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To cite this article: M. Loudiyi & A. Aït-Kaddour (2018): Evaluation of the effect of salts on chemical, structural, textural, sensory and heating properties of cheese: Contribution of conventional methods and spectral ones, Critical Reviews in Food Science and Nutrition, DOI: [10.1080/10408398.2018.1455637](https://doi.org/10.1080/10408398.2018.1455637)

To link to this article: <https://doi.org/10.1080/10408398.2018.1455637>



Accepted author version posted online: 21 Mar 2018.



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Publisher: Taylor & Francis

Journal: *Critical Reviews in Food Science and Nutrition*

DOI: <https://doi.org/10.1080/10408398.2018.1455637>

**Evaluation of the effect of salts on chemical, structural, textural, sensory and heating
properties of cheese:**

Contribution of conventional methods and spectral ones

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Abstract

Chemical composition, sensory characteristics, textural and functional properties are among the most important characteristics, which directly relates to the global quality of cheese and to consumer acceptability. A number of factors including milk composition, processing

conditions and salt content, influences these properties. The past decades many investigations were performed on the possibilities to reduce salt content of cheese due to its adverse health effects, the current lifestyle and the awareness of the consumers for nutrition quality products. Due to the multiple potential effects of reducing NaCl (simple reduction or substitution) on cheese attributes, it is of utmost importance to identify and understand those effects in order to control the global quality and safety of the final product.

In the present review a collection of the different results and conclusions drawn after studying the effect of salts by conventional (e.g wet chemistry) and instrumental (e.g. spectral) methods on chemical, structural, textural, sensory and heating properties of cheese are presented.

Key words: cheese, salt, spectroscopy, rheology, sensory, chemometrics

1. Introduction

Cheese is one of the most popular food products in the world. Its popularity can be mainly attributed to its numerous end-use applications. It has proliferated to suit varied conditions and requirements, especially during the last decade.

Cheese can be used as a major component of a meal, as a dessert, as a component of other foods or as a food ingredient; it can be consumed without preparation or subjected to various cooking processes. According to Sørensen (2001), the process cheese is one of the leading dairy product variety in the world that is used as an ingredient in various food preparations (i.e. processed foods and food service). Indeed, several cheeses satisfy varied requirements in order to be used as suitable ingredients in various dishes from baby foods to baked products (Gunasekaran and Ak, 2003). As an ingredient, cheese is generally required to provide different flavor, texture and cooking characteristics. The flavor of cheese is a key quality attribute in most of the applications in which cheese is used as an ingredient. The vast array of

cheese varieties provides a diversity of flavors (e.g. savory, picante, tangy, nutty, sweetish and salty) that contribute to the foods in which they are used.

In spite of the different advantages mentioned above, some cheeses are also considered as containing too many sodium chloride (NaCl) and fat that can participate to health problems. Cheeses have a salt content between 0.4 and about 4 g/100 g, specific to the variety (IDF, 2017). It is recognized that NaCl in cheese is a contributor to the high level of Na consumption. For example, it has been reported that cheese contributes to about 4% Na intake in the U.K (Ash and Wilbey, 2010), 5% in Australia (NHMRC, 2003) and 9.2% in France (Meneton et al. 2009) which highlights the need to reduce NaCl in most cheeses.

However, reducing the amount of NaCl in cheese represents a great challenge. Indeed, in cheese manufacture, the importance of NaCl is well established (Guinee, 2004). In addition to having a recognized role in improving the flavor, texture, and color of cheese, salt is a preservative due to its abilities to reduce the water activity (i.e. a_w) and to inhibit the germination of microbial spores by the chloride ion (Eckner, Dustman, and Ryś-Rodriguez 1994, Erkmen, 2001, Guinee and Fox, 2004, Guraya, Frank, and Hassan 1998). Moreover, consumers identify the amount of salt as a characteristic of each type of cheese, but they are also concerned about having cheeses with lower amounts of sodium (Johnson and Paulus, 2008). Hence, importance should also be given to reducing the contribution of NaCl as a sodium-carrier without affecting quality involving at least the following attributes: appearance, flavor, texture, functionality, microbiological safety and nutritional value (i.e. chemical composition) (Fox and Cogan, 2004). Thereby, numerous investigations were carried out in order to reduce the Na levels of cheese. The simplest way identified is to reduce

the level of added NaCl (El-Bakry et al. 2011a, McCarthy et al. 2016, Olson 1982, Schroeder et al. 1988). Other options were also proposed. The first one consisting in the partial or complete substitution of NaCl by other salts as KCl (i.e. potassium chloride), $MgCl_2$ (i.e. magnesium chloride), $CaCl_2$ (i.e. calcium chloride) (Ayyash, Sherkat, and Shah 2012, Ayyash et al. 2011, Ayyash and Shah, 2011a, Kamleh et al. 2012, Lindsay, Hargett, and Bush 1982, Lu and McMahon, 2015, McMahon et al. 2014, Patel, Patel, and Lee 2013, Reddy and Martin 1993a). The second one dealing with the reduction of the salt levels in combination with flavor enhancers (Grummer et al. 2013), and the last one by using ultrafiltration and reverse osmosis retentate-supplemented milks to alter the mineral levels of cheeses (Karimi, Mortazavian, and Karami 2012, Kosikowski, 1983, Lindsay, Karahadian, and Amudson 1985, Ozturk et al. 2015).

Cheese is a complex system, and the reduction and substitution of NaCl on its attributes and properties (i.e. appearance, flavor, texture, functionality, microbiological safety and nutritional value) could not be characterized in a simple way. So, multiple analytical methods such as sensory, empirical and instrumental techniques were used to quantify and evaluate the effect of NaCl reduction or substitution on cheese attributes and properties.

During the last 15 years, several novel techniques, e.g. various spectroscopic methods, like NIR (i.e. Near Infrared), MIR (Mid Infrared) and fluorescence have attracted the attention of researchers (Grossi et al. 2012, Mariette 2009). Some of these methodologies can be applied in automated and online monitoring systems. They allow much faster response, the possibility to screen in a nondestructive manner a high number of products and to obtain relevant information on product characteristics (molecular structure, texture, color...). Their application on milk and dairy products has been described in several book, book chapters and review articles (Karoui and Debaerdemaeker 2007, Wehr and Frank 2004).

However, there has never been a thorough review gathering the most important studies on the use of conventional and instrumental methods for quality control of cheeses with reduced or substituted sodium content. Thus, the present review paper aims to provide a comprehensive overview of the applications of those methods to assess the effects of reducing sodium content and its substitution with other salts on cheese attributes and properties (i.e. chemical, physico-chemical, structural, textural and functional ones).

2. Wet chemistry analysis

Cheese composition is mainly controlled by the initial composition of the milk (which is modified by the method used for milk standardization) and the manufacturing process (e.g., pH at renneting and draining, size of curd particles, ripening temperature, method of salting) used for cheese making. Moreover, factors such as species and breed of animal, stage of lactation, and seasonality can all affect the initial milk composition and alter the quality and functional characteristics of cheeses. Food composition is the basis for determining the nutritional value and overall acceptance, as chemical components are intrinsic reasons affecting food quality and functionalities.

The impact of NaCl concentration and its substitution by other salts on the cheese composition is of great importance because dairy products are considered as important for some consumer groups (e.g. children, pregnant women and elderly due to their nutritional values) (Souza et al. 2011). Moreover, sodium reduction in cheese can assist in reducing overall dietary Na intake. In this context many research investigated, on different cheeses (e.g. Cheddar, Mozzarella, Emmental, Halloumi, Akawi), the effect of NaCl reduction and its

substitution on the concentration of cheese components like proteins, organic acids, moisture, minerals. The methods used for those investigations were e.g. classical wet chemistry, electrophoresis, gas chromatography, and HPLC.

For example, different studies tried to evaluate the effect of NaCl and its substitution on e.g. moisture, fat, protein and minerals (**Table 1**). One of the first investigations concerned the study of unsalted cheeses and as expected different authors reported higher moisture content for Cheddar (Kosikowski 1983, Reddy and Marth 1994a, 1993a, Schroeder et al. 1988, Thakur, Kirk, and Hedrick 1975) and Mozzarella (Guo et al. 1997) compared to salted cheeses. This conclusion was recently confirmed on hard type cheese (Sheibani et al. 2015), Feta (McMahon, Motawee, and McManus 2009), and again on Cheddar cheeses (Lu and McMahon 2015, Murtaza et al. 2014). Rulikowska et al. (2013) demonstrated that the effect of NaCl on cheese composition depends on the NaCl reduction rate. They noted an effect of Na reduction in Cheddar cheese composition after studying different NaCl content (0.50, 1.25, 1.80, 2.25, 2.50 and 3.00%, w/w -weight/weight-). It was observed that in general, decreasing NaCl content resulted in a decrease in fat, protein, ash, sodium and an increase in moisture and L-lactic acid contents. When NaCl was added at 1.80, 2.25, 2.50 and 3.00%, no significant differences were observed between cheeses. Nonetheless, compared with the other samples, cheeses with 0.5 or 1.25% NaCl have significant ($P < 0.05$) higher moisture and lower protein, NaCl, ash and sodium contents.

Concerning the NaCl substitution by KCl, Reddy and Marth (1993a) observed no significant differences in moisture and fat-in-dry matter contents between Cheddar cheeses prepared with NaCl, KCl, or mixtures of NaCl/KCl (2:1, 1:1, 1:2 and 3:4, w/w basis). They reported that Cheddar cheese can readily be produced without affecting its composition when one-third or more of the NaCl added to cheese curd is replaced with KCl. Nonetheless, it was reported that cheeses salted with NaCl/KCl mixture retained more NaCl than did cheeses

salted with NaCl, whereas cheeses salted with KCl retained more KCl than did cheeses salted with NaCl/KCl mixtures. In Halloumi cheeses, Ayyash and Shah (2011b) observed that cheeses containing mixtures of NaCl/KCl (3:1, 1:1 and 1:3, w/w basis) did not depict significant ($P > 0.05$) differences in moisture, protein, fat, and ash contents among the experimental cheeses at the beginning of storage (i.e. 0 day at 4°C). The same conclusions were reported for Kefalograviera cheese by Katsiari et al. (1998). They reported that cheeses made with mixtures of NaCl/KCl (3:1 or 1:1, w/w) did not exhibit any significant differences in compositional (e.g. moisture, protein, fat and salt) and physicochemical (i.e. pH and a_w) characteristics compared to the control at the beginning and throughout storage (5 to 180 days at 2-3°C). The same conclusions were reported by Ayyash et al. (2012), on experimental Akawi cheeses containing different salt mixtures (Na/Cl : 3:1, 1:1 and 1:3, w/w) for moisture, protein, fat, and ash contents among the storage period (0 to 30 days at 4°C). Nonetheless, significant differences were observed in WSN (Water Soluble Nitrogen) and phosphotungstic-Soluble Nitrogen (SN) between experimental cheeses during storage, in contradiction with Ayyash and Shah (2011b) study on halloumi cheese.

In 2006, Upreti, McKay, and Metzger (2006) examined the influence of salt-to-moisture (S/M) on organic acids and residual sugars (lactose, galactose) of Cheddar cheese during 48 weeks of ripening. Cheeses with high S/M have higher levels of unfermented lactose compared with cheeses with low S/M. This difference was associated to the differences in bacterial activity due to alteration in cheese a_w . The concentration of lactic acid was significantly influenced by S/M, time and the interaction of time and S/M. More lactic acid was produced in low S/M treatments compared with high S/M treatments. This difference has been associated to the influence of S/M on a_w (Marcos et al. 1981). In accordance with Bassit, Cochet, and Lebeault (1993) reporting that reduction of a_w leads to a decrease in the growth of lactic acid bacteria (LAB) and concomitant decrease in lactic acid production.

Thus, in their study (Upreti, McKay, and Metzger 2006), increase in lactic acid from curds to day 1 was larger in low S/M treatments compared with high S/M treatment, leading to a higher lactic acid concentration in cheeses during ripening in low S/M compared to high S/M treatments. The increase in lactic acid from curds to day 1 of ripening was associated to the largest drop in lactose from cheese curds after salting.

Concerning galactose, cheeses with low S/M have higher levels of galactose compared with cheeses with high S/M. The authors suggested that this difference was related to higher lactose hydrolysis or higher lactococcal activity of cheese with low S/M.

For citric and uric acids, cheeses with low S/M presented high concentration for citric and uric acids and lower one for acetic acid compared to cheeses with higher S/M. No significant differences were observed on formic and orotic acids.

Equivalent investigation in older cheeses (i.e. 30 days of ripening) was reported by Ayyash et al. (2012). They found differences in organic acid concentrations in Akawi cheese when K was used to replace Na. Higher concentrations of citric, lactic, and acetic acids were found in cheeses containing a (1:3) NaCl/KCl mixture compared with the control (100% of added NaCl). This difference in organic acids concentrations were assigned to the impact of probiotic and lactic acid bacterial (i.e. *Streptococcus thermophiles*, *Lactobacillus casei*, and *Lactobacillus acidophilus*) growth and metabolism in cheeses that presented significant differences at the same storage period. The same approach was recently performed by McMahon et al. (2014) during storage on full-fat Cheddar cheeses containing different salts NaCl, KCl and two other salts MgCl₂ and CaCl₂. Potassium substitution levels of 10, 25, 50, and 75% were compared with the standard cheese (1.7% NaCl) and the effect of including a low level (10%) of MgCl₂ or CaCl₂ was investigated as well as a low-salt cheese with 0.7% NaCl. Those authors studied cheese differences in organic acids (i.e. lactic, propionic, acetic and other acids) in conjunction with survival of LAB and nonstarter LAB (NSLAB). They

observed a greater decrease in organic acid concentrations as a function of Na content. When Na content of cheeses was reduced by 25% or more, a delay in the increase in propionic acid concentration was observed during ripening. This was attributed to synthesis of propionic acid by NSLAB and the NSLAB becoming dominant later during the aging process of such lower-Na cheese. They also reported that decreasing Na lead to higher lactic acid concentration in the cheese. Moreover, as the percentage of K replacement increased in the cheeses, a corresponding increase in lactic acid concentration was detected. This was assigned to a slower die-off or inactivation of the starter bacteria, as K is less inhibitory than Na. Nonetheless, including 10% Mg or 10% Ca, along with 40% K in the salting mixtures did not have any apparent major effect on changes in organic acids during storage (9 months).

The impact of Na and K salts on mineral content of cheeses was also evaluated. Paulson, McMahon, and Oberg (1998), Schroeder et al. (1988), and Loudiyi et al. (2017a) observed that Ca content remained the same on Mozzarella, Cheddar and Cantal-type cheeses containing selected levels of added NaCl ranging from 0 to 1%, 0 to 1.44% and 0.5 to 2% (w/w), respectively. Contradictory observation was reported during the investigation of Rulikowska et al. (2013) on Cheddar cheeses. Nonetheless, in this study, among all samples salted with different NaCl content, only Ca content in the 0.5% presented statistical differences ($P < 0.05$) from the control (3.0% NaCl). However, they noted a decrease in Ca with decreasing NaCl content. This result was assigned to additional losses of minerals after pressing due to the enhanced lactose metabolism.

In the above studies, soluble Ca was not determined, therefore, whether adding salt to cheese would cause mobilization of Ca from caseins and into solution remains uncertain. Pastorino, Hansen, and McMahon (2003) confirmed in their study the above results concerning total Ca content and noted also that soluble Ca was unaffected by adding salt to cheese. In contrast, in Cheddar cheeses with added NaCl ranging from 0.8 to 2.3% (w/w) and equal level of

moisture (37.6%), Moller et al. (2013) reported that the level of total Ca decreased significantly from 169 to 158 mmol/kg with increasing salt content. This is a confusing result, since the Ca concentration is determined by the quantity of colloidal calcium phosphate (CCP) lost from the curd, which in turn depends on 3 principal factors during cheese manufacture: (1) preacidification of the cheese milk, (2) pH at whey drainage, and (3) cooking treatment (Lucey and Fox 1993). The lower pH at whey drainage for low-salt versus high-salt cheese ($\Delta\text{pH} = 0.22$) would be expected to favor Ca solubilization (Lucey and Fox 1993). Thereby, additional Ca analysis of the whey at drainage, salting, and pressing would clarify the significance of the mechanisms involved.

Concerning the partial substitution of NaCl by KCl, no significant difference in total concentration of Ca among experimental cheeses at the same storage period was reported by Ayyash et al. (2012). In contrast, Loudiyi et al. (2017a) reported that the partial substitution of NaCl with KCl induced a significant difference in total Ca content. This may be attributed to the manufacturing process of Cantal-type cheeses that were ripened for 5 days at 9 °C and dry-salted with 1.5NaCl:0.5KCl and 1NaCl:1KCl. Those conditions differs from the study of Ayyash et al. (2012) on Akawi cheeses salted in brine solutions (at 10%): 3NaCl:1KCl (B), 1NaCl:1KCl (C), and 1NaCl:3KCl (D) and stored at 4 °C for 1 month. However, their results (Loudiyi et al. 2017a) still in accordance with Ayyash and Shah (2011b), on Halloumi cheese, reporting that total Ca content decrease with increasing K content.

pH is one of the most important parameter in cheese processing (e.g. curd coagulation and ripening). The pH decrease accelerates the rennet activity (Zoon, Van Vliet, and Walstra 1989), reduces the electrostatic repulsion between casein micelles and alters the distribution of Ca between the micelle and serum phases. Therefore, determining the effect of NaCl and its replacement by other salts on the pH is of great importance. In this context, Pastorino, Hansen, and McMahon (2003) in agreement with previous studies (Cervantes, Lund, and

Olson 1983, Guo et al. 1997, Kindstedt, Kiely, and Gilmore 1992) and in contrast to the results of Thomas and Pearce (1981), demonstrated that increasing salt content (NaCl) of cheese did not affect cheese pH. Nonetheless, Reddy and Marth (1994a) reported, for unsalted cheeses, lower pH values (0.07-0.14 unit less) compared to salted cheeses (NaCl, KCl or mixtures of NaCl/KCl) probably due to higher level of bacterial growth. Indeed, it was reported that bacteria in cheese without added salt, continue to grow and ferment lactose to lactic acid leading to decrease the pH (Reddy and Marth 1993a, Thakur, Kirk, and Hedrick 1975). Moreover, the authors (Reddy and Marth 1994a) noted that the mean pH values of cheeses salted with NaCl/KCl or KCl were lower (0.04 to 0.07 unit) than pH of cheese salted with NaCl. These results agree with those of Koenig and Marth (1982), who identified that Cheddar cheese prepared with NaCl/KCl mixtures had lower pH than cheese salted with NaCl. McMahon et al. (2014) reported equivalent conclusions, for Cheddar cheeses. They observed a faster initial (day 1 of storage) decrease in cheese pH with low NaCl or K substitution. This difference in pH was maintained until the end of ripening (9 months), when considering the pooled pH (average pH all other the ripening period).

On the contrary, other studies reported no effect or an increase in the cheese pH when NaCl was replaced partly or totally with KCl. For Cheddar cheese different authors (Fitzgerald and Buckley 1985, Lindsay, Hargett, and Bush 1982) reported similar pH in cheeses salted with various NaCl/KCl mixtures when compared with the control. Although, cheeses containing KCl showed slightly higher pH than those with NaCl. In accordance with Silva et al. (2017) reporting that Prato Cheeses with partial replacement of NaCl by KCl showed higher pH values when compared with the control after 30 days of storage. These results agree also with Ayyash et al. (2012) and Gandhi and Shah (2016). Those differences can be due to the higher pH of the KCl solution as compared to NaCl solution (Ayyash et al. 2012). Ayyash and Shah (2011b, 2011c) reinforced conclusions presented above, because they reported similar pH on

Mozzarella and Halloumi cheeses salted with various NaCl/KCl mixtures (3:1 and 1:1, w/w) when compared with the control. However, the pH values of cheeses salted with 1NaCl:3KCl (w/w) were identified as higher ($P > 0.05$) compared to the control. Those results highlighted, that the pH increased as the KCl content increased above 50%. These observations were recently confirmed by Felicio et al. (2016) and Loudiyi et al. (2017a) reporting similar pH on Minas and Cantal-type cheeses salted with 75/25%, and 50/50% NaCl/KCl compared to the control.

Proteolysis is one of the most important biochemical event that occurs in cheeses during ripening and it greatly affects the general final cheese quality (e.g. texture and flavor properties) (Fox and McSweeney 1996). As it is a keystone of cheeses quality, different investigations were reported concerning the effect of NaCl reduction and its substitution with other salts and especially with KCl.

Schroeder et al. (1988) investigated the effect of different amount of added NaCl (0 to 1.44%) on Cheddar cheese during 7 months of ripening. One of the main conclusions of this study was that cheese with the lowest NaCl content showed a significant ($P < 0.05$) increase in proteolysis. This finding was later confirmed by Kelly, Fox, and McSweeney (1996) on younger (22 weeks) Cheddar cheeses with different S/M (0.37 to 5.41%). The results of Kelly, Fox, and McSweeney (1996) were confirmed recently by Moller et al. (2013) and Rulikowska et al. (2013) on equivalent cheeses ripened during 9 months and 1, 14, 28, 56, 112 and 224 days respectively. Moller et al. (2013) reported that the degradation of both α_{S1} - and β -casein increased with NaCl reduction. A positive correlation was observed between NaCl content and α -casein, β -casein degradation (α S: $P = 0.002$; β -CN: $P = 0.01$) and with β -casein major breakdown products, γ 1- casein A2 ($P = 0.03$) and γ 3-casein ($P = 0.004$). Rulikowska et al. (2013) reported also in their study that overall decreasing NaCl has for consequence an increase in proteolysis and a concomitant reduction of cheese ripening. However,

contradictory results were reported by Baptista et al. (2017) and Loudiyi et al. (2017a) on Prato and Cantal-type cheeses, respectively. They reported that salt reduction (25 and 50%) did not affect proteolysis. This difference could be related to bacterial stress influenced by the combined effect of salt, acid, aw, and cold storage of cheese. Enzyme activity is also affected by salt level because of its relationship with aw (Rallu, Gruss, and Maguin 1996).

One of the first study on NaCl substitution with other salts was performed in 1985 by Fitzgerald and Buckley on Cheddar cheese. They investigated the effect of NaCl, MgCl₂, CaCl₂ and KCl, or 1:1 mixtures of NaCl/KCl, NaCl/CaCl₂ and NaCl/MgCl₂ on cheese proteolysis. They reported that substitution of KCl for NaCl (up to 50%) did not influence proteolysis rates in Cheddar cheese, whereas a 100% substitution did enhance proteolysis. Moreover, proteolysis was higher in the cheese salted with CaCl₂ and MgCl₂. The authors reported that extensive proteolysis in these cheeses may be the consequence of the low salt in moisture. In the same way, McMahon et al. (2014) reported recently that including 10% Mg or 10% Ca, along with 40% K in the salting mixtures of Cheddar cheese did not have any apparent major effect on changes in proteolysis during storage (9 months).

Compared to previous conclusions, contradictory results were reported by Reddy and Marth (1993a) on Cheddar cheeses. They noted that NaCl substitution did not significantly affect trichloroacetic acid-soluble Nitrogen (TCA-SN) and phosphotungstic-soluble nitrogen (PTA-SN) (as proteolysis indicators). Similarly, Katsiari et al. (2001a, 2000) reported no significant differences ($P < 0.05$) in proteolysis in Feta and Kefalograviera cheeses, respectively, made with NaCl/KCl mixtures, when compared to the control (only added NaCl). The same conclusions were drawn during the investigation of Halloumi cheese (Milci et al. 2005, Guven et al. 2008, Ayyash and Shah 2011b), White (Guyen and Karaca 2001) and Cantal-type cheeses (Loudiyi et al. 2017a). Ayyash and Shah (2011c) performed equivalent investigations on the effect of substitution of NaCl with KCl on proteolytic and angiotensin-converting

enzyme inhibitory activities in low-moisture Mozzarella cheese. These authors pointed out that salt treatment (3NaCl:1KCl, 1NaCl:1KCl, 1NaCl:3KCl) unaffected WSN, TCA-SN and total free amino acids during storage. Nonetheless, WSN and TCA-SN increased significantly during storage within a salt treatment.

In contrast, Silva et al. (2017) evaluated the effect of partial substitution of NaCl with KCl and the flavor enhancers addition (arginine, yeast extract and oregano extract) on probiotic Prato cheese. It was reported that, all cheeses with reduced sodium content and with added flavor enhancer showed increased proteolysis when compared to the regular sodium cheese. The authors related this difference, to more favorable environment due to the increased moisture content, considering water is essential for the microbial growth.

Concerning lipolysis, another enzymatic degradation observed during ripening, little information is available regarding the effects of salt in this attribute. Nevertheless, investigations of Thakur, Kirk, and Hedrick (1975) and Reddy and Marth (1993b) on Cheddar cheeses indicated that NaCl may inhibit lipolysis. Godinho and Fox (1981) reported that lipolysis in Blue type cheese, showed to be highest in NaCl concentrations from 4 to 6% and higher the concentrations of NaCl, the lower the lipolysis is. Similar results were found in Picante type cheeses and Portuguese cheeses, with variation in lipolysis depending on the concentration of NaCl. Usually the lipolysis appeared to increase in concentrations from 9 to 16% and to decrease in concentrations from 20 to 23% (Freitas and Malcata 1996). More recently, Rulikowska et al. (2013) depicted no statistical influence ($P > 0.05$) of NaCl on lipolysis (free fatty acid (FFA) content was between 908-1020 ppm) when the concentration of NaCl was between 0.5 and 3% (w/w). The above results could depict that the concentration of NaCl should be in a specific range in order not to affect lipolysis.

Lindsay, Hargett, and Bush (1982) studied Cheddar cheese manufactured with either NaCl or a 1:1 blend of NaCl and KCl at various final salt levels (1.25, 1.5, and 1.75%). Cheeses made

with NaCl and KCl blends exhibited higher lipolysis compared with the cheeses made with NaCl alone. In accordance with Fitzgerald and Buckley (1985) reporting extensive lipolysis (as measured by FFA development) in Cheddar cheese salted with $MgCl_2$, $CaCl_2$, or KCl compared to the cheeses salted with NaCl.

In contrast, Reddy and Marth (1994b) found no significant differences in the lipolysis when cheeses produced with NaCl, KCl or with mixtures of both salts were compared. In accordance with Katsiari et al. (2001b), who investigated the effect of different Na content, on lipolysis in reduced sodium Kefalograviera cheeses, by varying the salting processes (brine- and dry salting with NaCl (control) or a mixture of NaCl/ KCl (3:1 or 1:1, w/w). They reported that the partial replacement of NaCl with KCl did not significantly influence the lipolysis during Kefalograviera cheese aging (5, 25, 50 and 180 days). This was later confirmed when the partial substitution of NaCl by KCl (0, 25, and 50%) in probiotic Minas cheese was investigated by Felicio et al. (2016).

3. Rheology methods

A lot of research has been undertaken on the effects of reducing NaCl and its substitution, as a mean of providing insights for the development of reduced-salt cheeses with quality characteristics (flavor, texture and cooking properties) as close as possible to the control cheese (**Tables 2 and 3**). As reported in the previous part of the manuscript, salt reduction is generally paralleled by modification of the chemical characteristics of cheeses (e.g. moisture content, proteolysis) (Kelly, Fox, and McSweeney 1996, McCarthy et al. 2015, McMahon et al. 2014, Murtaza et al. 2014, Rulikowska et al. 2013). These changes coincide with significant influence on rheology and texture properties of cheese.

Guinee and Fox (2004) reported that final salted cheese presented a firmer shape, whereas salt-free cheese had a rather soft and creamy or pasty consistency. On the other hand, high concentrations of NaCl resulted in hard and brittle cheese, observed in cheeses such as Gaziantep (Kaya 2002) and Feta (Prasad and Alvarez 1999). In accordance with Cervantes, Lund, and Olson (1983) on Mozzarella cheese and Schroeder et al. (1988) on Cheddar cheese. They reported an increase in hardness and a decrease in cohesiveness when increasing salt content. The above results were confirmed by Pastorino, Hansen, and McMahon (2003) on Muenster cheese. They noted that, at salt contents above 0.5%, salt appears to further increase hardness and decrease cohesiveness of cheese. These variations were associated to the fact that increasing salt content, increases ionic strength. Increasing ionic strength can affect protein interactions at more than one level. More extensive short-range interactions, such as those involving increased hydration and thickness of strands in the protein matrix, could contribute to increase hardness of cheese. In contrast, weaker and/or decreased long-range interactions, and water loss from pockets throughout the cheese may lead to a less elastic cheese matrix, resulting in a decrease in cheese cohesiveness.

In Cheddar cheese, Rulikowska et al. (2013) investigated during ripening (1, 14, 28, 56, 112 and 224 days) the influence of incremental NaCl reduction (0.5 to 3.0% w/w) on a wide range of key aspects associated with Cheddar cheese quality such as textural properties using TPA. The results showed that firmness (hardness), fracture stress (toughness) and fracture strain (shortness/crumbliness) decreased with decreasing NaCl content and ripening time. Moreover, these parameters were significantly affected by NaCl addition levels, ripening time and their interaction. Decrease in these textural attributes was associated with increased proteolysis,

moisture-in-non-fat substance (MNFS) and lower S/M, which impact directly the cheese texture as previously reported (Guinee 2004, Kelly, Fox, and McSweeney 1996, Kishor and Thakur 2015, Mistry and Kasperson 1998, Schroeder et al. 1988, Thomas and Pearce 1981). However, the authors noted that it is also likely that casein hydration was involved. Indeed, as reported by Guinea (2004) and in accordance with Johnson (2000), addition of NaCl to cheese increases the water-holding capacity of the cheese matrix by increasing the hydration of the casein, the volume of the matrix and the casein solubilisation. Therefore, reducing casein hydration decreased the capacity of the cheese matrix to withstand deformation, resulting in decreased cheese hardness, toughness and shortness/crumbliness.

Recently Akkerman et al. (2017), investigated the effect of NaCl reduction in Danish brined semi-hard cheese on textural properties (firmness and compressibility) during cheese ripening (1-12 weeks). They demonstrated that cheese firmness increased and compressibility decreased linearly as the NaCl content increased. The increase in firmness was expected, as NaCl is a major contributor to the formation of a strong gel network (Guinee 2004, Mistry and Kasperson 1998, Schroeder et al. 1988). However, the authors concluded that it seems possible to reduce the NaCl content in semi-hard cheeses without compromising the textural properties by adjusting the DL-starter cultures and the chymosin, during cheese process.

In the same approach, Moller et al. (2013) investigated the effect of NaCl reduction on the textural properties of Cheddar cheese with different contents of salt (0.8, 1.3, 1.8 and 2.3%) and equal moisture, corresponding to S/M ratios of 2.3, 3.4, 4.6, and 6.0%. Rheological analysis (uniaxial compression) was performed by measuring the force during the compression of the samples. The parameters considered in their study were, Young's modulus (E), fracture stress (σ_f) (both describing cheese firmness/rigidity), and the deformation needed for fracture ($\epsilon_{H,f}$) (characterizing the cheese in terms of shortness/brittleness; the smaller $\epsilon_{H,f}$, the shorter the cheese). The results showed no effect on the cheese firmness with salt. Those

results indicated that if the moisture content of cheeses made with variable amounts of salt is kept constant; such softening effect may be avoided upon a decrease from 4.6 to 2.3% S/M. In contrast, slightly lower firmness/rigidity was found for high-salt cheese (6.0% S/M), which was related to a salting-in effect that, in combination with the Na-Ca exchange mechanism (Na ions may have displaced colloidal Ca in an ion-exchange effect), facilitated casein hydration and solubilization. Once solubilized, casein molecules or aggregates no longer contribute to the continuous casein network, which consequently loses strength and imparts a more viscous texture to the cheese (Guinee and Fox 2004, Lucey et al. 2003). Like firmness (E), the measure of shortness ($\epsilon_{H,f}$) revealed a high degree of similarity between low-, reduced-, and normal-salt cheeses, whereas the highest salt level (6.0% S/M) resulted in a significantly longer cheese texture. This difference was associated to the pH of high-salt cheese (pH = 5.36) compared with the other cheeses (pH = 5.10 to 5.17). Indeed, it was reported that the fracture strain is closely related to the pH, upon which cheese texture may be overall more dependent than on any other physicochemical parameter (Lawrence et al. 1987). The authors suggested that, analogous to the characteristic changes of casein micelles in milk upon a pH decrease from 5.4 to 5.2 (Roefs et al. 1985), the higher pH of high-salt cheese would appear to favor casein hydration and aggregate integrity, whereas colloidal calcium phosphate solubilization and casein dissociation proceeded in the lower pH cheeses, creating smaller and more compact casein aggregates. In accordance with Lawrence et al. (2004) reporting that such changes, along with parallel enzymatic degradation of the casein network, are responsible for the short texture of Cheddar. The above results depict that the rheological changes observed at high salt content seems mainly attributable to the direct and indirect effects of salt related to casein hydration and pH, respectively. In accordance with Euston et al. (2002) who noted an effect of the interaction between salt level and pH on the rheology of model cheeses. Indeed, in addition to the interaction effect of NaCl and ripening time on

cheese texture, the authors (Euston et al. 2002) demonstrated an interactive effect between the level of salt and the pH on the microstructure and rheology of cheeses. When the content of NaCl increased from 1.5 to 3.5% at pH 4.6, there was an increase in the degree of dilation of protein structures (indicating an increase in the level of hydration), which reduced the elasticity of cheese, making it more brittle (increase in the plastic property). A similar effect was observed on increasing the pH from 4.6 to 5.2 and maintaining the concentration of salt at 1.5%.

The effect of substituting NaCl with KCl on texture profile was reported in several studies. Kamleh et al. (2012) investigated the effect of partial replacement of NaCl with KCl (100% NaCl, 70/30% NaCl/KCl, 50/50% NaCl/KCl) on texture characteristics (adhesiveness, chewiness, cohesiveness, hardness and springiness) of fresh and matured Halloumi cheese using texture analyzer. They reported that salt treatment had a significant effect on chewiness, cohesiveness, and hardness, whereas age of cheese had a significant effect on springiness, chewiness and hardness. Moreover, significant interaction effect between salt and ripening was observed for chewiness, hardness and cohesiveness.

Ayyash and Shah (2011d) reported an effect for equivalent texture parameters (hardness, cohesiveness) and on gumminess and adhesiveness for Nabulsi cheese during storage. Nonetheless, they reported that adhesiveness did not change significantly, whereas cohesiveness increased and hardness decreased during storage. These variations were associated principally to increase in proteolytic activity that reduce the protein network which may decrease hardness and increase cohesiveness and adhesiveness (Johnson and Lucey 2006, Lawrence et al. 1987) depending on cheese process. Regardless of salt treatment, no significant difference was observed among experimental cheeses during most of the storage period. This finding is in accordance with other studies that reported no effect on textural profile of Cheddar (Fitzgerald and Buckley 1985), Halloumi (Ayyash et al. 2011), Akawi

(Ayyash et al. 2012), Feta (Katsiari et al. 1997) and Kefalograviera (Katsiari et al. 1998) cheeses.

Functional properties of heated cheese (e.g. meltability, amount of oil released, stretchability, browning) are also altered by the level of NaCl reduction (Ganesan et al. 2014, Henneberry et al. 2015, Mistry and Kasperson 1998) (**Table 2**).

Ma et al. (2013) investigated the blistering and browning of Mozzarella with different moisture and salt (from 1.07 to 1.60 % w/w) contents on a number of functional properties like meltability, free oil, viscoelasticity, transition temperature (i.e softening point) and stretching properties. The blistering and browning properties were evaluated using machine vision and image analysis techniques. The viscoelastic properties were determined by Small strain dynamic rheological method. Meltability and free oil were measured by modified Schreiber test. Stretchability was measured using a modified three-prong-hook test. The results showed that all Mozzarella samples showed similar browning appearances. This similarity was assigned to similar galactose contents known to be implied in the browning reaction. Nonetheless, Mozzarella cheeses with high salt concentration presented higher transition temperature ($\tan \delta = 1$; index of cheese melting point), elastic and stretching resistances, which resulted in smaller blisters on the pizza. It was proposed that cheese with higher transition temperature had a shorter time to flow. Moreover, the elastic and stretching resistances of the melted cheese restrained the size of the blisters and impeded their growth during heating.

El-Bakry et al. (2011a) investigated the influence of reducing NaCl (0–1.5% NaCl) content on, imitation cheese, functional properties (assessed by flowability and dynamic rheology tests). Flowability was measured in a conventional oven at 180 °C for 10 min (Mounsey and O’Riordan 1999) and dynamic rheology test was performed from 20 to 95 °C. The results showed that flowability was not significantly affected by the NaCl reduction. Regarding

viscoelastic parameters, the G' values were significantly lower at 0% NaCl in comparison to the standard NaCl level between 20 and 95 °C. The $\tan \delta$ profile at all levels of NaCl concentration was broadly similar, while the main effects of NaCl concentration were observed at temperatures above 40 °C. At this temperature, the values of $\tan \delta$ were higher at lower levels of NaCl. Moreover, the cheese melting point ($\tan \delta = 1$) progressively decreased from 66.11 to 55.44 °C on reducing the NaCl concentration from 1.5 to 0%.

In the same approach, El-Bakry et al. (2010) investigated the effect of emulsifying salts (ES: trisodium citrate (TSC) and disodium phosphate (DSP) at a ratio of 2.16:1 (w/w)) reduction on imitation cheese manufacture and functionality. Flowability was measured according to Mounsey and O'Riordan (1999), by using tubes containing cheese cylinders of 25 mm diameter and 20 mm height and placed horizontally in a conventional oven at 180 °C for 10 min. The results showed that reducing emulsifying salts decreased flowability. In comparison to standard ES, a reduction of up to 20% produced cheeses with slightly altered functionality and reducing ES by 40% increased flowability. At ES reduction above 40% the product obtained bore little resemblance to cheese.

McCarthy et al. (2016) investigated the effects of altering NaCl and fat followability using Schreiber method, of cheddar cheese manufactured with different levels of fat (33% (FF), 22% (RF) or 16% (HF), and salt 1.9% (FS), 1.2% (RS) or 0.9% (HS)) and ripened at 14, 30, 90, 150, 210 and 270 days. The results demonstrated that, in general, reducing salt in Cheddar cheese below critical levels (e.g., < 1.2%) impairs cooking properties. The results showed that the response to salt reduction depends on the extent of salt reduction and the variable studied. Indeed, significant interactive effect between salt and fat on the flow has been observed at approximately all ripening days. Salt significantly affected the mean flow over ripening, with the mean flow of the reduced- and half –salt variants of the FF, RF and HF cheeses being significantly higher than the corresponding FS variants. The increase in flow on reducing salt

level from 1.9% to 1.2% or 0.9% (w/w) concurs with the results previously published on Mozzarella containing 0.7 up to 1.9% salt (Ganesan et al. 2014, Henneberry et al. 2015). In addition, Henneberry et al. (2015) by using uniaxial extension on a TA-HDi Texture Analyser in the range of 90 to 95 °C, to mimic cooking temperatures on a pizza pie, reported that, reduced-salt and reduced-fat cheeses required higher work to extend the molten cheese.

Lawrence, Gilles, and Creamer (1983), tried to compare the effect of NaCl and Ca content on cheese functionality. They reported that NaCl affected cheese functionality to a lower extent than did Ca content. This finding can be seen when the observations of Olson (1982) are compared to those of Paulson, McMahon, and Oberg (1998). Indeed, Olson (1982) reported that higher salt content (2% compared with 1%) decreases the melting of Mozzarella cheese (with high Ca content, 0.7%). In contrast, in nonfat Mozzarella cheese (with low Ca content, 0.4%), Paulson, McMahon, and Oberg (1998) reported increased melting when the salt content increased from 0.14 to 0.4%, with further increases in salt content, up to 2.2%, having no effect on cheese melting. This difference could be assigned to the difference in the ionic strength. Indeed, as reported previously, adding salt to cheese increases the ionic strength. Moreover, in cheese with relatively high Ca content (0.7% for Mozzarella made by standard procedures), such as in the study of Olson (1982), this may lead to increased interactions between proteins that decrease cheese melting. However, in the study of Paulson, McMahon, and Oberg (1998), similar increase in salt content was at lower ionic strength because the cheese had low Ca content (0.4%). As a result, interactions between proteins did not increase significantly, and cheese melting was unaffected. Thus, a relatively small increase in the salt content of unsalted cheese with low Ca content seems to promote protein-to-water interactions. As a result, interactions between proteins are impaired and protein hydration increases, which results in increased melting of cheese.

Pastorino, Hansen, and McMahon (2003) in contrast to Olson (1982) and in accordance with Paulson, McMahon, and Oberg (1998) reported that cheese melting (using UW Meltmeter) was not significantly affected by increasing salt content (from 0.1 to 2.2%), of cheeses ripened for more than 24 days. Those conclusions agree with the result of Lawrence, Gilles, and Creamer (1983) who noted that salt content affected cheese functionality to a lower extent than did Ca content.

Alternatively, different authors investigated the effect of NaCl substitution by other salts on cheese functionality. El-Bakry et al. (2011b) studied the effects of different levels of substitution of the added sodium emulsifying salts (Na-ES) and NaCl with their KCl equivalents on the manufacture of imitation cheese. The authors demonstrated that imitation cheeses, in which sodium salts were fully or replaced partially with their potassium equivalent, could be manufactured successfully. However, slight difference was identified because, the replacement of sodium salts with potassium equivalents increased the flowability of cheeses. The authors suggested that this increase was associated to an increase in pH values, enhancing protein hydration and leading to a softer, less cohesive structure, with lower protein-protein interactions. Nonetheless, a difference in the water binding capacities of K compared to Na can also explain this difference. A weaker binding of the water molecules by the potassium compared to the sodium cations, led to a more plasticized matrix with increased flowability values. In the same approach, Hoffmann et al. (2012) studied the partial substitution of sodium by potassium in emulsifying salts of Cheddar like cheese. Different blends of emulsifying salts containing sodium polyphosphate, sodium and potassium citrate, and potassium phosphate were used. The best result, without loss functional properties (cheese melting point), was obtained with 0.39-0.55% sodium and 0.47-0.72% potassium.

Recently, in Cantal-type cheese, Loudiyi et al. (2017b) investigated the changes in dynamic rheological properties (G' , G'' and $\tan \delta$) during heating and cooling for the evaluation of cheeses with different salt contents (0.5% NaCl, 1% NaCl, 2% NaCl, 1.5/0.5% NaCl/KCl, 1/1% NaCl/KCl). It was reported that compared to the control, the heating of samples with low NaCl content generally present lower G' and G'' values, whereas an opposite trend was observed for cheeses containing KCl. The $\tan \delta$ values of all the cheeses were nearly always higher than those of the control. Moreover, between heating and cooling cycles G' , G'' and $\tan \delta$ of all cheeses during heating were lower than those observed throughout cooling. $\tan \delta = 1$ ($G' = G''$) was also used as an index of cheese melting and congealing points as reported by Mounsey and O'Riordan (1999) and Guinee et al. (2015). The results showed that the melting of cheese matrix during heating and its congealing during cooling occur at different temperatures. Thus, the comparison between cheeses suggested that decreasing NaCl content and increasing NaCl substitution by KCl decreased the melting and congealing points of investigated cheeses. In another study, the same authors (Loudiyi et al. 2017a) investigated variation of $\log(\eta^*)$ versus temperature during heating (from 20 to 60 °C) to determine the melting point of fat in Cantal-type cheeses. The results showed that compared to the control (2% NaCl) all cheeses exhibited significant difference except cheese salted with 1.5/0.5% NaCl/KCl.

4. Sensory analysis

The quality of cheese involves at least six attributes, appearance, flavor, texture, functionality, microbiological safety and nutritional value (Fox and Cogan 2004). Appearance, flavor and

texture are important criteria for acceptability of the cheese by the consumer. The flavor of cheese was considered as the most important attribute for most consumers but it is very much a matter of personal preferences between varieties and within a variety. The appearance of cheese is usually the only attribute the consumer can assess when purchasing and hence is critically important. Appearance includes color, the presence or absence, as appropriate for the variety, of mold and the presence or absence, as appropriate, of eyes or other openings. The cheese texture was defined as a composite sensory attribute resulting from a combination of physical properties that are perceived by the senses of touch (tactile, including muscular and mouth-feel) and sight.

As indicated above in this review, the decrease in NaCl content of cheeses can be conducted by reducing the amount of added NaCl (Ganesan et al. 2014, Mistry and Kasperson 1998, Rulikowska et al. 2013), the total or partial substitution of NaCl by other salts such as KCl, CaCl_2 or MgCl_2 (Grummer et al. 2012, Thibaudeau et al. 2015) or the addition of flavor enhancers such as hydrolyzed vegetable protein, yeast extract, disodium inosinate, and disodium guanylate (Grummer et al. 2013). However, all these substances may promote salty taste, but only NaCl offers what is really recognized as pure salty taste. The salty perception of NaCl is attributed either to the cation (70-85%) or to the anion (30-15%) (Formaker and Hill, 1988, Mattes 2001) and involves the passage of sodium ions through a narrow ionic channel, being almost impossible to find a similar substance, except toxic ions (lithium), that might pass through these channels (McCaughy 2007). Additionally, one of the most important functions of salt is to create a base for the perception of other flavors, that is, it works as an enhancer of taste in some cases and inhibitor in other, in addition to promoting higher perception of aromas (Kilcast and Rider 2007).

Various types of cheeses were developed with reduced sodium content by decreasing NaCl or partial/total substitution of this salt with other salts. The first technological intervention that

needs to be considered is typically to decrease the amount of added NaCl. Thus, the simple reduction of salt to levels that do not compromise the physicochemical and sensory quality of the product is a promising approach. Indeed, it was suggested by the group of specialists (Consensus Action on Salt and Health) that a reduction in the amount of salt between 10 and 25% cannot be detected by consumers. Fox and Walley (1971) and Lawrence and Gilles (1980) reported that S/M of 4 to 6 are usually recommended for best Cheddar cheese flavor development and preventing excessive proteolysis and bitterness. This finding was confirmed by Guinee and Sutherland (2011) reporting that, there is an ideal S/M for each cheese variety, and the sensory defects can be perceived only below that range, possibly due to the growth of undesirable bacteria and/or uncontrolled enzymatic activity.

In Cheddar cheeses, Lindsay, Hargett, and Bush (1982) reported that reduction of S/M from 4.9 to 3.5% did not significantly affect the flavor and texture of the product. Schroeder et al. (1988) showed non-detectable differences in Cheddar with reduced concentration of salt (from 4.1 to 3.1%). However, when the levels of NaCl were reduced to 0.7%, the cheeses became excessively viscous with acid, sour and unpleasant residual flavor, indicating an increase in proteolysis. Mistry and Kasperson (1998) reported no differences in flavor, when Cheddar type cheeses with reduced amount of fat and salt concentrations between 2.7% and 4.5% were compared. In cottage cheese, Wyatt (1983) reported that a reduction of 50% in the amount of NaCl resulted in low acceptance compared to the control. However, a reduction of 35% in the amount of NaCl (from 1 to 0.65%) did not influence the evaluation by consumers.

In the production of Edam type, Federal Dairy Research Center (1988) reported that cheese with low amount of sodium (concentration of sodium between 0.4 and 0.5%) also resulted in the product with satisfactory quality. Recently, Ganesan et al. (2014) concluded that consumers can distinguish even a 30% salt reduction on Mozzarella and Cheddar cheeses, and

a gradually phased sodium reduction is needed to improve acceptability of lower sodium cheeses.

Furthermore, Karahadian and Lindsay (1984) reported that salt-free Cheddar-type cheese demonstrated undesirable sensory characteristics when compared to cheeses with intermediate and conventional levels of salt. In accordance with Baptista et al. (2017) who studied the effect of salt reduction (25 and 50%) in Prato cheeses. They reported that Cheeses with 50% salt reduction were less firm and less sensory acceptable than the control cheese and the cheese with 25% salt reduction. In the study of Rulikowska et al. (2013), they reported that reducing NaCl adversely impacted Cheddar flavor and texture. Thus, cheese with lower salt content exhibited low level of flavor development and high level of bitterness. The increase of bitterness was associated with increased hydrolysis of β -casein (Guinee and O’Kennedy 2007). These results are in accordance with Moller et al. (2013), who studied the flavor and the texture parameters of Cheddar cheese with different NaCl levels (0.8, 1.3, 1.8 and 2.3%) and equal moisture. In addition to previous study, they concluded that the flavor of mature Cheddar with a constant moisture content deteriorated gradually upon a 50% salt reduction, whereas moisture regulation during manufacture contributed to largely unaffected textural properties.

Mooster (1980) reported that compared to Na, other cations (K, Mg and Ca) give sourness and less saltiness perception. Compared to chloride, other anions (phosphates and citrates) interfere more directly in taste, decreasing saltiness and leaving a residual metallic taste due to phosphate. Fitzgerald and Buckley (1985) reported that the use of $MgCl_2$, KCl and $CaCl_2$ at a concentration of 1.5% resulted in an unacceptable Cheddar cheeses, as they exhibited an extremely sour taste. At this concentration, they also presented high level of proteolysis and lipolysis. Both flavor and texture scores for the $CaCl_2/NaCl$ and the $MgCl_2/NaCl$ salted cheese were significantly ($P < 0.05$) lower than the controls. Thus, cheese salted with Ca and

Mg had a bitter metallic flavor and a flaky, crumbly, and greasy texture. However, cheese salted with the mixture (1:1 KCl/NaCl) resulted in a cheese comparable to standard (Fitzgerald and Buckley 1985). These results were confirmed by Grummer et al. (2012) reporting that Both CaCl_2 and MgCl_2 produced considerable off-flavors in the cheese (bitter, metallic, unclean, and soapy). While, KCl can be used successfully to achieve large reductions in sodium when replacing a portion of the NaCl in Cheddar cheese in accordance with Guinee and O’Kennedy (2007). In the production of Gruyere type cheese, Lefier et al. (1987) reported that cheese produced by replacing NaCl with MgCl_2 , (reducing the residual sodium by 80% and doubling the content of magnesium) showed a slight residual sour taste and alterations in the body (increased smoothness). However, the cheese was acceptable in the sensory analysis.

In the same way, Guinee (2004) reported that at high concentration ($> 1\%$) KCl caused considerable sourness perception and the alternative of replacing NaCl with KCl must be carefully studied. In 2007, Guinee and O’Kennedy (2007) reported that the proportions higher than 50:50 of NaCl/KCl, that is, 70:30 or 60:40, tend to be more attractive, as they reduce the sodium content satisfactorily and maintain the characteristic flavor of cheese. Recently, Thibaudeau, Roy, and St-Gelais (2015) confirmed that it is possible to reduce Na in Mozzarella cheese containing mixture of NaCl/KCl. However, regardless of the metallic taste detected for cheeses with a high K concentration, they reported that the substitution of KCl for NaCl should not exceed a ratio of 25%.

Nonetheless, according to Demott, Hitchcock, and Sanders (1984) on Cottage cheese with total substitution of NaCl with KCl or a partial substitution (50%) had similar acceptance levels between them and when compared to a standard cheese. In Kefalograviera type cheeses salted with NaCl (control) or a mixture of NaCl/KCl (3:1 or 1:1, w/w), Katsiari et al. (1998) reported also no significant differences in any aspects when compared to standard cheeses.

However, cheeses exhibited approximately 25 and 50% less sodium, respectively. In accordance with Quattrucci et al. (1997) reporting that a reduction in the salting period in Cacciota type cheese caused a decrease in sodium by 75% when compared to the control group. Furthermore, partial substitution of NaCl with KCl in the mass resulted in a cheese with 55% less sodium with no significant differences regarding flavor and texture.

Kamleh et al. (2012) studied the effect of NaCl reduction and partial replacement with KCl on the sensory characteristics of fresh and matured Halloumi cheese. The results obtained from their study revealed significant difference between cheeses for bitterness, crumbliness, and moistness, whereas age of cheese was significant for saltiness and squeakiness. Nonetheless, they reported that salt treatment had no significant effect on any of the acceptability variables for all Halloumi samples. Equivalent conclusions were reported by Patel, Patel, and Lee (2013) on processed cheeses, where the replacement of NaCl using KCl had a significant effect on sensory properties including hardness, bitterness and saltiness. The authors reported that KCl has high potential for use as a salt replacer without changing the chemical properties, but it has limited application for use in processed cheese because of short shelf life and low sensory qualities compared with the control. Moreover, the authors recommended the combination of 50% NaCl and 50% KCl for use as a salt replacer in the processed cheese with other ingredients like flavor enhancers, which can mask the bitter flavor produced by KCl. Indeed, in addition to the options reported above, there is also a great interest in adding flavor enhancers, compounds that activate the receptors in the mouth and throat, helping to reduce the level of salt (Brandsma 2006). Flavor enhancers are responsible for their umami, brothy, and savory taste which opens the opportunity to produce low sodium products with high saltiness intensity and can mask bitter flavour (Desmond 2006).

In this context, flavor enhancers such as monosodium glutamate (MSG), hydrolyzed vegetable protein, yeast extract, disodium inosinate, and disodium guanylate were tried by

Grummer et al. (2013) who investigated the partial (60%) replacement of NaCl with KCl with and without flavor enhancers. They reported that low-Na Cheddar cheeses made with NaCl and KCl blends were well liked by the sensory panelists and were comparable to the control cheese (which contained only NaCl). However, the type of flavor enhancers used either positively or negatively affected the quality of reduced-NaCl cheese. Some modified the flavor in a positive way in the judgment of consumers in a sensory panel, whereas another resulted in a significant reduction in liking scores. More recently, Silva et al. (2017) evaluated the effect of partial substitution of NaCl with KCl and the flavor enhancers addition (arginine, yeast extract and oregano extract) on five experimental probiotic Prato cheese as reported previously. Compared to the control cheese, lower score was observed in cheese without the flavor enhancers, since 50% w/w NaCl was replaced by KCl. However, a good performance in the consumer test was obtained principally by the addition of yeast extract and oregano extract. In contrast, a decrease overall liking of the cheese added with arginine was observed and was associated to the water retention in the cheese matrix caused by the arginine, which negatively affected the visual appearance of the product. In accordance with Felicio et al. (2016) on low sodium probiotic Minas cheese (the most popular cheese in Brazil), added with arginine, reporting that the addition of arginine positively affected cheese flavor and appearance. While with respect to the attribute texture, similar scores were observed for the cheese containing arginine and the control cheese.

A new approach by using experimental design (response surface methodology) has been reported by Khetra, Kanawjia, and Puri (2016) to reduce sodium content of Cheddar cheese by using potassium based salt replacer in combination with flavor enhancers and bitter blockers to mask the off-flavors and bitterness recognized in Cheddar cheese containing potassium salts. Three types each of salt replacer (Saloni Saloni K and KCl), flavor enhancer (hydrolyzed vegetable protein, yeast extract and IMP (5 nosine-50 monophosphate)) and

bitter blocker (Lysine, AMP (adenosine-50-mono- phosphate) and Glycine) were used in this study. Saloni and Saloni K are vegetable origin potassium rich salt replacers manufactured from the plant of *Salicorniabranchiata* and *Kappaphycusalvarezii*, respectively. The authors reported that low sodium cheese with 75% sodium chloride substitution, 2 g.L⁻¹ hydrolyzed vegetable protein and 300 mg.L⁻¹ AMP gave best results without sensory loss. However, the effect of these ingredients on ripening behavior of Cheddar cheese needs to be explored.

It is known that low-sodium cheeses often exhibit an acidic flavor due to excessive acid production during the manufacturing and the initial stage of ripening, which is caused by ongoing starter culture activity facilitated by the low S/M levels. In this approach, rather than replacing Na, Kosikowski (1983) proposed a different approach consisting in the fortification of cheese milk with UF (i.e. ultrafiltration) retentate for the manufacture of reduced-salt (~1%) Cheddar cheese. Control cheese (1% NaCl, with no UF retentate fortification) was acidic, bitter, pasty, and lacked typical Cheddar cheese flavor, whereas cheeses fortified with UF retentate exhibited good to excellent quality, probably due at least partially to their higher pH values. In the same approach, Ozturk et al. (2015) investigated, more recently, a combination of approaches involving cheese milk fortification with ultrafiltration UF retentates (to increase curd buffering), along with high-hydrostatic pressure (HHP) treatment (to reduce residual starter numbers) of cheese, on the properties of low-sodium Cheddar cheeses manufactured with fermentation produced camel chymosin. Camel chymosin was used as a coagulant to help reduce bitterness development (a common defect in low-sodium cheeses). Four treatments were used: one regular salt (2% NaCl) non-UF fortified, no HHP applied (R-Na); 3 low-salt (0.8% NaCl), non-UF fortified, no HHP applied (L-Na); UF-fortified, no HHP applied (L-Na-UF); and UF-fortified, HHP-treated (L-Na-UF-HHP; 500 MPa for 3 min applied at 1 day after cheese manufacture). The authors reported successfully that the L-Na-UF-HHP cheese did not significantly differ in bitterness and acidity from R-Na

cheese during ripening. Moreover, it was reported that HHP treatment lead to reduce the rate of proteolysis and microbial numbers which could help to provide increase shelf-life of L-NA cheese. Thus, the combination of HHP and UF retentate fortification were helpful approaches to improve the quality of L-Na cheese.

5. Spectroscopic methods

VIS-NIR (i.e. Visible Near Infrared), MIR, and fluorescence spectroscopies are probably the most used techniques for rapid, accurate, and reliable characterization of cheese properties (molecular structure, molecular interactions, authentication, melting...) without preparation and destruction of the sample.

The MIR range is a privileged spectroscopic region to study molecular vibrations because precise and directly accessible information concerning X-H chemical bonds (X: C, H, O, and N) can be depicted. The potential of MIR spectroscopy for monitoring and charactering cheese origins, chemical parameters, texture and physicochemical modifications during ripening have been the subject of several research (Dufour et al. 2000, Irudayaraj, Chen, and McMahon 1999, Karoui et al. 2004, Kulmyrzaev et al. 2005). Dufour et al. (2000) demonstrated that MIR spectroscopy has the ability to discriminate between Cheddar cheese samples at various ripening stages. Irudayaraj, Chen, and McMahon (1999) also investigated the potential of this method to follow texture development in full-fat and reduced-fat Cheddar cheese during ripening. They reported that texture parameters were correlated with the absorbance of peak areas corresponding to the reactive groups of fat, protein and water. Moreover, models predicting sensory texture attributes were reported by Fagan et al. (2007a) for processed cheeses varying from approximate to excellent depending of the parameter considered. Coefficient of determination varying from 0.66 up to 0.81 was reported for hardness, springiness, mass-forming, and mouth-coating; while an excellent model was provided for the fragmentable parameter ($R^2 > 0.91$). In another study, the same authors

(Fagan et al. 2007b) demonstrated that MIR spectroscopy coupled with chemometrics was successfully used to develop calibration models to predict instrumental texture and meltability attributes of processed cheese samples. In the same approach, Boubellouta and Dufour (2012) showed that MIR spectroscopy is a useful tool to delineate the molecular structure changes of cheese matrices during melting. In addition, it was possible to derive the melting temperatures of cheese fats and cheese matrices from MIR spectral data. Those studies demonstrated that MIR spectroscopy is a suitable method for analyzing texture and molecular structure of cheeses.

Nonetheless, as far as we know very few studies investigated the potential of MIR to study molecular structure and functionality modifications of cheeses depending on salt content by MIR spectroscopy. Recently, Loudiyi and Aït-Kaddour (2017) investigated the ability of MIR spectroscopy coupled with Independent Components Analysis (ICA) to monitor the molecular structure changes of model cheeses (Cantal-type cheese) with different salt content during gentle heating (20 to 60 °C) and ripening (5 and 15 days). Their investigation suggested that decreasing NaCl content of cheese (2 to 0.5%) decreased the fat and the cheese melting temperatures and increasing the NaCl substitution by KCl (0.5 to 1%) tended to decrease and increase the cheese melting and the fat melting temperatures, respectively. The authors successfully demonstrated that MIR spectroscopy has a huge potential to delineate through ripening and heating the molecular structure changes of cheeses with different salt contents. Moreover, similar fat melting temperatures for each kind of cheese were obtained regardless the technique used (dynamic rheology and MIR spectroscopy); while MIR spectroscopy showed its limit to predict the melting temperature of cheese matrix.

Fluorescence concerns emission of lower energy light by a fluorescent molecule (i.e. fluorophore) following the absorption of UV or VIS light. Fluorophores encountered in food products are generally substances which possess one or more conjugated bonds. Fluorophores

are hydrophilic, hydrophobic or even amphiphilic molecules (Herbert 1999, Lakowicz 1999). Fluorescence spectroscopy, offers several advantages for the characterization of molecular interactions and reactions, and molecule environments. For examples, caseins contain the amino acid tryptophan, fat globules contain the Vitamin A, the riboflavin was also in presence, and fluorescent Maillard reaction products were also reported. The fluorescent properties of tryptophan can give information on its surrounding environment. The fluorescence of this amino acid in a hydrophobic environment is different from its fluorescent properties when it is in a hydrophilic environment (Lakowicz 1983). Consequently, this method was used to investigate changes of the structure of milk components and their interactions during heating and acidification (Boubellouta and Dufour 2008). This method was also used for structural changes during cheese melting (Boubellouta and Dufour 2012, Karoui, Laguet, and Dufour 2003), for predicting some chemical components of cheeses (Abbas, Karoui, and Ait-Kaddour 2012) and for characterizing molecular structure and texture properties of cheeses (Karoui and Dufour 2006, Tang et al. 2008). Most of the studies, that investigated the potential of fluorescence spectroscopy to studies cheeses, were performed in classical front-face fluorescence (FFF) and synchronous fluorescence (SF) excitation modes. SF spectroscopy, as reported by different authors, presents an interesting advantage compared with FFF spectroscopy. Indeed, compared with a FFF spectroscopy emission or excitation spectrum that is mainly specific to a fluorophore a SF spectrum generally depicts bands related to multiple fluorophores (Aït-Kaddour et al. 2016, Boubellouta and Dufour 2008, Divya and Mishra 2007).

Concerning fluorescence, only two studies, conducted by the same research team (Loudiyi et al. 2017a, Loudiyi et al. 2017b) were investigated to assess the effect of NaCl and its substitution by KCl on molecular structure and functional properties (melting and congealing) on model Cheddar like cheese. The authors successfully demonstrated that fluorescence

spectroscopy has a huge potential to delineate the molecular structure changes of cheeses depending on the added salts (NaCl and KCl). Moreover, As reported in the second section (Rheology methods), Loudiyi et al. (2017b) suggested that decreasing NaCl content (2 to 0.5%) and increasing NaCl substitution by KCl decreased the melting and congealing points of investigated cheese. However, a slight decrease was noted in terms of melting and congealing values of cheeses obtained with the fluorescence method compared to those obtained with the rheological method. This suggested that fluorescence showed its limit to predict the melting temperature of cheese matrix. This finding is in accordance with Loudiyi and Aït-Kaddour (2017), reporting also that MIR spectroscopy showed its limit to predict the melting temperature of cheese matrix.

6. Conclusions and future trends

An effective approach for the production of cheese with reduced sodium content is an important issue for the dairy industry as there is a public health concern regarding the use of high level of sodium. This review has documented the current state of art in relation to the influence of salt reduction on cheese quality, as well as an overview of conventional and spectroscopic methods for quality control of cheeses with reduced sodium content.

Potassium chloride or a mixture of NaCl and KCl with NaCl have been the most widely and successfully used strategy for partial replacement for NaCl in cheeses. Although most of the results showed that the substitution of NaCl for mixtures of 50:50% of NaCl and KCl normally does not cause any biochemical, textural or structural changes, some discrepancies among studies report that the use of mixtures with that proportion affects the sensorial quality of cheese.

Many analytical methods were used to quantify and evaluate the effect of NaCl reduction or substitution on cheese attributes and properties. Traditional wet chemistry, for compositional analysis was widely used in several studies. However, this method is labour intensive, time

consuming and require sample destruction. The use of approach based on sensory and instrumental methods appears pertinent to better understanding the effect of salt on cheese quality. However, many of these methods are either time consuming, destructive or require trained personnel, and are therefore not suited for online or large-scale operations.

Several studies demonstrated the value of further integration and implementation of modern instrumental analysis in the quest for a more detailed understanding of cheese structure and functionality. Recent studies showed that spectroscopic techniques such as fluorescence and MIR can be successfully applied for rapid, accurate, and reliable characterization of cheese properties (molecular structure, molecular interactions, authentication, melting...) without preparation and destruction of cheese samples. The potential role of these technologies in monitoring the molecular structure of cheeses seems very promising. This capability may be useful for dairy food analysis at the laboratory research scale and further for the development of both laboratory and online monitoring technologies in the dairy industry.

Future research could focus on considering the suitability of spectroscopic techniques to monitor the molecular structure, to predict functional properties and the use of more flavor enhancers to improve the quality of cheese with reduced sodium. The spectroscopic methods in 1D (classical spectroscopy) and 2D (e.g. fluorescence, MIR, NMR spectroscopic images) configurations could help to understand the effect of salt on the molecular and microstructure properties of cheese and help the dairy industries to develop low salt cheeses without decreasing its technological, nutritional and sensory qualities. Reducing sodium in cheese, while maintaining quality and safety, continues to be a challenge for the dairy industry, worldwide.

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Table 1. Example of the principal conclusions of studies concerning the effect of NaCl reduction and/or substitution on physicochemical properties of cheeses

| Cheese product | Principal conclusion | Reference |
|---|--|--|
| Cheddar, Mozzarella, Hard type- cheese, Feta | - Decreasing NaCl content resulted in an increase in moisture content | Guo, Gilmore, and Kindstedt 1997, Kosikowski 1983, Reddy and Marth 1994a, 1993a, Schroeder et al. 1988, Thakur, Kirk, and Hedrick 1975 |
| Kefalograviera, Halloumi, Akawi | - Cheeses made with the mixtures of NaCl/KCl 3:1 or 1:1 did not exhibit any significant difference in moisture, protein and fat contents at the same storage period | Ayyash, Sherkat, and Shah 2012, Ayyash and Shah 2011a, Katsiari et al. 1998 |
| Cheddar | - Cheeses with low salt/moisture ratio have lower lactic acid and higher galactose contents - Decreasing Na and increasing K replacement levels in the cheese lead to higher lactic acid concentration - Unsalted cheeses have lower pH values compared to salted cheeses with NaCl, KCl or mixtures of NaCl/KCl - Cheese salted with NaCl/KCl or KCl had lower pH compared to cheese salted with NaCl - Decreasing NaCl content increased proteolysis rates | Bassit, Cochet, and Lebeault 1993, Upreti et al. 2006 McMahon et al. 2014 Reddy and Marth 1994a, 1993a, Thakur et al. 1975 Koenig and Marth 1982, McMahon et al. 2014, Reddy and Marth 1994a Kelly, Fox, and McSweeney 1996, Moller et al. 2013, Rulikowska et al. 2013, Schroeder et al. 1988 |
| | - Substitution of KCl for NaCl up to 50% did not influence proteolysis rates, whereas a 100% substitution did enhance proteolysis rates | Fitzgerald and Buckley 1985 |
| | - Cheeses made with NaCl/KCl mixtures or KCl alone exhibited higher lipolysis compared with the cheeses made with NaCl alone | Fitzgerald and Buckley 1985, Lindsay, Hargett, and Bush 1982 |
| Akawi | - Cheese made with 3:1 NaCl/KCl mixture has higher concentrations of lactic, citric and acetic acids compared to the control | Ayyash et al. 2012 |
| Mozzarella, Cheddar, Cantal | - Ca content remained the same in cheeses containing selected levels of added NaCl ranging from 1 to 2% | Loudiyi et al. 2017a, Paulson, McMahon, and Oberg 1998, Schroeder et al. 1988 |

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| Muenster, Cheddar, Akawi, Halloumi | - Soluble Ca was unaffected by adding salt to cheese | Ayyash et al. 2012, Ayyash and Shah 2011b, Pastorino, Hansen, and McMahon 2003, Ozturk et al. 2015 |
| Halloumi, Cantal | - Total Ca content decreased with increasing KCl content | Ayyash and Shah 2011b, Loudiyi et al. 2017a |
| Muenster, Mozzarella | - Increasing NaCl content did not affect cheese pH | Cervantes, Lund, and Olson 1983, Guo, Gilmore, and Kindstedt 1997, Kindstedt, Kiely, and Gimore 1992, Pastorino, Hansen, and McMahon 2003 |
| Mozzarella, Halloumi, Cantal Minas Prato, Cantal | - Increasing KCl content above 50% increased cheese pH | Ayyash and Shah 2011b, 2011c, Felicio et al. 2016, Loudiyi et al. 2017a |
| Cheddar, Kefalograviera, Halloumi, White, Cantal, Mozzarella | - NaCl reduction (25 and 50 %) did not affect proteolysis rates | Baptista et al. 2017, Loudiyi et al. 2017a |
| | - Cheeses made with NaCl/KCl mixtures showed no significant differences in proteolysis rates compared with the control containing only NaCl | Ayyash and Shah 2011b,c, Guven et al. 2008, Guven and Karaca 2001, Katsiari et al. 2001a 2000, Loudiyi et al. 2017a, Milci et al. 2005, Reddy and Marth 1993a |
| Bleu, Cheddar, Picante, Portuguese | - Increasing NaCl content may inhibit and decrease lipolysis | Freitas and Malcata 1996, Godinho and Fox 1981, Reddy and Marth 1993b, Thakur, Kirk, and Hedrick 1975 |
| | - The concentration of NaCl should be in a specific range in order not to affect lipolysis | |
| Cheddar, Kefalograviera Minas | - Cheeses produced with NaCl, KCl or with mixtures exhibited no significant differences in lipolysis compared to the control | Felicio et al. 2016, Katsiari et al. 2001b, Reddy and Marth 1994b |

Table 2. Examples of principal conclusions of studies concerning the effect of NaCl reduction and/or substitution by other salts on textural and functional properties of cheeses

| Cheese product | Principal conclusion | Reference |
|---|---|--|
| Gaziantep, Domiati, Feta, Cheddar, Mozzarella, Muenster | - Increasing NaCl content increased hardness and decreased cohesiveness | Cervantes, Lund, and Oslon 1983, Guinee and Fox 2004, Pastorino, Hansen, and McMahon 2003, Schroeder et al. 1988 |
| Nabulsi, Cheddar, Halloumi, Akawi, Feta, Kefalograviera | - Cheeses made with NaCl/KCl mixtures exhibited no significant difference on texture parameters hardness, cohesiveness, gumminess and adhesiveness | Ayyash et al. 2012, 2011, Ayyash and Shah 2011d, Fitzgerald and Buckley 1985, Katsiari et al. 1997, 1998 |
| Cheddar | - Firmness, hardness, toughness and shortness/crumbliness decreased with decreasing NaCl content and ripening time | Rulikowska et al. 2013 |
| | - Salt reduction with equal moisture content showed no effect on the cheese firmness, shortness while the highest salt level 6.0% S/M resulted in a significantly longer cheese texture | Moller et al. 2013 |
| | - Reducing salt below critical levels e.g., < 1.2% impairs cooking properties | McCarthy et al. 2016 |
| | - Reducing salt level from 1.9 to 0.9% increased flowability | |
| Mozzarella | - Cheeses with NaCl content (1.6 %) presented higher transition temperature ($\tan \delta = 1$), elastic and stretching resistances | Ma et al. 2013 |
| | - Reducing salt level from 1.9 to 0.7% increased flowability | Ganesan et al. 2014, Henneberry et al. 2015 |
| | - Higher NaCl content from 1 to 2% | Olson 1982 |

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| | decreased cheese melting point | |
| Non fat Mozzarella cheese | <ul style="list-style-type: none"> - Increasing NaCl content from 0.14 to 0.4%, increased cheese melting point Paulson et al. 1998 - Increasing NaCl content, up to 2.2%, had no effect on cheese melting point | |
| Imitation cheese | <ul style="list-style-type: none"> - Flowability and $\tan \delta$ profile were unaffected by NaCl reduction 0–1.5% El-Bakry et al. 2011a - G' values were significantly lower at 0% NaCl in comparison to the standard NaCl level between 20 and 95°C - Cheese melting point decreased with decreasing NaCl content - The replacement of Na salts with K equivalents increased flowability El-Bakry et al. 2011b | |
| Cantal | <ul style="list-style-type: none"> - Decreasing NaCl content and increasing NaCl substitution by KCl decreased the melting and congealing points Loudiyi et al. 2017a | |
| Danish | <ul style="list-style-type: none"> - Cheese firmness increased and compressibility decreased linearly as the NaCl content increased Akkerman et al. 2016 | |
| Muenster | <ul style="list-style-type: none"> - Cheese melting was not affected by increasing NaCl content from 0.1 to 2.2% Pastorino, Hansen, and McMahon 2003 | |
