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Study on Physico-Chemical, Rheological and Sensory Properties of Mozzarella Cheese Made by Direct Acidification

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Abstract: The effect of two types of organic acids (Lactic and Citric acid) at three pH levels (5.3, 5.6 and 5.8) on yield and chemical (calcium), rheological (elasticity) as well as sensory (taste and flavor) properties of cheeses made by direct acidification in comparison with conventional Mozzarella cheese were investigated, using a factorial experiment in Completely Randomized Design. Results indicated that the type of acid and pH used for initial pH adjustment had a significant effect on the yield, chemical and elasticity ($p<0.01$). The interaction of acid type and pH had a significant effect on chemical and elasticity ($p<0.05$), but had no effect on the yield ($p>0.05$). Taste and flavor were affected significantly by type of acid and pH ($p<0.05$), but the interaction of pH and acid type had only a significant effect on stretchability ($p<0.05$) and had no effect on taste and flavor ($p>0.05$). The results of sensory evaluation revealed that conventional cheese and cheese made by citric acid at pH = 5.3 and pH = 5.6 respectively have scored higher values than other samples.

Key word: Mozzarella · direct acidification · pH · physico-chemical · rheological · organoleptic properties

INTRODUCTION

Currently, more than 4000 types of cheeses are produced in the world. Among all, global production of Italian processed cheese has grown dramatically during last decade [1]. The main impetus for this growth has been the ubiquitous increase in the popularity of pizza, in which Mozzarella is the main cheese used. Low moisture Mozzarella cheese is a prominent member of the pasta filata group and includes Mozzarella, Provolone and kashkaval [2]. Pasta Filata cheeses are distinguished by a unique plasticizing and kneading treatment of the fresh curd in hot water. In recent years this cheese is produced in Iran in amounts far less than processed or the so-called pizza cheeses. Low moisture mozzarella cheese is traditionally produced using a mixed thermophilic starter cultures. However, it is also produced by direct acidification, using an organic acid in some countries. It is generally accepted that pH and Ca concentration influence the ability of curd to plasticize in hot water. Many studies have been done on the application of organic acids in production of mozzarella cheese during last two decades. Olson [3] produced mozzarella cheese by direct acidification method using citric, lactic, phosphoric, hydrochloric and acetic acids. He investigated different properties of cheese and the effect of acid type on time of coagulation and the amount of rennet that is necessary for coagulation. According to that finding, more hardness and least moisture content of mozzarella cheese is produced by using phosphoric acid while mozzarella cheese which is produced using citric and lactic acid had the most moisture content and was softer. The effect of acid type (in constant pH and rennet) on coagulation time was also investigated by this researcher. It was found that using citric acid to acidify milk resulted in slowering the coagulation than other acidulants. Yun *et al.* [4] and Michael and Nolan [5] studied the effect of direct acidification by citric acid on mozzarella cheese properties. These researchers produced cheeses with pH = 5.6 and 5.8 and found out that the hardness of cheese has been increased and chewingness and melt properties has been decreased by increasing the pH to 5.8. They also indicated that by decreasing the pH from 6 to 5.8, the amount of calcium content decreased about 20 percent. McMahon *et al.* [6] investigated the effect of direct acidification on microscopy and macroscopy structure of mozzarella cheese. They pointed out that cheese with 6 and 10

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percent of moisture, 0.3 and 0.6 percent of Ca and pH = 5-3 and 0.6 percent of Ca and pH = 5-3 and 5.8 was selected. The functional attributes of Mozzarella cheese are influenced by many factors, including milk pretreatment; make procedure, composition and proteolysis [7, 8]. Comparatively little information is available on the effect of pH and Ca content and their interaction on the functional attributes of Mozzarella cheese when made by direct acidification.

The objective of this study was to investigate the effect of two types of organic acids (lactic and citric acid) at three pH levels (5.3, 5.6 and 5.8) on physicochemical, rheological and organoleptic properties of Mozzarella cheese in comparison with conventional mozzarella cheese at pH = 5.3.

MATERIALS AND METHODS

Cheese manufacture: In each of three trials, four types of Mozzarella cheese were manufactured, using either starter culture, as conventional manufacture of the control cheese at pH 5.3, or by direct acidification using lactic and citric acids at pH 5.3, 5.6 and 5.8. All experimental as well as control cheeses were made in a 5 l water bath as a vat. The six cheese trials differed with respect to pH of milk at rennet addition (setting), pH of the curd at whey drainage and pH of the curd at milling and plasticization.

The details for manufacture of the different cheeses are not given here but it should be noted that the procedure was similar to the control cheese unless otherwise specified and the pasteurized milk used for all treatments had protein-to-fat ratio of 1.2 and its proximate analysis was as follow: Fat: 2.5%, protein: 3.01%, acidity: 15.8-16, Density: 1.025-1.03, Ca: 120 mg/ 100 g, K: 150 mg/ 100 g, pH = 6.6-6.7. Preliminary bench scale cheesemaking studies were undertaken to establish the levels of lactic acid and citric acid required to reduce the initial pH of milk to the appropriate values. Milk for control cheese was inoculated with a starter culture TCC-5 (Chr-Hansen Co.), consisting of *Streptococcus thermophilus* and *Lactobacillus. delbreuckii* ssp *bulgaricus.* Fungal rennet (Meito Co.) was added to milk at appropriate level depend on the pH of milk in such a way that the cutting time for all treatments was similar. The coagulum was cut when it attained firmness sufficient to withstand cutting, as assessed subjectively. After cutting (~30 min), the curd-whey mixture was allowed to heal for 6 min and then stirred continuously, with the speed of stirring increasing gradually from 10 to 22 rpm over 30 min. The curd-whey mixture was cooked to 40°C, at a rate of 1°C /5 min and continuously stirred until desired stretching is achieved. To examine the streachability, the mixture is put under the hot water for 1 min and then its taken out of water and is pulled genteelly by hand. This action is repeated 4-5 times, if the curd breaks, then it is not yet ready and must left undisturbed for a few more minutes (5 to 10) before trying the test. On a further trial, if the water was nonturbid and if the curd stretches like chewing gum then it is ready for next operation. After whey drainage, the curds were cheddared and milled. The curd was then heated to 85°C in brine (5%) and kneaded manually. All treatments were produced at triplicate.

Packaging: The product was then packed in airtight plastic bag (Nescofilm), to avoid moisture loss and then was kept in freezer at -4°C until the day of experiment.

Cheese firmness: Texture analyzer Model QTS 25 was used to determine the texture of all cheese trials according to the machine guideline. Firmness was the force required to compress the samples of cheese to 30% of the original height.

Sensory analysis: Organoleptic properties of pizza cheese were tested by skilled panelists. Color, stretchability and aroma of all treatments have scored by a panel of 12 judges and the data were statistically analyzed using a five points hedonic method [9] with1 being devoid of attributes and 5 being extremely strong. Judges were selected by their ability to differentiate the aforementioned attributes using sequential triangle tests on dairy products [9].

Statistical analysis: A factorial experiment in Completely Randomized Design was used to analyze the response variables relating to the overall acceptability of cheeses. Analysis of variance (ANOVA) was carried out using a SAS procedure. Duncan's multiple comparison test was used as a guide for pair comparisons of treatment means. The level of significance was determined at $p<0.05$.

RESULTS AND DISCUSSION

The effect of pH on Ca and the interaction of acid type and pH on Ca content of all cheese trials have been shown in Fig. 1. The amount of Ca is known to be direct ly correlated with physical and rheological properties of cheese. In general, the lower pH at setting results in increased solubilization of micellar calcium phosphate and a concomitant increase in the concentration of soluble Ca

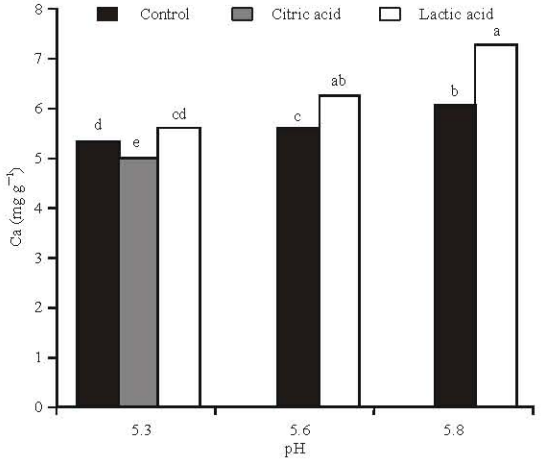

Fig. 1: Effect of pH and acid type on Ca content of Mozzarella cheese

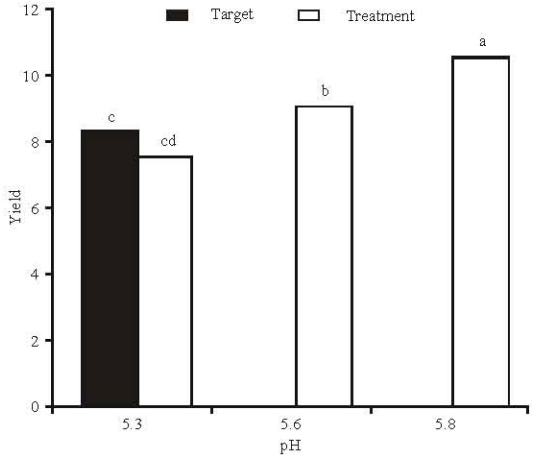

Fig. 2: The effect of pH and acid type on yield

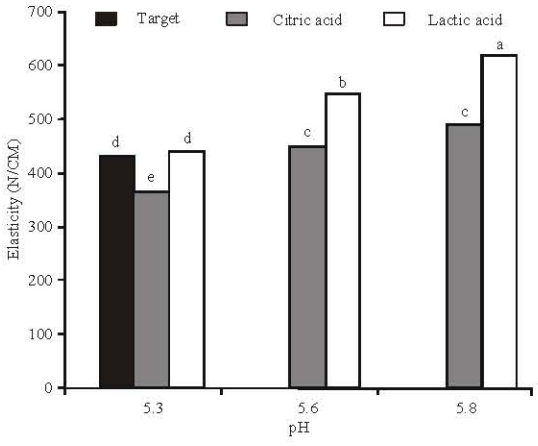

Fig. 3: The effect of pH and acid type on elasticity of Mozzarella cheese

in the whey while in contact with the curds [10]. Acid type and pH used for initial pH adjustment of milk for cheesemaking significantly affect the Ca content of all treatment as illustrated in Fig. 1. It should be mentioned that cheeses produced using starter culture or acid to adjust the pH at setting to 5.3, had no significant difference in terms of Ca content. In general, the effect of acid on micellar calcium phosphate is as follow:

Ca phosphor-caseinate + Organic acid → undissolved casein + Ca salt

In similar pH, Ca content of cheese trail produced by citric acid was less than the one which was produced by lactic acid. This was observed in all cheese trials and indicated that the type of acid used for acidification of milk, had significant difference on Ca content of cheese at the same pH.

This effect concurs with the results of others (Keller *et al.* and Metzger *et al.*) who reported a reduction in the Ca content of cheese made from milk that had been preacidified before setting to a degree depending on the extent of pH reduction at setting and the type of acid used [11-13].

The effect of pH on yield has shown in Fig. 2. Yield of cheese has a direct relation to moisture content and Solid non Fat (SNF) level. The higher moisture content of directly acidified cheeses may be due to its relatively low Ca-to casein ratio, which would be conductive to a greater degree of casein hydration. This result concurs with those of Keller *et al.* [11] which showed a linear increased in the moisture content of directly acidified Mozzarella as the Ca level of the cheese decreased. In other words, with reduction the setting pH from 5.8 to 5.3, the yield was decreased.

The interaction effect of pH and acid type on the yield of cheese was not significant. The analysis of variance of the interaction effects of pH and acid type on yield is depicted in Table 1.

One of the most important rheological properties of Mozzarella cheese is stretchability of melted cheese, which has a direct relation to cheese stretchability before melting. The effect of acid type and pH on elasticity changes is shown in Fig. 3. Significant differences in elasticity of cheeses made at pH 5.3 and 5.8 was observed ($p<0.01$). No significant difference was observed in elasticity between control cheese acidified by starter culture to pH 5.3 and directly acidified cheese to pH 5.3. The elasticity of Mozzarella cheese was increased as the setting pH decreased. The elasticity of cheese has an inverse relationship to Ca content. A reduction of pH

Table1: Analysis of variance of changes in pH, Ca, yield, elasticity, stretchability, taste and color in Mozzarella cheese made by different acid type, pH and Ca level

Treatment	pH	Ca	Yield %	Elasticity	Stretchability	Taste	Pizza color
Acid type	1	0.019**	0.011**	28.376**	0.500**	0.889*	0.056*
pH	2	1.751**	9.726**	29.587**	8.167*	3.167*	13.722*
Acid type + pH	2	0.018*	1.151^{NS}	5.562*	1.167*	0.056^{NS}	0.722^{NS}
Error	1266$^{(1)}$	0.103	0.348	5.761	1.111	0.389	0.667

* and **: Significant at 5 and 1% levels respectively, NS: Non Significant

Table 2: The mean of interaction effect of pH and acid type on yield, taste and color of Mozzarella cheese

Acid type	pH	Yield (%)	Taste of cheese	Pizza color
Citric acid	5.3	8.00^{cd}	3.12^{c}	2.40^{d}
	5.6	9.67^{b}	4.31^{bc}	4.90^{a}
	5.8	10.11^{a}	4.90^{b}	3.10^{c}
Lactic acid	5.3	7.53^{cd}	3.15^{c}	2.40^{d}
	5.6	9.41^{b}	4.50^{bc}	5.00^{a}
	5.8	10.50^{a}	4.71^{b}	3.00^{c}

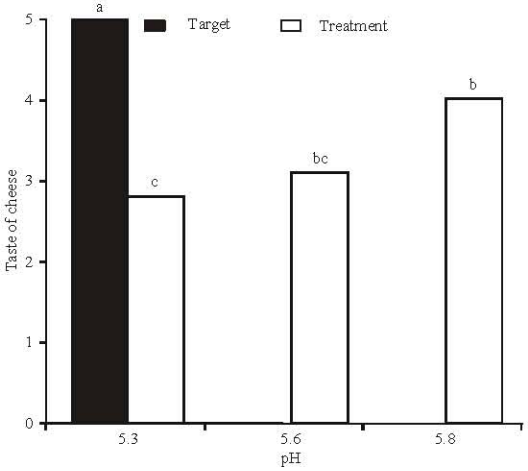

Fig. 4: The effect of pH and acid type on taste of cheese

before rennet coagulation as it is the case in preacidified cheeses, probably effects a more efficient removal of Ca from the curd than the slow reduction of pH after gelation and gel cutting, as in control cheese. The more Ca content of cheese leads to the higher firmness of the texture due to increase in the content of intact casein [14]. Results from analysis of variance indicated that setting pH has a significant effect on the organoleptic properties of Mozzarella cheese (Fig. 4). The control cheese was scored higher than all direct acidify cheese trials, probably mainly due to production of flavor compounds by starter culture rather than direct acidification. On the other hand, cheeses made by pH adjustment with direct acidification were scored as non pleasant cheese due to increase the

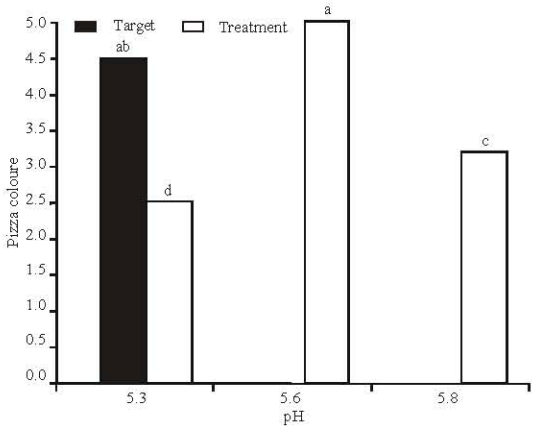

Fig. 5: The effect of pH and acid type on pizza color

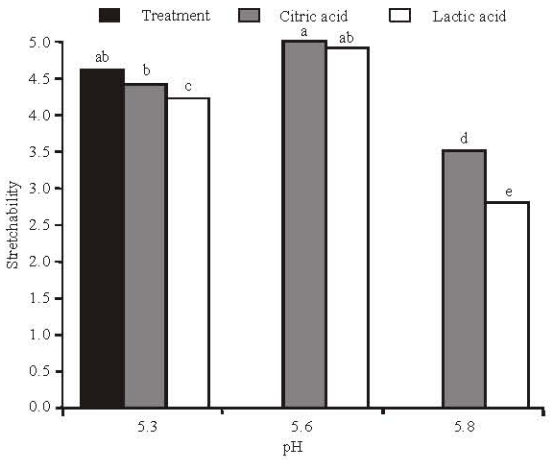

Fig. 6: The effect of pH and acid type on stretchability of Mozzarella cheese

amount of acid in the absence of aroma. Our finding is in agreement with other workers [15, 16]. Acid type, which was used to adjust the pH had no significant difference on taste scores ($p>0.05$). The analysis of variance of the interaction effects of pH and acid type on taste is also depicted in Table 1.

pH of cheese had significant effect on the color ($p<0.05$) as shown in Fig. 5. There was a decrease in the

score of directly acidified cheese color than control cheese color. In fact, Millard reaction occurred between the cheese constituents under high temperature conditions during the melting process of cheese which affect the color of cheese. Millard browning reaction is occurred between reduced sugars and proteins during heat induction of Pizza. Both heat and moisture have direct effect on the reaction. Reaction rate has direct relationship to pH and inverse relationship to moisture. By decreasing the pH form 5.8 to 5.3, significant difference is achieved in the color. Also, by increasing the reduced sugars, reaction rate will increased [17]. In directly acidified cheeses, lactose, the nonreduced sugar, remains intact whereas in starter culture cheeses, lactose is degraded by starter organisms and converted to reduced sugars which give stronger color on melting.

Decrease in setting pH had significant effect on cheese stretchability ($p<0.05$), so that with reduction of pH form 5.8 to 5.3, stretchability was increased. No significant difference was shown on the stretchability of cheeses adjusted at 5.3 and 5.6 as illustrated in Fig. 6. In general, pH and acid type concurrently have significant effect on stretchability of pizza cheese ($p<0.05$) and effect of cheeses made by direct acidification had higher stretchability than starter culture cheeses mainly due to reduction the Ca content and the casein network compactness [3]. The stretchability of cheese acidified by citric acid was greater than that acidified by lactic acid mainly due to the higher chelating ability of citric than lactic acids which in turn absorb more Ca and result in reduction the casein network compactness [18].

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