

Szajnar K., Znamirowska A., Kalicka D., Kuźniar P. 2017. Fortification of yoghurts with various magnesium compounds. J. Elem., 22(2): 559-568. DOI: 10.5601/jelem.2016.21.3.1226

ORIGINAL PAPER

FORTIFICATION OF YOGHURTS WITH VARIOUS MAGNESIUM COMPOUNDS

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Abstract

Owing to the presence of B vitamins, proteins and lactose, milk and dairy products fortified with magnesium could function as transporters of magnesium to the human body. The aim of the study was to determine the effect of the type of an applied magnesium compound on the dynamics of fermentation, syneresis, texturometric profile and colour of yoghurts. The yoghurts produced were thickened with 3% addition of skimmed milk powder and fortified with various magnesium compounds in the amount of 30 mg Mg 100 g⁻¹ of milk. The fermentation of milk was carried out with starter yoghurt cultures at the temperature of 45°C for 4.5 hours. During the milk fermentation, changes in acidity were examined immediately after the addition of the starter, after 2 h of incubation and after 4 h of incubation. After 24 h of cold storage, texture, syneresis, pH, titratable acidity and colour of yoghurts were determined. The magnesium compounds used for the fortification of milk in the dose of 30 mg Mg 100 g⁻¹ of milk did not inhibit fermentation. After 24 h of storage, all yoghurts fortified with magnesium had a pH typical of fermented milk. Among the magnesium compounds used, magnesium bisglycinate had the strongest alkalizing impact on acidity of milk prior to fermentation. Fortification of yoghurts with magnesium compounds reduced syneresis of the whey in yoghurts. The texturometric profile of yoghurts changed depending on the type of an applied magnesium compound. Comparable components of texture were determined in control yoghurts and in yoghurts enriched with magnesium D-glukonate hydrate. Magnesium L-pidolate and magnesium chloride hexahydrate significantly reduced hardness and adhesiveness of yoghurt curd. Enriching yoghurts with magnesium bisglycinate, magnesium L-lactate, magnesium chloride hexahydrate, magnesium L-pidolate and magnesium acetate caused a darker colour compared to the colour of yoghurts which were not enriched.

Keywords: yoghurt, texture, magnesium, syneresis, colour, fermentation process.

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INTRODUCTION

Food enrichment has a significant impact on the health of consumers, and therefore this process must be carefully planned and consistent with the state's nutritional policy. The amount of an enriching substance added to a food product should be sufficient to achieve the intended health effect but it must not interfere with the absorption and metabolism of other nutrients or lead to overdosing (Gahruie et al. 2015).

With the consumption of yoghurts in Poland on the increase, enrichment of these dairy products can limit or prevent diseases related to deficiency of minerals. As numerous medical studies show, irrespective of a country, age or gender, a poor supply of magnesium in human diet causes a major health threat (Wojtasik et al. 2009, Grzebisz 2011). The average use of magnesium from a diet by the human body is 40-50%, and its absorption increases in the presence of B6 vitamin, lactose and protein. Milk has been determined to contain 0.03-0.06 mg 100 g⁻¹ of B6 vitamin, 4.77% of lactose and 3.48% of protein (Kozikowski, Przybyłowicz 1994, Wszołek 1997, Król et al. 2011, Bobrowska-Korczak 2015). Therefore, milk and dairy products fortified with magnesium could function as transporters of magnesium into the human body (Ziarno 2004, Gerhart, Schottenheimer 2013, Szeleszczuk, Kuras 2014, Gahruie et al. 2015). An appropriate magnesium compound should be selected for fortification of dairy products, one that would meet technological, economic and nutritional expectations. Achieving the desired effect of the enrichment and production of a food product that satisfies the above requirements is not always possible, which may account for the modest range of fortified products on the Polish market.

Magnesium from an appropriate source and added to dairy products should be well absorbed by the body, safe in use, have the desired solubility, chemical and heat stability, should not interact with other components or cause changes in sensory attributes and shorten the shelf-life of a final products (LAWLESS et. al. 2003, *Regulation (EC) No. 1925/2006*).

The aim of the study was to determine the effect of the type of a several magnesium compound added to yoghurt on the dynamics of fermentation, syneresis, texturometric profile and colour of yoghurts.

MATERIAL AND METHODS

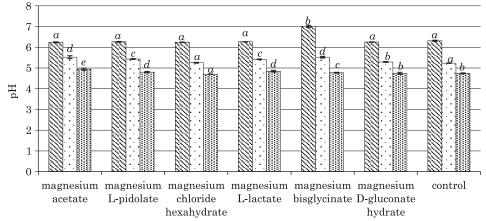
The experiment was composed of two stages. The first stage included production of yoghurts (2% fat milk) thickened with 3% of milk powder and fortified with magnesium compounds 30 mg Mg 100 g⁻¹ of milk: magnesium acetate pure p.a. $(CH_3COO)_2Mg \cdot 4H_2O$ (Chempur, Poland), magnesium L-pidolate $C_{10}H_{12}N_2MgO_6$ (Sigma, France), magnesium chloride hexahydrate pure

p.a. MgCl₂·6H₂O (Chempur, Poland), magnesium L-lactate hydrate C₆H₁₀MgO₆· H₂Ō (Sigma - Aldrich, Spain), magnesium bisglycinade MgC₄H₄O₅ · 3H₂O (Sigma – Aldrich, USA), magnesium D-gluconate hydrate C₁₉H₉₉MgO·H₉O (Sigma – Aldrich, USA), and pasteurized at the temp. of 72°C for 15 seconds. To be in line with Commission Regulation (EC) No 1662/2006 of 6 November 2006 amending Regulation (EC) No 853/2004 of the European Parliament and of the Council laying down specific hygiene rules for food of animal, the temperature used during research was 72°C. Magnesium compounds increase the risk of denaturisation of milk proteins at high temperature, and the choice of the above pasteurization temperature not only minimized the denaturisation process, but it also helped us to avoid using different pasteurization temperatures for various magnesium compounds (ZIARNO et al. 2009). The established amount of magnesium did not cause thermal denaturation of proteins of milk during its pasteurization (at the temp. of 72°C for 15 s). The fermentation with the use of the yoghurt starter culture YC-X16 (Chr. Hansen) was performed at 45°C for 4.5 h and then fermented beverages were cooled down to 5°C. The beverages were examined during incubation for the change of yoghurt's acidity directly after addition of the starter, after 2 h and also after 4 h of incubation. In the second stage of the study, after 24 hours of cold storage (at 5°C), yoghurts were analyzed for syneresis, to which end a sample of yoghurt weighing 25 g was placed on a paper filter in a funnel. The funnel was placed in a measuring cylinder (of known weight) and was left for 120 min at 5°C. Whey leakage (the quantity of whey) was weighed and the percentage was calculated in relation to the weight of the sample (Cueva, Aryana 2008). The pH determination was performed with the electrometric method by measuring the activity of hydrogen ions with a pHmeter Mettler Toledo FiveEasy (Switzerland). Potential acidity was established by titration of 0.25N samples with standard solution of sodium hydroxide in the presence of phenolphthalein as an indicator. Texture was assessed with an analyzer Brookfield CT3 (USA) equipped with Brookfield Texture Pro CT software. For determination, the TPA (Cycle Count 2) was selected with a sample of solid state yoghurt put without mixing in a 100 ml container, at the following settings: sample – cylinder 66.00 mm x 33.86 mm, force 0.1 N, head speed 1 mm s⁻¹, table TA-BT-KIT, probe TA3/100 (acrylic cylinder 35 mm length). Colour was analyzed by a A. Minolta chromameter CR-400. A microcolour tristimulus colorimeter (Konica Minolta Sensing Inc., Milton Keynes, UK) was used for the determination of L*, a*, and b* values.

The experiment was repeated three times and each parameter was measured five times. The mean and standard deviation were calculated from the results using the software Statistica 12.0 (StatSoft, USA). The univariate analysis of variance was applied to evaluate the influence of the type of a magnesium compound used on the properties of fermented milk beverage. Significance of differences between the averages ($p \leq 0.05$) was estimated with the Tukey's test.

RESULTS AND DISCUSSION

Among the extensive literature on fermented dairy products there are few works describing changes in acidity during fermentation of milk beverages fortified with mineral compounds. However, as it is known, the presence of metals such as Cu, Zn, Cd, Pb and Hg in milk may cause changes in the fermenting activity of dairy bacteria, resulting in either a stimulatory or an inhibitory effect on fermentation (Ocak, Köse 2010). The type of magnesium compounds applied for enriching milk (30 mg Mg 100 g⁻¹ of milk) had a significant impact on pH and titratable acidity of milk during fermentation. The highest pH directly after the addition of starter was determined in milk fortified with magnesium bisglycinate (pH = 7.01), and this value was statistically significantly higher than in other samples. The lowest value of pH was obtained in milk enriched with magnesium acetate and magnesium chloride (pH = 6.24), but it was not significantly different from the pH of the control milk. High pH obtained after the addition of magnesium amino acid chelate results from alkalizing properties of this compound (HARTLE 2006). When controlling pH after two hours of fermentation, the smallest decrease in the value of pH compared to the control milk was found in milks enriched with magnesium bisglycinate and magnesium acetate. The differences turned out to be statistically significant. After 2 h of fermentation, the pH closest that of the control milk was found in milk with the addition of magnesium chloride. After four hours of fermentation, significantly higher ($p \le 0.05$) pH than in the control sample was determined in milks enriched with magnesium acetate, magnesium L-lactate hydrate, magnesium L-pidolate and magnesium bisglycinate (Figure 1). However, significantly lower pH compared to the control sample was found in milk with the addition of magnesium chloride



 \boxtimes after addition of the starter \square after 2 h of incubation \boxtimes after 4 h of incubation Fig.1 The effect of the type of a magnesium compound on pH of yoghurts during incubation process: a, b, c, d — different letters indicate a statistically significant difference between the magnesium compounds at the same time ($p \le 0.05$)

hexahydrate. Among all the magnesium compounds used for fortification, the highest pH was caused by the addition of magnesium acetate during fermentation. In the study by Ocak, Köse (2010) on the effects of fortifying milk with Cu, Fe and Zn minerals on the production and texture of yoghurt, the presence of Cu and Zn in milk was found to have had an inhibitory effect on the fermenting activity of a yoghurt starter culture. The incubation time was longer in Cu and Zn fortified yoghurt than control yoghurts and the Fe fortified one.

The added magnesium compounds also affected titratable acidity of processed milk compared to the value of acidity of the control milk (Figure 2).

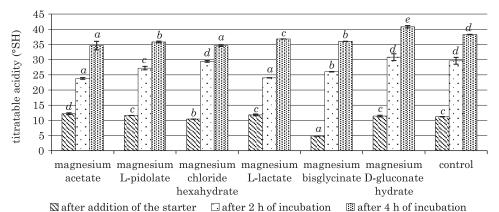


Fig. 2 The effect of the type of a magnesium compound on titratable acidity of yoghurts during incubation process: a, b, c, d – different letters indicate a statistically significant difference between the magnesium compounds at the same time ($p \le 0.05$)

Significantly higher titratable acidity of milk after the addition of starter was determined in milk magnesium acetate compared to the acidity of the control milk ($p \le 0.05$). The addition of magnesium chloride hexahydrate and magnesium bisglycinate resulted in a significant decrease in milk's acidity, particurally in comparison with the control milk ($p \le 0.05$). After 2 h of fermentation, significantly lower titratable acidity was found in milks with the addition of magnesium acetate and magnesium L-lactate hydrate in relation to the control sample ($p \le 0.05$). In contrast, the addition of magnesium chloride hexahydrate and magnesium D-gluconate hydrate did not significantly change the acidity of milk in the second hour of fermentation ($p \le 0.05$). At the end of the fermentation process, significantly higher acidity was obtained in milks enriched with magnesium D-gluconate hydrate compared to the rest of milks ($p \le 0.05$). Compared to the control sample, significantly lower titratable acidity was determined in milks enriched with magnesium acetate and magnesium chloride ($p \le 0.05$).

The value of pH after 24 h of cold storage was reduced in all analyzed yoghurts (Table. 1). Significantly higher pH was obtained in yoghurts enriched with magnesium acetate and magnesium L-lactate hydrate than in

Table 1

The effect of the type of a magnesium compound on pH, titratable acidity (°SH), syneresis (%) and colour of yoghurts after 24 h of cold storage

Pr	Properties	Magnesium acetate	Magnesium L-pidolate	Magnesium chloride hexahydrate	Magnesium L-lactate	Magnesium bisglycinate	Magnesium D-gluconatehydrate	Control
	$_{ m Hd}$	4.540 ± 0.04^{b}	4.475 ± 0.02^a	4.450 ± 0.01^a	4.530 ± 0.00^{b}	4.530 ± 0.00^b 4.495 ± 0.02^{ab}	4.465 ± 0.02^a	4.460 ± 0.01^a
Tir	Titratable acidity (°SH)	35.60 ± 0.00^a	36.00 ± 0.00^{b}	$36,60\pm0.28^{b}$	$36,20\pm0.01^b$	$37,20 \pm 0.28^{ab}$	$40,80 \pm 0.20^{\circ}$	$35,60 \pm 0.10^{a}$
Syne	Syneresis (%)	43.93 ± 1.51^{ab}	$41,85 \pm 3.16^{ab}$	$41,01 \pm 0.36^a$	$44,23 \pm 1.68^b \qquad 44,44 \pm 1.02^b$	$44,44 \pm 1.02^{b}$	$42,38 \pm 0.63^{ab}$	$44,48 \pm 1.33^b$
J	r*	95.33 ± 0.78^{a}	96.92 ± 0.49^b	94.38 ± 0.79^a	98.57 ± 0.87^c	96.42 ± 0.56^b	97.47 ± 1.62^{bc}	98.23 ± 1.95^c
noĮo	ъ*	-4.020 ± 0.10^{a}	-4.020 ± 0.05^a	-4.020 ± 0.05^a -4.113 ± 0.05^{ab} -4.080 ± 0.04^a -4.043 ± 0.17^a	-4.080 ± 0.04^a	-4.043 ± 0.17^a	$-3.968 \pm 0.09^{\circ}$	-4.235 ± 0.13^{b}
)	, q	14.52 ± 0.22	14.50 ± 0.07	14.54 ± 0.31	14.58 ± 0.22	14.51 ± 0.19	14.44 ± 0.43	14.71 ± 0.35

Explanatory notes:

Table shows mean values and standard deviation.

a,b,c - mean values denoted with different letters within lines differ statistically significantly $(p \le 0.05)$

The effect of the type of a magnesium compound on texture of yoghurts after 24 h of cold storage

Table 2

Properties	Magnesiumacetate	Magnesium L-pidolate	Magnesium chloride hexahydrate	Magnesium L-lactate	Magnesium bisglycinate	Magnesium D-gluconatehydrate	Control
Hardness (N)	1.258 ± 0.08^a	0.891 ± 0.06^{b}	0.891 ± 0.06^{b} 0.933 ± 0.21^{b} 2.690 ± 0.04^{c} 2.217 ± 0.54^{c}	2.690 ± 0.04^c	2.217 ± 0.54^c	3.060 ± 0.09^d	3.113 ± 0.037^d
Adhesiveness (mJ)	2.831 ± 0.36^a	0.200 ± 0.00^{b}	0.200 ± 0.00^{b} 0.354 ± 0.15^{b} 3.933 ± 1.78^{a} 5.733 ± 1.96^{c}	3.933 ± 1.78^{a}	$5.733 \pm 1.96^{\circ}$	9.133 ± 1.07^d	7.933 ± 1.35^{dc}
Springiness (mm)	13.50 ± 0.14^a	13.71 ± 0.63^a	13.71 ± 0.63^{a} 13.45 ± 0.73^{a} 11.53 ± 0.58^{b} 13.28 ± 1.58^{a}	11.53 ± 0.58^{b}	13.28 ± 1.58^a	12.85 ± 0.70^{a}	12.82 ± 0.71^a

Explanatory notes:

Table shows mean values and standard deviation.

a,b,c – mean values denoted with different letters within lines differ statistically significantly $(p \le 0.05)$

the remaining ones ($p \le 0.05$). In the study by PIRKUL et al. (1997), fortification of yoghurt with calcium L-lactate or calcium D-gluconate had statistically significant influence on titratable acidity and pH of yoghurt ($p \le 0.05$). However, there were no statistically significant differences between the acidity values of the control yoghurt and the yoghurts fortified with a mixture consisting of both compounds of calcium: calcium L-lactate combined with calcium D-gluconate.

After 24 h, titratable acidity of yoghurts containing L-pidolate, chloride hexahydrate, L-lactate hydrate, and D-gluconate hydrate was significantly higher compared to the acidity determined in the control yoghurts ($p \le 0.05$).

Fortification of milk with magnesium compounds in the amount of 30 mg Mg 100 g⁻¹ of milk caused a decrease in syneresis by 3.47% in yoghurts with magnesium chloride hexahydrate, by 2.63% in yoghurts with magnesium L-pidolate and by 2.10% in yoghurts with magnesium D-gluconate hydrate as compared to the control yoghurts. In contrast, the addition of magnesium bisglycinate and magnesium L-lactate hydrate did not impair syneresis. In the experiment by ACHANTA et al. (2007), enrichment of yoghurt with 420 mg Mg 170 g⁻¹ in the form of magnesium carbonate also lead to reduced syneresis in comparison with the control yoghurt, as did the other compounds tested in the cited work, i.e. zinc gluconate, manganese sulfate monohydrate, sodium molybdate dihydrate, chromium chloride and sodium selenite. According to several authors, syneresis may be limited by increasing the amount of protein and total dry matter in yoghurts (Bhullar et al. 2002, Martin--Diana et al. 2004, Amatayakul et al. 2006, Akalm et al. 2008). Martin-Diana et al. (2004) explained this decrease in syneresis by the water retention capacity of proteins, which increases with the denaturation caused by heat treatment.

Adding 30 mg Mg 100 g⁻¹ of milk in the form of acetate, L-pidolate, chloride hexahydrate and bisglycinate prior to fermentation caused some darkening of the colour of yoghurts compared to the control yoghurt, as evidenced by the values L* coefficient (Table 1). In contrast, the addition of magnesium L-lactate hydrate and magnesium D-gluconate hydrate did not affect significantly the brightness of the colour of yoghurts. The values of parameters a* and b* indicate a colour shift in space toward green and yellow colours. All yoghurts fortified with magnesium were characterized by a lower portion of green and yellow colours than the control yoghurts. ACHANTA et al. (2007), examining fat free plain set yoghurts fortified with various minerals, found that the mean L* values of yoghurts fortified with chromium, magnesium, manganese and molybdenum were higher than the control. In turn CLOWLEY et al. (2014), in their research on fortification of reconstituted skim milk powder with different calcium salts, emphasize that the used calcium compounds do not significantly affect the colour brightness. Only fortification with calcium hydroxide – Ca(OH)₂ significantly contributed to a lower intensity of yellow colour in the tested yoghurt.

Individual distinguishing features of the texture of yoghurts (Table 2) were found to depend on the type of a magnesium compound used for enrichment. The highest adhesiveness was in the yoghurt with the addition of D-gluconate hydrate, and the highest elasticity was determined in the yoghurt enriched with L-pidolate. In addition, fortification of milk with magnesium definitely reduced hardness of yoghurts, where the lowest value of hardness of clot was caused by the addition of L-pidolate and magnesium chloride hexahydrate, especially in comparison with the yoghurts without enrichment. In the case of L-pidolate and magnesium chloride hexahydrate, they were also found to have resulted in the lowest adhesiveness of yoghurt curds. The texturometric profile most similar to the control yoghurt was determined in yoghurt with the addition of magnesium D-gluconate hydrate. Ocak and Köse (2010), examining the texture of yoghurt with Cu, Fe and Zn minerals, found that Cu addition of 0.2 mg l⁻¹ to yoghurt caused significantly $(p \le 0.01)$ higher firmness and cohesiveness of the product, while the firmness and cohesiveness values for 3.5 mg l⁻¹ addition of Zn were significantly lower than those connected with the other doses and metals $(p \le 0.05, p \le 0.01)$. In order to increase the firmness of yoghurts enriched with magnesium, hydrocolloids or whey protein isolate (WPI) may be used; optionally prebiotics can be added (Guvenet al. 2005, Gustaw et al. 2007, 2011).

CONCLUSIONS

- 1. The magnesium compounds used for fortification of milk (magnesium acetate, magnesium L-pidolate, magnesium chloride hexahudrate, magnesium L-lactate hydrate, magnesium bisglycinate, magnesium D-glukonate hydrate) at a dose of 30 mg Mg 100 g⁻¹ of milk did not inhibit the fermentation process. After 24 h of storage, all yoghurts fortified with magnesium had pH typical of fermented milks.
- 2. Among the applied magnesium compounds, the greatest impact on acidity of milk prior to fermentation was shown by magnesium bisglycinate acting alkalization.
- 4. Fortification of yoghurts with magnesium compounds caused reduced syneresis of whey in yoghurts.
- 5. Depending on the type of the applied magnesium compound, the texturometric profile of yoghurts changed. Comparable components of texture were determined in control yoghurts and in yoghurts enriched with magnesium D-glukonate hydrate. Magnesium L-pidolate and magnesium chloride hexahydrate significantly reduced hardness and adhesiveness of yoghurt clot.
- 6. Enrichment of yoghurts with magnesium bisglycinate, magnesium L-lactate hydrate, magnesium chloride hexahydrate, magnesium L-pidolate and magnesium acetate caused some darkening of the colour in comparison with the colour of yoghurts which were not enriched.

7. D-gluconate magnesium, out of all the examined magnesium substances, seems to be the most preferred form of Mg for yoghurt supplementation because of the least influence on yoghurt's texture.

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