

## THE EFFECT OF INDIVIDUAL PHOSPHATE EMULSIFYING SALTS AND THEIR SELECTED BINARY MIXTURES ON HARDNESS OF PROCESSED CHEESE SPREADS

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### ABSTRACT

The aim of this work was to observe the effects of emulsifying salts composed of trisodium citrate and sodium phosphates with different chain length (disodium phosphate (DSP), tetrasodium diphosphate (TSPP), pentasodium triphosphate (PSTP) and sodium salts of polyphosphates with 5 different mean length ( $n \approx 5, 9, 13, 20, 28$ )) on hardness of processed cheese spreads. Hardness of processed cheese spreads with selected binary mixtures of the above mentioned salts were also studied. Measurements were performed after 2, 9 and 30 days of storage at 6 °C. Hardness of processed cheese increased with increase in chain length of individually used phosphates. Majority of applied binary mixtures of emulsifying salts had not significant influence on hardness changes in processed cheese spreads. On the other hand, a combination of phosphates salts (DSP with TSPP) was found, which had specific effect on hardness of processed cheese spreads. Textural properties of samples with trisodium citrate were similar compared to samples with DSP.

**Keywords:** emulsifying salt; processed cheese spread; hardness; optical density

### INTRODUCTION

Processed cheese are dairy products made by heating cheese, emulsifying salts, butter and water until a homogenous mass is formed. Wide range of dairy products (cream, anhydrous milk fat, curd, milk powder, whey powder, caseinates, etc.) and non-dairy ingredients and additives (hydrocolloids, colouring, sensory active mixtures etc.) could be used during processed cheese production (Carić & Kaláb, 1997; Lee et al., 2004; Shirashoji et al., 2010). The production of processed cheese is composed of this six steps: i) ingredients formation (with respect to the desired parameters of the final product) ii) ingredients cleaning, milling and cutting iii) blending all ingredients in melting device iv) the melting process (melting temperature between 85-105 °C with a dwell time of several minutes) v) packaging vi) cooling and storing (Kapoor & Metzger, 2008).

In processed cheese matrix, fat emulsion and water binding provide proteins. In natural cheese, caseins do not possess their emulsifying properties, because they are bound in three-dimensional calcium bond matrix. Therefore emulsifying salts are used during processed cheese production. They are capable of exchanging ions and from insoluble calcium paracaseinate more soluble sodium paracaseinate is created, which possesses emulsifying and stabilization function in processed cheese matrix (Kapoor & Metzger, 2008; Cunha & Vuitto, 2010). Emulsifying salts are able to change textural properties of processed cheese during cooling and storing process. It is a complex of interactions including calcium bridges, disulphide bridges, hydrophobic interactions,

electrostatic interactions, hydrogen bonds, calcium-phosphate complexes etc. (Lee et al., 2004; Shirashoji et al., 2006; Dimitreli & Thomereis, 2008; Kapoor & Metzger, 2008).

The ability to exchange ions and form three-dimensional structure during cooling is different for individual emulsifying salts and decreases in following order: polyphosphates > triphosphates > diphosphates > monophosphates  $\approx$  citrates (Abdel-Hamid, 2010; Sádliková et al. 2010; Buňka et al. 2013).

El-Barky et al. (2011) declares that citrates have higher ability to exchange ions than monophosphates. Longer phosphates lead to a higher dispersion of casein due to more intensive ability of ion exchange. The more dispersed casein are, the more these proteins show their emulsifying and hydrating properties and stabilize oil and water present in the mixture. The increasing range of the hydration process of proteins and emulsifying fats leads to an increase in the intensity of interactions in the melt and crosslinking casein, resulting in processed cheese with higher hardness (Mizuno & Lucey, 2005; Shirashoji et al., 2010; Bayarii et al., 2012).

Emulsifying salts in their binary mixtures have different properties and effect on processed cheese. Weiserová et al. (2011) and Kalliappan & Lucey (2011) declared that there exist a specific ratio between monophosphates and diphosphates (1:1-3:4) that strongly supports gel-formation and therefore the processed cheese has the highest hardness value. Polyphosphates in binary mixtures are supposed to inhibit gel formation. Polyphosphates are

strong bind to complexes with caseins and increase the intensity of negative charge on casein chain. It leads to increase in repulsive force between caseins and gel formation (especially hydrophobic interactions) is weakened (Lee & Klostermeyer, 2001; Mizuno & Lucey, 2005).

Over the past few years a number of studies have been published (e. g. Awad et al., 2002; Weiserová et al., 2011; Buňka et al., 2012; 2013) where the influence of individual or binary and ternary mixtures of emulsifying salts on processed cheese textural properties was described. But these works are limited on phosphate emulsifying salts including polyphosphate with mean-length  $n \approx 20$ . Sodium salts of polyphosphate with different chain length are supposed to be practised. Intensity of ion exchange and therefore casein dispersion could be influenced with different length of polyphosphate chain. Effect of citrate and phosphate salts on pH values is not in previously literature described. The role of individual polyphosphate with different chain length or their binary mixtures with trisodium citrate, disodium phosphate, tetrasodium diphosphate and pentasodium triphosphate on casein dispersion has not been published before.

The aim of this work was to describe the influence of individual emulsifying salts composed of citrates and phosphates with different chain length and their selected binary mixtures on hardness of processed cheese spreads. The second aim of this work was to study the effects of individual and selected binary mixtures of emulsifying salts on optical density in the model milk system dispersion.

## MATERIAL AND METHODOLOGY

### Processed cheese production

Model samples of processed cheese spread ( $\approx 40\%$  w/w dry matter content,  $\approx 50\%$  w/w fat in dry matter content) were produced from Dutch-type cheese blocks ( $\approx 50\%$  w/w dry matter content and  $\approx 30\%$  w/w fat in dry matter content, 7-week maturity), butter ( $\approx 82\%$  w/w dry matter content and  $\approx 80\%$  w/w fat content), water and emulsifying salts (Fosfa, Břeclav-Poštorná, Czech Republic). Added emulsifying salts were trisodium citrate (TSC), disodium phosphate (DSP), tetrasodium diphosphate (TSPP), pentasodium triphosphate (PSTP) and five sodium salts of polyphosphate (POLY<sub>xx</sub>) with different medium-length ( $n \approx 5, 9, 13, 20$  and  $28$ ; signed POLY05, POLY09, POLY13, POLY20 and POLY28, respectively). Binary mixtures of emulsifying salts composed of DSP:TSPP, DSP:PSTP; DSP:POLY<sub>xx</sub> (POLY05, POLY09, POLY13, POLY20 and POLY28), TSPP: POLY<sub>xx</sub> (POLY05, POLY09, POLY13, POLY20 and POLY28), TSPP:PSTP, PSTP:POLY<sub>xx</sub> (POLY05, POLY09, POLY13, POLY20 and POLY28), TSC: DSP, TSC:TSPP; TSC:PSTP, TSC:POLY20. The total applied concentration of emulsifying salts was  $3\%$  (w/w) and the added amount was calculated on total weight of the melt.

For model processed cheese production Vorwerk Thermomix TM 31-1 (Vorwerk & Co. GmbH, Wuppertal, Germany) was used. The melting temperature was  $90\text{ }^\circ\text{C}$  and was kept for 1 minute.

The same procedure was used in the work by Lee et al. (2004), Buňka et al. (2012; 2013) and Lee et al. (2013). The hot melt was put into polypropylene cups of cylindrical shape (52 mm in diameter; 50 mm high) and sealed with aluminium lids. Samples were cooled on temperature  $6 \pm 2\text{ }^\circ\text{C}$  during 2 hours after production and kept at this temperature until analyses started.

### Model milk dispersion samples preparation

Model samples of milk dispersion were prepared from skim milk powder (Moravia Lacto PLC, Jihlava, Czech Republic) and deionized water in an amount corresponding with  $5\%$  (w/v) solution. The mixture was carefully stirred for 1 hour and dissolved by  $22 \pm 1\text{ }^\circ\text{C}$  and then the sodium azide ( $0.2\%$  (w/v)) was added. Then pH of the solution was adjusted to  $5.80 \pm 0.01$  using  $1\text{ mol/L}$  or  $0.1\text{ mol/L}$  HCl. Emulsifying salts were added after 18 hours of stirring by  $22 \pm 1\text{ }^\circ\text{C}$ . Applied concentration was  $0.3\%$  (w/v) and composed of trisodium citrate (TSC), disodium phosphate (DSP), tetrasodium diphosphate (TSPP), pentasodium triphosphate (PSTP) and five sodium salts of polyphosphate (POLY<sub>xx</sub>) with different medium-length ( $n \approx 5, 9, 13, 20$  and  $28$ ; signed POLY05, POLY09, POLY13, POLY20 and POLY28, respectively). Binary mixtures were created from DSP:TSPP; DSP:POLY<sub>xx</sub> (POLY05, POLY09, POLY13, POLY20 and POLY28); TSPP:POLY<sub>xx</sub> (POLY05, POLY09, POLY13, POLY20 and POLY28); TSC: DSP; TSC:TSPP and TSC:POLY20. After 10 minutes of mixing the pH of the solution was adjusted again on  $5.80 \pm 0.01$  using  $1\text{ mol/L}$  or  $0.1\text{ mol/L}$  HCl. The mixture was finally stirred for 50 minutes at  $22 \pm 1\text{ }^\circ\text{C}$ . Optical density was measured at wave length  $700\text{ nm}$  (UV VIS Spectrofotometer, UV Mini 1240, Germany) according to Kalliappan & Lucey (2011). The results were expressed with respect to the optical density of the milk system without emulsifying salts addition where the pH was adjusted to  $5.80 \pm 0.01$  after 18 hours of storage. All samples were measured three times.

### Chemical analysis

Dry matter content (according to ISO 5534 (2004)) was measured in processed cheese samples. On the other hand, pH (puncture pH meter with glass electrode, Malaysia) was investigated only in processed cheese and also in milk dispersion. All the samples were measured three times.

### Hardness analysis

Hardness of processed cheese samples was measured using TA.XT.plus texture analyser (Stable Micro Systems Ltd., Godalming, UK) with  $20\text{ mm}$  diameter cylindrical probe (strain of deformation  $25\%$ , a probe speed  $2\text{ mm/s}$ ) (Weiserová et al., 2011; Bayarii et al., 2012; Cunha et al., 2013). All the samples were measured three times.

### Statistical analysis

The results of chemical analysis and optical density were evaluated using non-parametrical analysis of variance by Kruskal-Wallis and Wilcoxon test (Unistat® 5.5 software, unistat, London, UK). The significance level used in the test was  $0.05$ .

## RESULTS AND DISCUSSION

### Results of chemical analysis

Dry matter content was measured in each processed cheese sample after 2, 9 and 30 storage days. Because the dry matter content is an important parameter, which can influence processed cheese textural properties; it is necessary to detect it in order to ensure the comparability of the individual samples (Lee et al., 2004).

The pH values of samples for individual applied emulsifying salts are shown in Table 1. The highest pH-values have samples with shorter phosphates (DSP, TSPP, PSTP) and with trisodium citrate. After 30 storage days at  $6 \pm 1$  °C a slight increase in pH (range from 0.10 to 0.20) was found ( $P < 0.05$ ).

Values of pH of processed cheese with binary mixtures are summarized in Table 2. Values of pH of samples decrease with increase in chain length. After 30-day storage, the results were the highest ( $P < 0.05$ ; compared samples with the same mixture of emulsifying salts).

### Results of hardness of processed cheese spreads

The lowest hardness values of processed cheese spreads were measured after 2 storage days with trisodium citrate addition and then the results increase with increasing chain length of phosphate emulsifying salts (DSP < TSPP < PSTP < POLY05 < POLY09 < POLY13 < POLY20 < POLY28) as can be seen in Figure 1. Slightly higher values were found for each sample after 30 storage days at  $6 \pm 1$  °C but the trend stayed unchanged.

Majority of binary mixtures of emulsifying salts did not significantly influenced hardness of processed cheese spreads (Figure 2) and an increasing hardness with increasing phosphate chain length was measured only in a not substantial intensity. Two samples were the exception of phenomena described higher. The first one was the binary mixture of DSP and TSPP which significantly reached hardness values. Opposite situation (smaller values in comparison with the others) was in binary mixture TSC:DSP. For each sample of binary mixture an increase of hardness after 30 storage days was measured.

### Results of skim milk dispergation

An intensity of optical density decreased with increasing chain length of phosphate emulsifying salts and the highest results were measured for samples including trisodium citrate as can be seen in Figure 3. When polyphosphates were included in to binary mixtures of emulsifying salts, optical density of model dispersal was decreased (Figure 4). Phosphates with longer chain length have bigger ability to exchange calcium and sodium ions and therefore were their optical density results lower. The results of optical density influence ability of emulsifying salts disperse casein what can in finally impact change hardness values of processed cheese spreads (an intensively casein dispersion leads to lower values of skim milk dispersion optical density and higher results of processed cheese spreads hardness).

**Table 1** Influence of processed cheese pH values on citrates and phosphates with different chain length addition\* after 2 storage days

Emulsifying salts	pH value
DSP	6.48 $\pm$ 0,02
TSPP	6.42 $\pm$ 0,03
PSTP	6.41 $\pm$ 0,02
TSC	6.39 $\pm$ 0,02
POLY05	5.95 $\pm$ 0,02
POLY09	5.44 $\pm$ 0,02
POLY13	5.38 $\pm$ 0,03
POLY20	5.36 $\pm$ 0,02
POLY28	5.35 $\pm$ 0,02

\* pH values mentioned like values average  $\pm$  standard deviation.

**Table 2** Influence of processed cheese values on binary mixtures of emulsifying salts addition\* after 2 storage days

Binary mixtures	pH value
DSP:TSPP	6.08 $\pm$ 0.01
DSP:POLY05	6.05 $\pm$ 0.01
DSP:POLY09	5.99 $\pm$ 0.01
DSP:POLY13	5.81 $\pm$ 0.02
DSP:POLY20	5.80 $\pm$ 0.02
DSP:POLY28	5.79 $\pm$ 0.02
TSPP:PSTP	6.08 $\pm$ 0.02
TSPP:POLY05	6.02 $\pm$ 0.01
TSPP:POLY09	5.88 $\pm$ 0.02
TSPP:POLY13	5.80 $\pm$ 0.01
TSPP:POLY20	5.79 $\pm$ 0.02
TSPP:POLY28	5.74 $\pm$ 0.01
PSTP:POLY05	5.81 $\pm$ 0.02
PSTP:POLY09	5.80 $\pm$ 0.02
PSTP:POLY13	5.79 $\pm$ 0.01
PSTP:POLY20	5.72 $\pm$ 0.02
PSTP:POLY28	5.70 $\pm$ 0.01
TSC:DSP	6.08 $\pm$ 0.01
TSC:TSPP	6.02 $\pm$ 0.02
TSC:PSTP	5.80 $\pm$ 0.02
TSC:POLY20	5.70 $\pm$ 0.01

\* pH values mentioned like values average  $\pm$  standard deviation

### Discussion

When individual polyphosphate salts are added, it could be seen, that with increase in number of phosphate atoms in chain length the pH-values of processed cheese decreases (Table 1). This phenomenon can be explained by the amount of hydrogen atoms, which can be released into the melt. Polyphosphates have higher amount of these atoms and therefore could decrease pH more intensive (Remy, 1961; Nagyová et al., 2012). The same phenomenon (decrease of pH with increase in chain length) was found likewise in binary mixtures (Table 2).

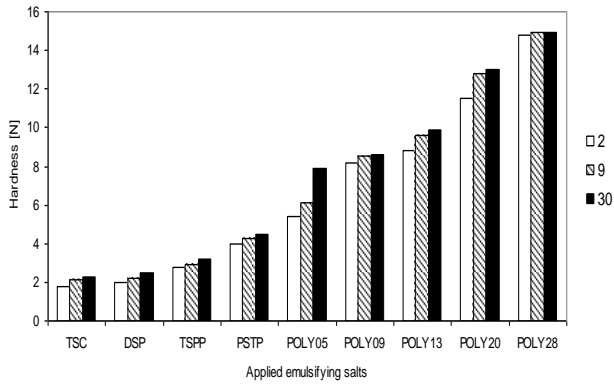


Figure 1 Influence of processed cheese hardness [N] on citrate and phosphates with different chain length addition

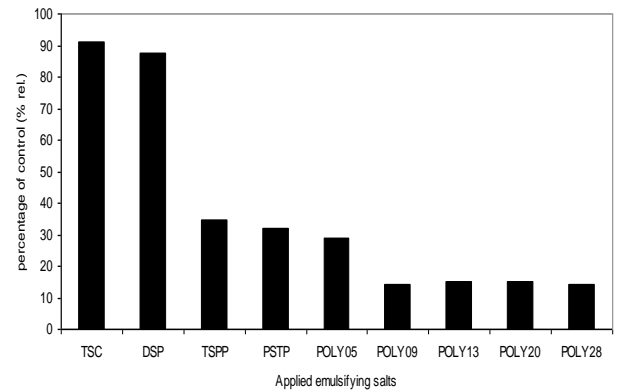


Figure 3 Influence of optical density (samples expressed with respect to optical density of the milk system without emulsifying salts addition) on citrate and phosphates with different chain length addition

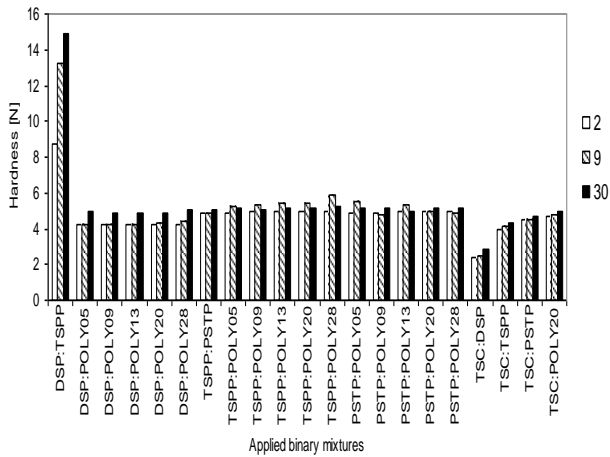


Figure 2 Influence of processed cheese hardness [N] on binary mixtures of citrate and phosphates with different chain length addition

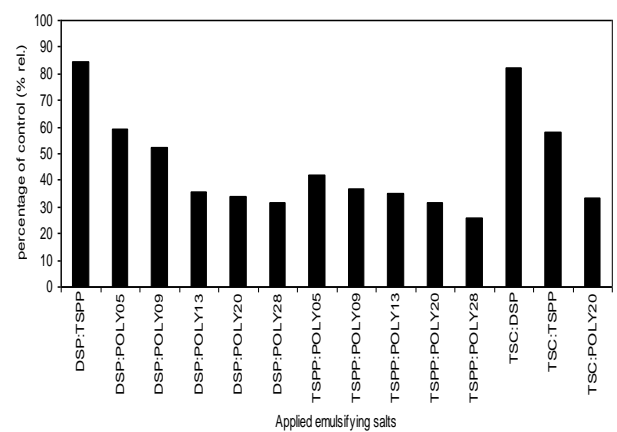


Figure 4 Influence of optical density (samples expressed with respect to optical density of the milk system without emulsifying salts addition) on binary mixtures of citrate and phosphates with different chain length addition

Hardness of processed cheese increases with increase in chain length of individual applied phosphates (Fig. 1). This phenomenon could be explain by larger ability of phosphates with longer chain length to ion exchanging, disperse caseins, stabilize water and fat in systems. The higher the presence of crosslinking in the matrix of the product, the harder processed cheese can be expected (Mizuno & Lucey, 2005; Shirashoji et al., 2010; Bayarri et al., 2012).

Low values of hardness in samples with trisodium citrate could be explained with their low affinity to calcium ions and low ability to protein hydratation. Low values in samples with trisodium citrate could be explained with their low affinity to calcium ions and low ability to protein hydratation. It is not capable to crosslink protein matrix and it could reduce hardness values of processed cheese (Mizuno & Lucey, 2005; 2007).

Hardness of samples with DSP and TSPP could be explained by a strong ability of the mixture of mono and diphosphate to enhance the formation of bridges between

diphosphate complexes with calcium ions and casein. Monophosphates are able to bind water there and therefore hardness of processed cheese increased (Mizuno & Lucey, 2007; Shirashoji et al., 2010; Kaliappan & Lucey, 2011; Buňka et al., 2012). Slight differences in hardness of binary mixtures including polyphosphates (in our case with chain length POLY05 and POLY09) could be explain mainly in their ability to give caseins multiple negative charges which probably leads to a lower intensity of hydrophobic interactions of the dispersed caseins and thus lower final matrix hardness (Shirashoji et al., 2010; Buňka et al., 2012; 2013).

After 30 storage days a little increase in hardness values was found. It could be caused by a hydrolysis of the emulsifying salts including more than two phosphorus atoms in a molecule, slight decrease of pH of the processed cheese, possible changes in the form of binding of the salts and thus a change in their dissociative characteristics and polymorphism of dairy fat and gradual change of its crystal forms (Carić & Kaláb, 1997;

Muslow et al., 2007; Shirashoji et al., 2010; Nagyová et al., 2012).

Intensity of casein dispersion is affected by the type of added emulsifying salts. Phosphates with longer chain length are capable of casein dispersion in larger intensity in comparison with phosphates with shorter chain length (Figure 3). This phenomenon is linked to idea that polyphosphates exchange more ions and therefore the intensity of milk optical intensity decrease. The hardness values are related to casein dispersion intensity. The more dispersed casein are, the more these proteins utilize their emulsifying and hydrating properties and stabilize oil and water present in the mixture. It leads to an increase in the intensity of interactions in the melt and crosslinking casein, resulting in processed cheese with higher hardness (Mizuno & Lucey, 2005; Shirashoji et al., 2010; Bayarii et al., 2012). Binary mixtures of emulsifying salts added in skim milk dispersion (Figure 4) had equal trend like individual emulsifying salts (decreasing values with increasing chain length). Citrate salts are comparable with monophosphates if individual or binary mixtures were applied.

## CONCLUSION

The aim of this work was to describe hardness of model processed cheese spreads with individual and binary mixtures of trisodium citrate and disodium phosphate, tetrasodium diphosphate, pentasodium triphosphate and polyphosphates with different chain length (n ≈ 5, 9, 13, 20 and 28) addition. The smallest values of processed cheese hardness were measured for trisodium citrate, followed by monophosphates and subsequently rose with increasing number of phosphorus atoms in phosphate chain. Majority of applied binary mixtures did not significantly influenced hardness of processed cheese spreads. A binary mixture (DSP:TSPP) was an exception.

Optical density of skim milk dispersion decreased with increase of phosphate chain length and the samples with trisodium citrate addition have higher values. The considerable trend was found if binary mixtures of emulsifying salts were applied.

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