) and 16 °C (right) and 16 % (w/w) and 23 % (w/w) NaCl. The 6 figures show the brining times of 24 h and 72 h.

30 cm under brine conditions identical to those of the present study. In the 1980s, a study with Romano-type cheese slices showed that the salt uptake for the three brine concentrations of 14.8, 18.9, and 24.9 % differed only slightly, whereas the water loss doubled between 14.8 and 24.9 % (Bisig et al., 2025; Guinee & Fox, 1986).

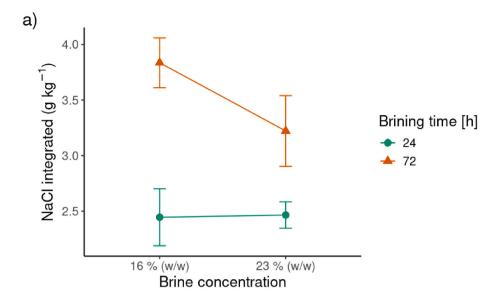
## 3.2. Analysis of NaCl uptake of model emmentaler cheese cuboids

NaCl diffusion performance in the model Emmentaler cheese cuboids for the different brine conditions is shown in Fig. 3. NaCl decreased in a non-linear manner along the cuboid cheese blocks from the immersed side to the other, as expected. For the brine temperature, a substantial difference was observed between 11 and 16 °C: the 11 °C brine treatment (Fig. 3 left) led to the highest NaCl levels at the immersion point, especially for the 72 h brining duration, meaning that more salt diffused into the cheese samples. To study the total amount of absorbed NaCl in the cheese cuboids ripened for 0.5, 1.5, and 3 months, the integral from distance 1.5–27.5 cm was calculated (Table 3). The integrated value was divided by 26 to estimate an equilibrium value for NaCl at  $t \to \infty$ . In fact,

the brine duration (24 h versus 72 h) was the only factor influencing equilibrium NaCl content in the cheese cuboids significantly (p < 0.05), as calculated by the integral, rather than the brine temperature (Table 3).

For the  $16^{\circ}$ C-treated cheese cuboids (Fig. 3, right), the levels of NaCl at the first sample point were highest; by contrast, these levels were only slightly higher for the 72 h compared to the 24 h brining duration. Against expectations, the NaCl levels were not higher at  $16^{\circ}$ C compared to  $11^{\circ}$ C brine temperature; rather, the opposite was observed. A suggested reason for this phenomenon could be a less permeable cheese surface at  $16^{\circ}$ C due to more liquefied milk fat and/or to the lower porosity of the interface at  $16^{\circ}$ C compared to  $11^{\circ}$ C (Melilli, Barbano, Licitra, Portelli, et al., 2003).

Alehosseini et al. (2023) reported a decrease in the diffusion coefficient by increasing the salting temperature from 10 to 40 °C in rennet-induced micellar casein concentrate model systems. The authors suggested that a stronger syneresis at higher temperatures, which led to moisture loss, was responsible for this observation. A different experimental study of NaCl diffusion in French Emmental during brining by



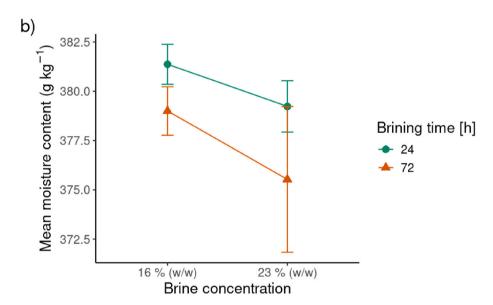


Fig. 4. Interaction plots of the  $11^{\circ}$ C-treated cheese cuboids for the equilibrium NaCl contents calculated by integration (a) and the mean moisture content (b) of all the ripening timepoints (0.5, 1.5, and 3 months) together (N = 3).

Table 3 Calculated equilibrium value (for  $t \to \infty$ ) for NaCl (g kg<sup>-1</sup>) calculated by integration from 1.5 to 27.5 cm and divided by the distance 26 cm (difference between 27.5 and 1.5 cm = 26 cm) (Experimental design according to Table 1.).

Variant	Total NaCl content [g kg <sup>-1</sup> ] 0.5 month	Total NaCl content [g kg <sup>-1</sup> ] 1.5 months	Total NaCl content [g kg <sup>-1</sup> ] 3 months	Mean NaCl content [g kg 1]	SD NaCl content [g kg <sup>-1</sup> ]
1 (T11; D24; C16)	2.079	2.317	2.940	2.45	0.445
2 (T11; D24; C23)	2.367	2.327	2.702	2.47	0.206
3 (T11; D72; C16)	3.388	4.035	4.085	3.84	0.388
4 (T11; D72; C23)	3.035	2.788	3.842	3.22	0.551
5 (T16; D24; C16)	2.415	2.092	2.487	2.33	0.210
6 (T16; D24; C23)	1.652	1.825	2.069	1.85	0.210
7 (T16; D72; C16)	2.035	2.763	3.081	2.63	0.536
8 (T16; D72; C23)	2.133	3.085	4.294	3.17	1.083
ANOVA					
Brine concentration	n.s.	n.s.	n.s.	n.s.	
Brining time	n.s.	0.025*	0.019*	0.017*	
Brine temperature	n.s.	n.s.	n.s.	n.s.	

n.s.: not significant; \* p-value 0.01 > x 0.05.

Pajonk et al. (2003) showed a clear increase in NaCl diffusion with increased temperature from 4 °C to 18 °C in the brine for the first 24 or 48 h within the first 2.5 cm. A higher brine temperature generally increases the salt uptake of cheese due to higher salt diffusivity and the reduction of cheese serum viscosity (Guinee & Fox, 2017).

Moisture loss causes shrinkage of the cheese structure and decreases porosity, which impedes moisture movement out and salt movement into the cheese, as observed by Melilli, Barbano, Licitra, Portelli, et al. (2003). In two cheeses of the same type, the cheese with a higher moisture content absorbs salt more rapidly. Melilli, Barbano, Licitra, Portelli, et al. (2003) studied the influence of brine temperature in Ragusano cheese between 12 °C and 24 °C on salt uptake. Generally, total salt uptake and moisture loss increased with increasing brine temperature. However, for the temperatures between 12 °C and 15 °C, no significant difference was visible.

In the present study, the higher brine temperature of  $16\,^{\circ}\mathrm{C}$  did not significantly increase the salt content of the cheese compared to  $11\,^{\circ}\mathrm{C}$ . Again, this is surprising, but similar findings were found by Melilli, Barbano, Licitra, Portelli, et al. (2003) with Ragusano cheese: the lowest uptake of NaCl was measured in the  $15\,^{\circ}\mathrm{C}$ -treated cheeses compared to cheeses treated at 12, 18, and  $21\,^{\circ}\mathrm{C}$ . A possible explanation can be found in the works of Melilli et al. (2003a), Melilli et al., 2004, who observed lower porosity at higher brine temperatures at the interface. A study with different brining conditions for Feta cheese showed that high temperatures as high as  $22\,^{\circ}\mathrm{C}$  can significantly reduce NaCl uptake due to a contraction of the cheese matrix. By contrast, at lower temperatures of 3, 6, and  $10\,^{\circ}\mathrm{C}$ , the protein matrix expanded (McMahon et al., 2009), as examined using laser scanning confocal microscopy.

Melilli, Barbano, Licitra, Portelli, et al. (2003) further concluded that a decrease in salt penetration at lower brine temperature may be caused by an increase in the viscosity of the water phase of the cheese, whereas the mechanism of the impact of changing brine concentration might be due to an impact on the porosity of the cheese at the interface between brine and cheese. Two competing factors (viscosity and porosity) appear to influence complex salt intake. This may explain the lack of a significant difference between 11 °C and 16 °C observed in the present study. Another explanation could be the fact that the lower brine temperature of 11 °C compared to 16 °C did not cause a relevant increase in the viscosity of the water phase of the cheeses.

Similar to the brine temperature, the difference in the calculated NaCl equilibrium value (for  $t\to\infty$ )—between  $16\,\%$  (w/w) and  $23\,\%$  (w/w)—was small and not significant for any of the maturation levels in this study (Table 3) (p > 0.05). Guinee et al. (2014) reported a generally greater salt uptake by cheese by increasing the brine concentration from  $5\,\%$  to  $25\,\%$  (w/w). It must be noted, however, that a NaCl concentration of  $\sim\!25\,\%$  (w/w) may cause a reduction of the salt uptake due to dehydration or shrinkage of the protein network on the surface of the cheese, enabling a barrier layer (Melilli et al., 2005). Furthermore, a laboratory-scaled experiment showed that a dense protein network (median pore size  $=2.92\,\mu m$  compared to  $4.45\,\mu m$ ) could slow down salt diffusion significantly (Alehosseini et al., 2023; Sharma, Sheehan, & Floury, 2021).

As shown above, for the 11 °C treatment, the difference of salt uptake between the two levels of the brining time (24 h or 72 h) was clear, but the difference between 16 % NaCl ( $\bullet$ ; w/w) and 23 % NaCl ( $\bullet$ ; w/w) was not strong and therefore not relevant (not significant, p>0.05). Although a higher brine concentration (23 % versus 16 %) is known to result generally in faster salt absorption, the higher water loss (Fig. 2) for 23 % brine may have led to a slight reduction in salt uptake. Water loss might have negatively influenced the porosity and lowered the salt uptake, and we found a similar salt uptake for 16 % and 23 % brine.

By contrast, in Fig. 3 (right), samples processed in the 16  $^{\circ}$ C salt brine showed generally lower NaCl values. As for the 11 $^{\circ}$ C-treated cheeses, a stronger impact on the salt content was seen from the brining time (72 h versus 24 h) than from the brine concentration 16 % and 23 % (w/w). Salt uptake by cheese generally increases with increasing brining time

(Guinee, 2007).

Melilli et al. (2003a, 2004, 2005, 2006) studied the phenomenon of salt uptake of non-saturated brine (18 % w/w) compared to saturated brine. The increased rate of salt uptake with 18 % brine compared to saturated brine (~26 % w/w) was supposed to be related to the impact of lower brine concentration on the moisture content and the porosity of the cheese near the surface of the Ragusano cheese block. Moisture loss causes shrinkage of the cheese structure and decreases porosity, which impedes moisture movement out and salt movement into the Ragusano cheese blocks, as indicated by Melilli et al. (2003a). Interestingly, the use of non-saturated 18 % brine compared to saturated brine for the first 8 days of brining reduced moisture loss and cheese shrinkage at the interface and allowed more salt penetration. This phenomenon was also found in other cheese variants, such as Parmigiano Reggiano (Resmini, Volonterio, Annibaldi, & Ferri, 1974), and it explains the observed good salt uptake in lower concentrated brine solutions.

However, we suggest caution when comparing the data of other studies, given the substantial differences in experimental outlines, such as the cheese variety or even model systems, the time point of analyses, the distance from the interface, and the size of the slice.

3.3. Estimation of mean diffusion coefficients of NaCl uptake with model emmentaler cheese by Fick's second law

For each of all eight experimental setups (variants 1–8; timepoints: 0.5, 1.5, and 3 months), the mathematical solution (Formula 2) of the mass diffusion equation was used to fit the experimentally obtained NaCl profiles and to determine "mean" diffusion coefficients. Formula 2 and experimental NaCl profiles from each variant at a time (0.5, 1.5, and 3 months) were loaded in R and by using non-linear least squares (nls), starting with initial values for M (starting value = 1) and D (starting value =  $1*10^{-10}$ ) and a maximum of 1000 iterations diffusion coefficients (D1–D8) were calculated for each experimental variant. M and D values resulted from fitting Formula 2 simultaneously for all three samples (with brining times of 0.5, 1.5, and 3 months) and salt concentration measured at the 10 different locations.

Hence, our approach resulted in one set of values M and D for each variant (Table 4). It is interesting that the values for the diffusion coefficients D1–D8 were in a rather narrow range between  $1.48 \times 10^{-10}$  and  $3.17 \times 10^{-10}$  m<sup>2</sup> s<sup>-1</sup> for all eight variants. This indicates that NaCl diffusion in Emmentaler cheese remains in a low range, even for practical standard ripening conditions for Emmentaler AOP cheese and

**Table 4** Modelled M and D values by the non-linear least-squares method, by fitting NaCl profiles for each experimental variant and at 0.5–3 months according to Formula 2. M represents the amount of NaCl deposited at time t=0 in the plane x=0, whereas t=0 indicates the time point after contact of the cheese with the brine (either 24 h or 72 h). D: Diffusion coefficient (Experimental design according to Table 1.).

Variant	M [ $kg_{NaCl} kg_{cheese}^{-1} \times m$ ]	$D [m^2 s^{-1}]$	r <sup>a</sup>
1 (T11; D24; C16)	$4.289 \times 10^{-4}$	$2.551 \times 10^{-10}$	0.987
2 (T11; D24; C23)	$4.676 \times 10^{-4}$	$2.397 \times 10^{-10}$	0.991
3 (T11; D72; C16)	$8.808 \times 10^{-4}$	$2.607 \times 10^{-10}$	0.995
4 (T11; D72; C23)	$7.507 \times 10^{-4}$	$1.802  imes 10^{-10}$	0.989
5 (T16; D24; C16)	$4.265 \times 10^{-4}$	$3.165 \times 10^{-10}$	0.975
6 (T16; D24; C23)	$2.528 \times 10^{-4}$	$1.479 \times 10^{-10}$	0.989
7 (T16; D72; C16)	$4.805 \times 10^{-4}$	$1.660 \times 10^{-10}$	0.985
8 (T16; D72; C23)	$5.927 \times 10^{-4}$	$2.045 \times 10^{-10}$	0.959
Mean:		$2.210 \times 10^{-10}$	
ANOVA			
Brine concentration	n.s.	n.s.	
Brining time	0.018*	n.s.	
Brine temperature	n.s.	n.s.	

n.s.: not significant.

<sup>&</sup>lt;sup>a</sup> Correlation coefficient between analysed and fitted data (Fig. 5; formula: 2).

between 0.5 and 3 months. Comparing these values with the diffusion coefficients reported by Floury et al. (2010) for different cheeses with D between 1 and 5.3 x  $10^{-10}$  m<sup>2</sup> s<sup>-1</sup> at 10–15 °C, the D1–D8 values of this study are similar. Pajonk et al. (2003) reported effective diffusion coefficients between 0.62 and 2.2  $10^{-10}$  m<sup>2</sup> s<sup>-1</sup> for Emmental cheese with different brining conditions (4–18 °C, 24–48 h) in a similar experimental setup. Guinee and Fox (1987) found a D value of 1.8  $10^{-10}$  m<sup>2</sup> s<sup>-1</sup> for Emmental cheese at 15–16 °C. These D values were all similar to those in this study. The differences in the study by Pajonk et al. (2003) compared to the present one can be explained by the fact that the cheese surface was in contact with the brine for 24 h or 48 h compared to 24 h and 72 h in this study. Pajonk et al. (2003) stopped the experiment after 24 h or 48 h, and the samples were analysed only from a 0-2.5 cm distance from the contact surface. By contrast, in our study, the samples were left for ripening, according to the standard Emmentaler AOP ripening process, for 0.5, 1.5, or 3 months, and we measured the NaCl profile across a distance of 30 cm at each of the three ripening times. The corresponding figures with NaCl profiles fitted by the nls method are presented in Fig. 5 (a and b). As the nls fitting was performed in this study for the three ripening times of 0.5, 1.5, and 3 months simultaneously to calculate a diffusion coefficient, the agreement of the experimental values with the fitted values were checked visually in Fig. 5 and by calculating the correlation coefficients (r) between the analysed and fitted data (Table 4). We observed some underestimation of the experimental data, especially for values for the time point of 3 months. The reason for these findings could be due to the ripening temperature that was increased between day 10 and day 70 (from 11  $^{\circ}\text{C}$  to 22  $^{\circ}\text{C}$ ) and lowered again on day 70 (from 22  $^{\circ}\text{C}$  to 12–13  $^{\circ}\text{C}$ ), as usual in practice for Emmentaler AOP.

Although Pajonk et al. (2003) found a similar range of diffusion coefficients, they were confronted with some over-estimation of the experimental values close to the interface zone and under-estimation of the experimental values in the low concentration range by using Fick's diffusion equation describing unsteady state mass transfer. These authors suggested that the "constant" diffusion coefficient model that was based on the second law of Fick was not quite ideal for interpreting the NaCl diffusion kinetics for the Emmental brining process. Compared to our study, a ripening time between 0.5 and 3 months can be viewed as sufficiently precise, at least for ripening times of 0.5 months and 1.5 months. The highest values for M in the model were found for experimental variants 3 and 4, and 7 and 8 (Table 4), which represented the brining time of 72 h. This was in agreement with the visual evaluation.

#### a) Salt diffusion at 11°C

#### Diffusion of NaCl in model Emmentaler at 11°C Ripening: 0.5 - 3 months 72 h 0.025 0.020 0.015 Time [months] 0.010 0.5 months 0.005 1.5 months [g kg 3 months 0.000 NaCI | 0.025 concentration [%] 0.020 0.015 16% **▲** 23% 0.005 0.000 0.2 0.3 0.0 0.1 Length of cuboid cheese samples [m]

## b) Salt diffusion at 16°C

#### Diffusion of NaCl in model Emmentaler at 16°C Ripening: 0.5 - 3 months 0.025 0.020 0.015 0.010 0.5 months 0.005 1.5 months NaCl [g kg] 3 months 0.000 0.025 Brine 0.020 [%] 16% 0.015 0.010 0.005 0.2 0.3 0.0 0.3 Length of cuboid cheese samples [m]

Fig. 5. Experimental (symbols) and modelled (dashed lines) NaCl profiles of model Emmentaler cuboids, following Ficks' second law. The diffusion coefficient was calculated by fitting all 3 time points (0.5–3 months) per variant (1–8) with the "non-linear least-squares" algorithm (Experimental parameters are presented in Table 1).

#### 4. Conclusion

The brining temperature, brining time, and salt brine concentration affected NaCl uptake and water loss during the ripening time of 3 months of experimental Emmentaler cheese differently. Brining time (72 h versus 24 h) was the main factor affecting NaCl uptake by cheese, which was in agreement with the literature: salt content in the 72 h-treated cheese cuboids was around 1/3 higher compared to the 24 h-treated cheese cuboids at 11  $^{\circ}$ C. Against expectations, the higher brine temperature of 16  $^{\circ}$ C seemed to slightly inhibit salt uptake and water loss. Furthermore, the water distribution within the cheese cuboids was more homogeneous. Our results suggest that the process of NaCl diffusion is influenced by a variety of complex processes, such as water diffusion in the opposite direction to NaCl diffusion, and surface phenomena, such as a decrease in the pore size or partial liquefaction of fat at the interface between brine and cheese matrix when brine temperature is elevated, leading to sealing.

As previously demonstrated by other researchers with similar experiments, the salt uptake between the two brine concentrations of  $16\,\%$  and  $23\,\%$  (w/w) did not vary significantly, in contrast to the water loss, which was higher in the cheeses treated with  $23\,\%$  (w/w) brine solution, and the water content, which was more heterogeneously distributed within the cheese cuboids. This phenomenon was seen only in the cheeses brined at  $11\,^\circ$ C and could lead to an interesting implication in practice for enhancing salt uptake with minimal water loss and optimal water distribution: a prolongation of the brine treatment at  $11\,^\circ$ C and at a lower concentration of  $16\,\%$  (w/w). NaCl diffusion in model Emmentaler samples was observed over a long period, starting from 0.5 to 3 months of ripening. From the experimental NaCl profiles, the calculated mean diffusion coefficients using one solution of Fick's second law were in agreement with the literature.

The results additionally showed that the salt diffusion in model Emmentaler cuboids was slow in one direction. The front of the NaCl was found to be between 10 and 20 cm after 3 months. An Emmentaler AOP has a height between 16 and 27 cm, indicating that 3 months is not sufficient to achieve NaCl equilibrium in the centre of the cheese. These findings are of high relevance for cheese production and should be implemented in the production of cheese varieties with very low salt contents, such as Emmentaler AOP. For full size Emmentaler AOP a salt brining time should be tripled as well starting from standard conditions of about 48 h.

#### CRediT authorship contribution statement

Dominik Guggisberg: Writing – original draft, Visualization, Project administration, Methodology, Formal analysis, Data curation, Conceptualization. Walter Bisig: Writing – review & editing, Writing – original draft, Supervision, Project administration, Methodology, Conceptualization. Florian Loosli: Methodology, Investigation, Data curation, Conceptualization. Ralph Stoop: Writing – review & editing, Writing – original draft, Methodology, Formal analysis, Conceptualization. Remo S. Schmidt: Writing – review & editing, Supervision, Project administration. Marie-Therese Fröhlich-Wyder: Writing – review & editing, Writing – original draft, Supervision, Project administration, Methodology, Conceptualization.

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## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Data availability

Data will be made available on request.

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