Creamed Cottage cheese enriched with *Lactobacillus* GG

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Summary

Cottage cheese was produced from reconstituted skimmed milk powder. Fermentation of skim milk with approximately 11.16% total solids was performed at 22 °C with 0,5% mesophilic starter culture, "O" type (DVS, Chr. Hansen's Lab), without rennet addition. Dry Cottage cheese samples contained an average 21,32% total solids; 17.85% proteins; 0.75% ash; and 90.33 mg Ca/100g. The Cottage cheese produced from the fresh skim milk was taken as a control. The dressings for salted and sweet Creamed Cottage cheese were made from commercial sour cream (12% fat) with salt (3%) and sucrose (25%) addition, respectively. Both kind of dressings are inoculated with probiotic bacterium Lactobacillus casei subsp. rhamnosus GG (Lactobacillus GG). The cheese grains and dressings with and without Lactobacillus GG bacteria, are mixed at 1:1 ratio. Addition of high level of Lactobacillus GG cells to salted or sweet Creamed Cottage cheese samples had no adverse effect on sensory criteria. During 14 days of storage at +8 °C, the sensory properties of all Creamed Cottage cheese samples were not changed substantially, while the viable count of Lactobacillus GG (CFU/g) was approximately doubled in both types of Creamed Cottage cheese.

Key words: Cottage cheese, composition, sensory evaluation, Lactobacillus GG, survival

Introduction

Cottage cheese is a soft unripened, and acid coagulated curd formed in the individual granules (Codex Alimentarius, 1984). It is low calorie cheese with high protein content produced from full skim milk and in the case of Creamed Cottage cheese it is covered with some kind of creaming mixture (Cambell, 1990; Kosikowski and Mistry, 1997; Rosenberg, 1993; Tong et al., 1994; Tratnik et al., 1995; Tratnik, 1998). Although the specific origin of this cheese is unknown the name "Cottage" implies that the cheese is originally produced on family farms (Guinee et al., 1993). Industrial production of Cottage cheese started about 1915 in the USA (Kosikowski and Mistry, 1997) and until today different ways for production of this cheese have been referred (Guinee et al., 1993; Kosikowski and Mistry, 1997; Mc Auliffe et al., 1999; Tratnik et al., 1995; Tratnik, 1998; White et al., 1984). Each change during Cottage cheese production may have the great influence on its

composition, yield, characteristics and nutritive value (Bruhn and Franke, 1988; Kosikowski and Mistry, 1997; Martin et al., 1993; Mc Auliffe et al., 1999; Mistry, 1990; Puhan et al., 1994; Rosenberg, 1993; Shelef and Ryan, 1998; Tong et al., 1994; Tratnik et al., 1995). It is also mentioned that standard quality of Cottage cheese grains could be obtained from reconstituted skim milk powder (Guinee et al., 1993; Kosikowski and Mistry, 1997; Tratnik, 1998; White et al., 1984), which is the case in this work.

The aim of this work was to increase the functional properties of Creamed Cottage cheese by addition of the probiotic bacterium of *Lactobacillus casei* subsp. *rhamnosus* GG (*Lactobacillus* GG) in salted or sweet cream dressing which was mixed with dry Cottage cheese grains.

There are relatively few reports concerning cheese as a carrier of some probiotic microorganisms Gardiner et al., 1998; Gomes and Malacata, 1998; Jordan and Cogan, 1999; Stanton et al., 1998; Tratnik, 1998), even though only few probiotic cheeses are currently on the market world-wide, such as traditional Emmentaler cheese. Cottage cheese may offer certain advantages as a carrier of probiotic organisms (Blanchette et al., 1996; Puhan et al., 1994; Tratnik, 1998). Having a higher pH-value than the more traditional fermented probiotic foods (Gilliland, 1998; Guinee et al., 1993; Kosikowski and Mistry, 1997; Tratnik, 1998), it may provide a more stable milieu to support their long-term survival.

The purpose of this study was to observe the survival of *Lactobacillus* GG bacteria in salted or sweet Creamed Cottage cheese samples during the 14 days of storage at refrigerator's temperature. Furthermore, it is very important to evaluate the influence of *Lactobacillus* GG bacteria on sensory characteristics of both Creamed Cottage cheese types during their storage.

Materials and methods

The Cottage cheese was produced from the reconstituted skim milk powder (240-280 g/2 L water at 40 °C). Control Cottage cheese was produced from fresh skim milk.

Fermentation of 2 L skim milk samples is performed at 22 °C with 0.5% mesophilic starter culture, "0" type (DVS, Chr. Hansens Lab., Denmark), without rennet addition.

The parameters during cheese making processes by long term method (Kosikowski and Mistry, 1997) are presented in Fig. 1. The control cheese samples from fresh skim milk were produced in a same way. The cheese yield is expressed in percentage (kg cheese/100 L skim milk). The dressing for salted and sweet Creamed Cottage cheese was made from commercial sour cream (12% fat) with addition of salt (3%) and sucrose (25%), respectively. Both types of cream dressing are inoculated with probiotic bacterium *Lactobacillus casei*

subsp. *rhamnosus*, ATCC53103, (*Lactobacillus* GG) and mixed with Cottage cheese grains. The mixture ratio, of cheese grains and cream dressing with or without inoculation of *Lactobacillus* GG bacteria, was 1:1.

The probioitic strain *Lactobacillus* GG was isolated from "BioAktiv" commercial product, (pasteurised milk with *Lactobacillus* GG) produced by "Dukat" dairy company, Zagreb, Croatia. *Lactobacillus* GG *bacterium* was propagated and maintained in MRS broth (Biolife, Milano). The cultivation of *Lactobacillus* GG was performed in 100 mL Erlenmeyer flasks, with 40 ml of MRS broth at 37 °C in water bath shaker. After 24 h of growth, *Lactobacillus* GG cells were harvested by centrifugation (4000 g, 10 min). The pellets were washed with saline and then resuspended in cream dressing to final concentration of $3x10^9$ CFU/g.

During 14 days of storage at 8 °C samples of Creamed Cottage cheese were sensory evaluated and the viable counts (CFU/g) of *Lactobacillus* GG were determined.

The sensory properties of cheese samples (with or without *Lactobacillus* GG), were evaluated by the panel group of 5 sensory analysts, using 20 points scoring system (Tratnik and Kršev, 1992). The points were obtained by multiplication of the scores for each properties (1-5) with weighted factor (Fw) (ISO, 1985). The sensory acceptability of cheese samples during storage, was also evaluated by the panel group of 8 chosen students, using hedonic scale according Mistry (1990). Than the sweet cheese samples are prepared using strawberry aroma addition into sweet cream dressing, regardless *Lactobacillus* GG bacteria addition.

The viable count of *Lactobacillus* GG (CFU/g), was determined by standard method on MRS agar plates at 37 °C/72 h. Cell morphology of *Lactobacillus* GG was analysed microscopically.

The chemical composition of skim milk samples and cheese samples was determined by standard methods (Official Methods of Analysis, AOAC, 1990). The calcium content was determined by atomic absorption spectrofotometry (Rowe, 1973). The pH-value (digital pH-meter Knick type 646) of cheese samples was measured by the method described before (Tratnik and Kršev, 1992).

Results and discussion

The composition variability of reconstituted skim milk (Table 1), used in cheesemaking processes (n=12), had an important influence not only on the duration of milk fermentation (1555-1735 hours) but on Cottage cheese yield (20.6-27.0%) as well (Fig. 1).

The chemical composition of dry Cottage cheese samples (Table 2) and the characteristics of formed cheese grains (Fig. 2) were more dependent on the

Table 1: Chemical composition and pH value of skim milk used for cheese production
Tablica 1: Kemijski sastav i pH-vrijednost uzoraka obranog mlijeka za proizvodnju sira

Composition and pH value Sastav i pH-vrijednost		Reconstituted skim milk samples Rekonstituirani uzorci obranog mlijeka		*Control samples Kontrolni uzorci
		from - to od - do	average (n=12) prosjek (n=12)	average (n=12) prosjek (n=12)
Total solids / Suha tvar	(%)	9.99 - 12.05	11.16	8.52
Proteins / Proteini	(%)	4.07 - 4.99	4.65	3.20
Lactose / Laktoza	(%)	5.06 - 5.98	5.36	4.48
Fat / Mast	(%)	nd**	nd**	0.05
Ash / Pepeo	(%)	0.79 - 0.98	0.92	0.68
Ca	(mg/100 g)	126.40 - 147.20	133.80	-
pH		6.76 - 6.87	6.82	6.64

* control samples of fresh skim milk / Kontrolni uzorci svježeg obranog mlijeka
** nd = not detectable / nije detektirano

Table 2: Chemical composition and pH-value of dry Cottage cheese samples
Tablica 2: Kemijski sastav i pH-vrijednost uzoraka Cottage sira

Composition and pH value Sastav i pH-vrijednost		Cottage cheese samples Uzorci Cottage sira		*Control samples Kontrolni uzorci
		from - to od - do	average (n=12) prosjek (n=12)	average (n=12) prosjek (n=12)
Total solids / Suha tvar	(%)	19.59 - 23.02	21.32	17.73
Proteins / Proteini	(%)	16.29 - 20.30	17.85	14.58
Fat / Mast	(%)	nd**	nd**	0.38
Ash / Pepeo	(%)	0.65 - 0.89	0.75	0.51
Ca	(mg/100 g)	73.19 - 104.80	90.33	-
	1st day	5.06 - 5.44	5.21	4.93
pH	7th day	4.87 - 5.47	5.18	5.01
	14th day	5.10 - 5.29	5.17	5.03

* control samples of Cottage cheese obtained from fresh skim milk
Kontrolni uzorci Cottage sira dobiveni od svježeg obranog mlijeka
** nd = not detectable / nije detektirano

processing parameters (Fig. 1) than on the composition of reconstituted milk samples used (Table 1). The control cheese grains (Table 2) possessed higher moisture content than defined by international standard for Cottage cheese (max. 80%). However, from reconstituted skim milk, with an average of 11% total solids (Table 1) the standard quality dry Cottage cheese with approximately 21.32% of total solids (Table 2), and rich in calcium (about 90 mg/100g) can be produced. Also, the cheese yield was much higher compared with control cheese, produced from fresh skim milk (an average 14.64%).

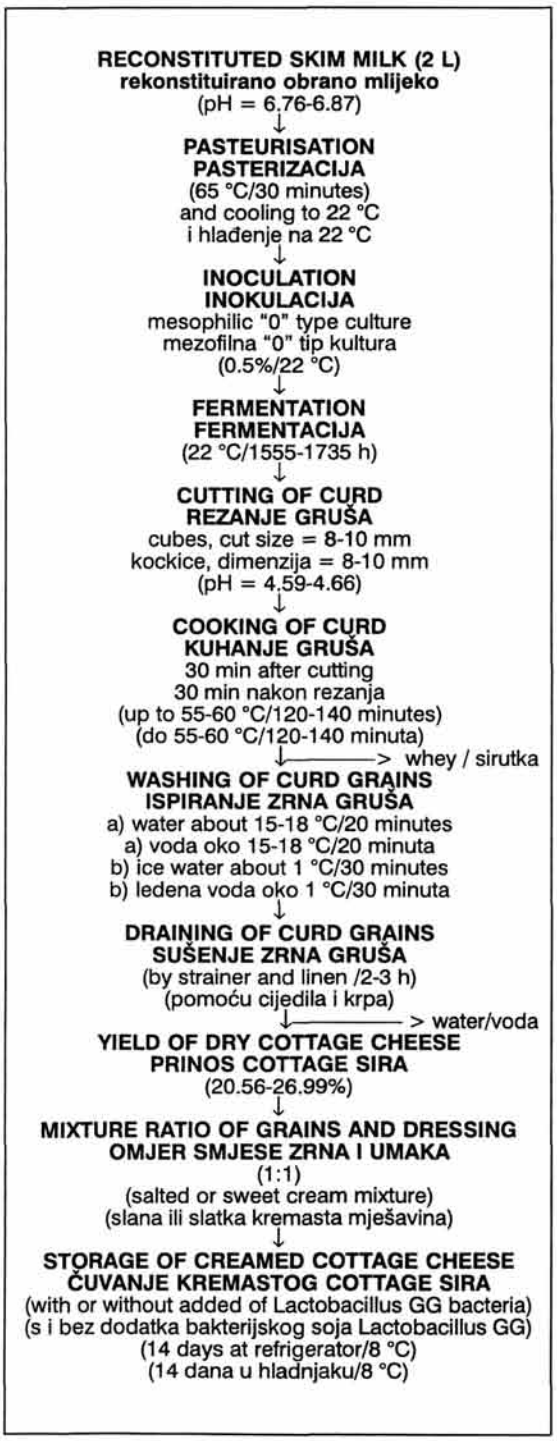


Fig. 1: Processing parameters during making of Cottage cheese

Slika 1: Procesni parametri tijekom proizvodnje Cottage sira

Table 3: Sensory properties evaluation of Cream Cottage cheese samples during storage*

Tablica 3: Senzorska ocjena Kremastog Cottage sira tijekom čuvanja*

Properties/points** Svojstvo/bodovi** (max. points/Fw) (maksimalni bodovi/Fw)	Sweet samples (n=3) Slatki uzorci (n=3)			Salted samples (n=3) Slani uzorci (n=3)		
	1st day 1. dan	7th day 7. dan	14th day 14. dan	1st day 1. dan	7th day 7. dan	14th day 14. dan
Appearance Vanjski izgled (max.=4/Fw=0.8)	3.04	3.04	2.64	3.20	3.20	3.04
Consistency Konzistencija (max.=4/Fw=0.8)	2.40	2.20	2.00	2.80	2.80	2.40
Colour Boja (max.=2/Fw=0.4)	2.00	2.00	2.00	2.00	2.00	2.00
Odour Miris (max.=2/Fw=0.4)	2.00	2.00	1.60	2.00	2.00	1.80
Flavour Okus (max.=8/Fw=1.6)	6.88	7.20	7.04	7.68	8.00	7.20
Total Ukupno (max.=20/Fw=4.0)	16.32	16.44	15.28	17.68	18.00	16.44

* Samples with or without addition of *Lactobacillus* GG bacteria have been equally evaluated Sweet samples prepared without aroma addition

* Uzorci s i bez dodatka bakterije *Lactobacillus* GG su jednako ocjenjeni Slatki su uzorci pripravljeni bez dodatka arome

** Points = score (1-5) (Fw (weighted factor) / Bodovi = ocjena (1-5) (Fw (faktor vaganja)

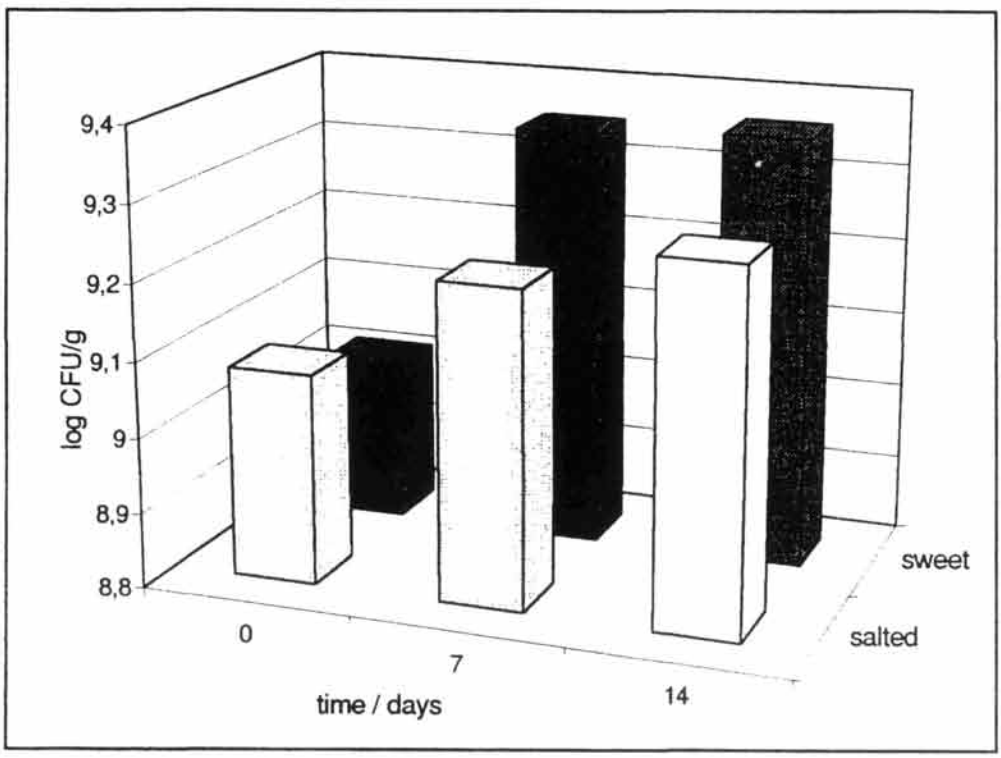
Fig. 2: Photographs of dry Cottage cheese samples

Slika 2: Fotografije uzoraka Cottage sira

first row =grains from fresh skim milk (control)
prvi red = zrna od svježeg obranog mlijeka (kontrola)
second row = grains from reconstituted skim milk
drugi red = zrna od rekonstituiranog obranog mlijeka

Fig. 3: Viable counts (log CFU/g) of Lactobacillus GG strain in salted and sweet Creamed Cottage cheese samples during storage at 8 °C (results are means of three replicates)

Slika 3: Broj živih stanica soja Lactobacillus GG (log CFU/g) u slanim i slatkim uzorcima Kremastog Cottage sira tijekom čuvanja pri 8 °C (rezultati su prosjek tri ponavljanja)



The general appearance of cheese grains, produced from reconstituted skim milk, were much alike to control Cottage cheese grains, produced from fresh skim milk (Fig. 2). In spite of lower moisture content, Cottage cheese grains obtained from reconstituted skim milk (Table 2), absorbed added cream dressing slightly stronger than control cheese grains. The addition of inoculum with high level of *Lactobacillus* GG to sweet or salted cream mixture had no adverse effect on sensory criteria of prepared Creamed Cottage cheese samples. The cheeses, enriched with *Lactobacillus* GG bacteria, possessed the flavour and texture comparable to control cheese samples, without *Lactobacillus* GG addition. During 14 days storage at +8 °C the sensory properties of both types of Creamed Cottage cheese samples (Table 3) did not change substantially, regardless *Lactobacillus* GG bacteria addition. Control samples of Creamed Cottage cheese, showed similar sensory properties to cheese samples obtained from

*Table 4: Acceptability evaluation of Creamed Cottage cheese samples during storage, using hedonic scale**

*Tablica 4: Ocjena prihvatljivosti uzoraka Kremastog Cottage sira tijekom čuvanja dobivena hedonističkom skalom**

Cheese sample/days Uzorci sira/Dani (n=3)		Flavour score Ocjena okusa (1-10)	Body texture score Ocjena teksture (1-5)	Appearance score Ocjena izgleda (1-5)	Σ (max. 20)
Sweet samples **	1st	8.66	3.00	3.69	15.35
Slatki uzorci	7th	9.00	2.75	3.86	15.61
	14th	8.00	2.50	3.50	14.00
Salted samples	1st	9.00	3.00	3.94	15.94
Slani uzorci	7th	9.00	3.00	3.94	15.94
	14th	7.75	2.75	3.50	14.00

* Flavour = 1 (dislike extremely) to 10 (like extremely); Okus = 1 (jako nepoželjan) do 10 (jako poželjan)
Body texture = 1 (poor) to 5 (excellent) Tekstura = 1 (loša) do 5 (izvrsna)
Appearance = 1 (poor) to 5 (excellent) Izgled = 1 (loš) do 5 (izvrstan)
** samples prepared using strawberry aroma addition / Uzorci pripravljeni s dodatkom arome jagode

reconstituted skim milk. The results for control samples are not presented, as they were not significantly different. However, the consistency of all cheese samples at the end of storage became slightly softer. In some experiments the odour and the flavour of Creamed cheese samples after 14 days of storage reminded to unfresh cheeses, probably as a result of not-hermetically packaging. Using the hedonic scale (Table 4), the differences of sensory quality between the samples with or without *Lactobacillus* GG bacteria addition also were not observed. In both evaluations (Tables 3 and 4) the best scores were obtained after 7 days of storage.

Lactobacilus GG survived well in Creamed Cottage cheese (salted and sweet samples) and retained viability of approximately 10^9 CFU/g during 14 days of storage at 8 °C in both types of cheese samples (Fig. 3). These results suggested that Creamed Cottage cheese can provide a suitable environment for the maintenance of probiotic strain - *Lactobacillus* GG at high levels over long period. Furthermore, the *Lactobacillus* GG can survive in fermented milk products together with mesophilic lactic acid bacteria as well as with yoghurt bacteria, even for longer storage time (21 day), which was observed by others as well (Borović et al., 1998). *Lactobacillus* GG *bacterium* had previously been isolated (1984) from healthy human intestine by Goldin and Gorbach (Goldin and Gorbach, 1992) and characterised in detail with regard to their probiotic potential (Borović et al., 1998; Goldin and Gorbach, 1992; Puhan et al., 1994; Rogelj, 1994; Saxelin, 1997; Siitonen et al., 1990; Stanton et al., 1998; Svensson, 1999; Šušković et al., 1997). The number of viable microbial

cells that should be present in a probiotic product has been the subject of many discussions, but is usually considered to be between 10^6 and 10^8 CFU/mL (or CFU/g) (Svensson, 1999).

The investigated Creamed Cottage cheeses contained over 10^8 CFU/g *Lactobacillus* GG, thus satisfying criteria for probiotic food product. The importance of these probiotic-containing products, commonly regarded as functional foods, in the maintenance of health and well being is a key factor affecting consumer choice. This has resulted in rapid growth and expansion of the market for such products, in addition to increased commercial interest in exploiting their proposed healthful attributes (Gardiner et al., 1998).

Conclusions

From reconstituted skim milk (with approximately 11% of total solids) the Cottage cheese of the standard quality can be produced (with approximately 21.32% of total solids). The composition of reconstituted milk used for Cottage cheese production (240-280 g/2L water) had important influence on the fermentation time and also on the cheese yield. The composition and sensory properties of dry Cottage cheese samples are more dependent on the processing parameters than on the composition of reconstituted milk used. During 14 days of storage at +8 °C, the sensory properties of salted and sweet Creamed Cottage cheese samples did not change substantially, regardless probiotic bacteria addition. The viable count (CFU/g) of inoculated *Lactobacillus* GG was approximately doubled during 14 days storage in both types of Creamed Cottage cheese. In this study, laboratory scale cheeses with high level of *Lactobacillus* GG addition were found to have flavour and texture comparable to those of control cheeses, indicating that addition of this probiotic strain to Creamed Cottage cheese had no adverse effects on sensory criteria.

KREMASTI COTTAGE SIR OBOGAĆEN S LACTOBACILLUS GG

Sažetak

Cottage cheese je izvorno ime za svježi meki sir, zrnatog tipa. Proizveden je od rekonstituiranog obranog mlijeka u prahu. Fermentacija uporabljenih uzoraka mlijeka, s prosječno 11,16% suhe tvari, provedena je pri 22 °C dodatkom 0,5% mezofilne kulture za izravnu inokulaciju u mlijeko za proizvodnju, "O" tipa (DVS, Chr. Hansen's Lab, Danska), bez uporabe sirila. Uzorci ocijeđenog zrnatog sira sadržavali su u prosjeku 21,32% suhe tvari; 17,85% proteina; 0,75% pepela i 90,33 mg Ca/100g. Kontrolni uzorci proizvedeni su od svježeg obranog mlijeka. Kremasta mješavina (umak) za pripravu slanih ili slatkih uzoraka kremastog zrnatog sira (Creamed Cottage cheese) načinjena je od komercijalnog kiselog vrhnja (12% masti) uz dodatak soli (3%) ili šećera (25%). Slatka ili slana kremasta mješavina

nacjepljena je inokulumom probiotičke bakterije Lactobacillus casei subsp. rhamnosus GG (Lactobacillus GG). Omjer mješavine sirnih zrna i kremaste mješavine (s i bez LGG bakterija) bio je 1:1. Dodatak visoke koncentracije Lactobacillus GG bakterija nije imao utjecaj na senzorske karakteristike ni slatkih ni slanih uzoraka kremastog zrnatog sira. Tijekom 14 dana čuvanja ovih uzoraka sireva pri temperaturi hladnjaka (+8 °C) nisu zapažene bitne promjene njihovih senzorskih svojstava, dok je broj živih stanica Lactobacillus GG (CFU/g) bio u prosjeku udvostručen u oba tipa pripravljenog kremastog zrnatog sira.

Riječi natuknice: Cottage sir, sastav, senzorska ocjena, Lactobacillus GG, preživljavanje

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