Preparation of Imitated Processed Cheese by Using Direct Acidification Technique to Resemble Mozzarella Cheese properties

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Abstract: Production of new imitated process cheese similar to stretched cheese was conducted to be used as a topping material for pizza in spite of Mozzarella cheese. Processed cheese have several advantages, compared to natural cheeses such as lower refrigerator cost, better keeping quality and longer storage stability. Direct acidification of cow's milk using three different types of acid was applied to produce acidic curds. Milk sample was divided into four portions .The first three portions were separately acidified with 2% diluted lactic acid; citric acid or lemon juice until pH 5.3 to create T1, T2 and T3 respectively. All individual acidic curds were mixed with 2 % emulsifying salt and cooked at 85°C, then turned to process cheese .The forth latter portion was manufactured as traditional Mozzarella and serve as control sample (C) . All samples were chemically analyzed; physically assayed and sensorial evaluated. The obtained results showed that the new process cheese made by direct acidification with lactic acid (T1) had the highest moisture and lowest acidity than other treatments. T3 had the lowest content of calcium and the highest acidity compared with the other treatments. Control Mozzarella cheese gained the best meltability and the highest stretchability followed by (T3). Fat leakage was the highest in control samples followed by T1. For the organoleptic properties; control Mozzarella samples gained slightly highest scores followed by T3 then T2. Data indicated that direct acidification technique succeed to produce new processed cheese which had semi- similar properties of the traditional Mozzarella cheese when used 2% diluted lemon juice. Sensory evaluation of pizza pastry samples; which prepared and topped by all individual cheese samples; showed that (T3) which prepared by using diluted lemon juice gained the highest scores than control and other treatments
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Key wards: Direct acidification, Lactic acid, Citric acid, Lemon juice, Mozzarella cheese, Imitated processed cheese.

1. Introduction

Mozzarella cheese is one of the cheeses that have achieved the most significant growth in production in the past century (Gwenole *et al.,* 2010). It has become one of the most popular cheese varieties in the world because of its primary use in pizza preparation. Its usage is expected to grow as global interest due to the over increasing demand for pizza and other foods that use mozzarella (Sameen *et al.,* 2010). This increase may be due to the increases in the popularity of pizza; high speed changes of consumers' behavior and the changes of life style of common people. Mozzarella is one of the cheeses that have special characteristics and physical properties which make it completely differ from all other types of cheese mainly stretchability (Nolan *et al.,* 1989; Kindstedt 1991; Tunick *et al*., 2000; Sameen *et al.,* 2010; and Abd El Gawad *et al.,* 2012). It's preparing need certain conditions; skill & carful manufacture; special precautions and long time to achieved these characters and adjustment of its technological steps. The main factors affect the success of Mozzarella cheese is the pH of curd in plasticization step. (Metzger *et al.,* 2000; Metzger *et al.,* 2001 Guinee *et al.,* 2002; and Najafi *et al.,* 2006). The pH value ranged between 5.1 - 5.4 and curd is scalded to about 57°C, Kneaded and stretched in hot water or diluted brine to about 80°C (Panthi, 2007). These conditions of low pH and high temperature are conductive to limited aggregation of Para- casein and formation of Para-casein fibers of relative high tensile strength (Guinee *et al.,* 2002). These fibers impart stringiness; strethchability; chewiness; or the other main physical characteristics of Mozzarella cheese (Fox *et al.,* 2000). The reduction of curd-pH, promote a number of physicochemical conditions which are conductive to the curd- flow during plasticization step, (VanHooydonk *et al*., 1986, Guinee *et al.,* 2002). So

the increasing of acidity; greatly; affected the properties of the resultant curd.

On the other side, direct acidification has been traditionally practiced in the manufacture of a number of un-ripened cheese varieties (Shehata & Olson, 1966; Olson, 1970; Kosikowski, 1977; Metzger *et al.,* 2000). The first scientific study of direct acidification technique was made on Cheddar cheese in 1955 by Mabbitt *et al*. to produce starter - free cheese. After that; many various studies had been done for applying this technique in cheese manufacture by different attitudes and for different targets (Metzger *et al.,* 2001; Panthi (2007) and Ahmed *et al.* (2011).

Therefore, the goal of this work was to prepared different acidic curds by applying the direct acidification technique using diluted lactic acid, citric acid or lemon juice for production of imitated processed cheese similar to mozzarella cheese properties to save time; give better keeping quality and longer storage stability.

2. Materials and Methods

Materials:

- Fresh raw cow's milk (3% fat) was obtained from the herd of faculty of Agric., Cairo Univ., Egypt.
- Kasomel K 2311 as emulsifying salt (which has a strong ion exchange capacity and has weak creaming action) produced by Doumer, France; was provided by International Dairy & Foods Co. Egypt.
- Food grade lactic and citric acids were parched from Sigma Company.
- Fresh Lemon juice was prepared in the laboratory.
- *Streptococcus thermophilus* and *delbrueckii* ssp *bulgaricus* obtained from Microbiological Resources Center (MIRCEN- Egypt) as starter culture; and Calf rennet powder (HA-LA) were used for preparing the traditional Mozzarella cheese sample.

Methods

Manufacture of traditional Mozzarella cheese samples

Control traditional Mozzarella cheese sample(C) was manufacture from cow's milk according to the standard method mentioned by Kosikowski (1982);

Imitated process cheese samples preparation

Imitated process cheese samples were prepared as flows: Cow's milk was divided into three portions: First portion was gradually direct acidified with 2 % diluted lactic acid until reached to pH 5.3 (T1). The second portion was also direct acidified with 2% diluted citric acid until pH 5.3 (T2), while the third portion was acidified with 2% fresh lemon juice until pH 5.3 (T3). Then the emulsifying salt (2%) was added to all individual acidic curd , cooked at 85°C and applied all the steps of manufacture as mentioned by Adrianson *et al*. (1997).

Three replicates were conducted for each treatment. The obtained cheese samples were freshly analyzed for their chemical composition; physical and sensory properties.

Methods of Analysis:

Dry matter (DM), fat, total protein (TP) and ash contents of fresh cheese samples were determined according to the AOAC (2007). The pH value was measured by using a digital pH meter (M4 1150 USA) equipped with glass electrode. Soluble nitrogen content (SN) was estimated by Kjeldah method (IDF, 1993). The calcium content was determined by titration with EDTA as described by Francesco and Raffaello (1980).

The cheese samples were also analyzed for meltability (tube test) as described by Savello *et al*. (1989). Fat Leakage test was determined according to Guinee *et al*. (1999); while oil separation was determined according to the method outlined by Thomas (1973).

Cheese samples were prepared for Texture Profile Analysis according to El-Zeny (1991). Instron universal testing Machine 4202 (Instron Co., Canton, OH) equipped with 50N (5kg) load cell was used. The chart was used to calculate the area under the response plot. Area 1 and 2 (cm^2) represent the area under the curves formed during the first and second compression cycle and the work done during compression. The height of the first peck during the first bite represented the extent of hardness (force) while the distance of the sample under compression during the second bite represented springiness (cm).Cohesiveness was derived from the ratio of Area 2/Area 1 whereas gumminess was equal to hardness x cohesiveness. Chewiness was equal gumminess x springiness. All texture measurements were done at room temperature (22 $\pm$2°C).

Sensory evaluation:

The organoleptic properties of cheese samples were evaluated by scoring panel of staff members; Dairy Department; National Research Center according to Nelson and Trout (1956). The sensory evaluation of Pizza pastry samples topped by experimental cheeses samples was carried out according to Abu-Foul (1990).

Statistical analysis:

Obtained data was statistically analyzed by the GLM procedure with SAS (2004) software.

3. Results and Discussion

Table (1) reflects the chemical composition of new imitated Mozzarella cheese. It is clear that new cheese made by direct acidified milk with diluted lactic acid (T1) had a lowest DM; followed by control (C) whereas imitated cheese which prepared

with acidified milk with lemon juice (T3) had highest DM than all treatments. This result is agreement with Shehata *et al*. (1967) who indicated that lactic acid tend to give a high moisture cheese. While Keller *et al.* (1974) reported that phosphoric acid which used to acidify the milk to pH 5.5 produced cheese of low moisture while moisture was significantly higher when malic or citric acids were used. However, Fox (1978) mentioned that lactic acid tented to give a high moisture cheese while hydrochloric yield gave satisfactory cheese. Najafi *et al.* (2006) mentioned that more hardness and lowest moisture content of Mozzarella cheese is produced by using phosphoric acid while Mozzarella produced using citric and lactic acid had the most moisture content and was softer. So the effect of acid type (in constant pH) was clear. From the same table it could be notice that new processed cheese made by acidified milk with lemon juice had a highest acidity followed by citric acid and control; whereas cheese samples prepared by milk acidified by diluted lactic acid (T1) had lowest acidity than all treatments. Ash/dry matter was the highest in process cheese made by acidified milk with lemon juice (T3) than all treatments followed by cheese made by acidified with diluted lactic acid and citric acid. Table (1) reveals also fat/dry matter in all treatments. Control and T2 had a highest content of fat/dry matter than all treatments followed by T1 and T3. Acid type affected the total composition of cheese in constant pH (Najafi *et al.,* 2006). From the same table it could be observed that control cheese had a highest content of TP/DM followed by T1. Control had highest content of soluble nitrogen/dry matter followed by (T1). The same table indicated that Cheese made by acidified milk with lemon juice (T3) had a lowest content of calcium than all treatments. This may be due to the high acidity which remove $Ca^{+}$from curd that lead to high firmness as result of increase casein which bond with $Ca^{+;}$ whereas control cheese had highest content of Ca^{+} than all treatments**.**

Table (1): Chemical composition of imitated process cheese samples:

properties	Treatments			
	C	T1	T2	T3
DM%	43.30	42.42	43.18	45.34
Fat/DM	40.42	40.10	39.37	37.49
TP/DM	45.54	45.47	44.97	43.23
Ash/DM	8.43^{b}	10.52^{a}	10.84^{a}	11.32^{a}
Acidity%	0.68	0.72	0.74	0.75
pH	5.4	5.2	5.1	5.0
SN/TP	0.402	0.377	0.366	0.344
Ca^{++} mg/100 g	0.860^{a}	0.691^{b}	0.542^{bc}	0.438^{c}

C = Control (Mozzarella cheese); T1: cheese made by direct acidification with diluted lactic acid. ; T2: cheese made by direct acidification with diluted citric acid. T3: cheese made by direct acidification with lemon juice.
*No significant difference between means with the same super script.

Rheological properties

Table (2) illustrates some rheological properties of new process cheese include meltability, fat leakage, oiling off and stretchability.

Meltability:

Meltability is an important character, which determines; to a great extent; the quality of process cheese as well as Mozzarella cheese. Meltability of the processed cheese was expressed as the distance of cheese flow in millimeter. From table (2) it could be notice that meltability of control Mozzarella cheese was the highest than all treatments followed by cheese which made by acidified milk by diluted lactic acid (T1) and cheese made by acidified milk with lemon juice (T3) ,finally cheese samples made by acidified with diluted citric acid(T2). Lawrence (1968) gives a wide explanation for the factors that affect the melting quality of the processed cheese. These are the heat induced interaction of casein and whey proteins resulting in reduced capacity to flow, binding of free water by denatured protein or casein complex, calcium binding by denatured β-lactoglobulin and reduced proteolysis. Process cheese made by acidified milk with lemon juice (T3) had a lowest pH and high acidity; which lead to weaken of protein bonds and de-emulsion of fat. (Shimp, 1985).

Fat leakage

From the same table; (2); it could be observed that control Mozzarella cheese samples had a highest fat leakage (cm^2) followed by (T1) process cheese prepared by acidified milkwith diluted lactic acid while (T2) process cheese acidified by citric acid had lowest leakage than all treatments .Table (1) reveals that control had a highest content of soluble nitrogen than all treatments. This increase in soluble nitrogen contents (more protein decomposition) resulted in lower degree of emulsification and high fat leakage. (Awad, 1996).

Table (2): Rheological properties of imitated process cheese samples:

Properties	Treatments*			
	C	T1	T2	T3
Meltability (mm)	80[a]	75a[b]	68[b]	70[b]
Fat leakage (cm^2)	45.50[a]	38.30[b]	34.00[b]	34.46[b]
Oiling off %	6[a]	3[b]	1.5[c]	1[c]
Stretchability (mm)	9[a]	2.0[c]	5.0[b]	6.0[b]

* See table (1)

Oil separation

Oil separation of cheese is affected by emulsifying salts which used to provide a uniform structure during the melting process. The emulsifying salt kasomel (K2311) has a strong ion exchange capacity and has a weak creaming. From table (3) ; it noticed that cheese made by acidified milk with lemon juice(T3) had a lowest oiling of followed by T2 made by acidified milk with diluted citric acid and T1 made by acidified milk with diluted lactic acid .

Stretchability

From table (3); it clear that control Mozzarella cheese had a highest stretchabilitythan allother treatment; followed by (T3) and (T2).

Texture profile of imitated process cheese samples

Hardness is the force required to attain a given deformation, springiness or elasticity is the rate at which a deformed material goes back to its un-deformed condition after the deforming force is removal; and cohesiveness is defined as the quantity simulating the strength of the internal bonds making up the body of the product.Table (3) reflect the texture profile of new processed imitatedMozzarella cheese. It is clear that control Mozzarella cheese had a highest hardness (kg) than all treatments followed by (T1) and (T3) and finally (T2). This may be due to pre-acidification of milk which resulted in reduction in both total calcium and water insoluble calcium (Metzger *et al.,* 2001). From the same table chewiness (kg/mm) in control Mozzarella cheesewas the highest than all treatments. Rudan and Barbano, 1998; show that melt chewiness of low fat Mozzarella cheese can be reduced with high levels of proteolysis or low level of water-insoluble calcium or a combination of both factors. However, high levels of proteolysis and low levels of water-insoluble calcium decrease post melt-whiteness may adversely affect the consumer acceptability of low fat Mozzarella cheese As shown in table (3) cheese made by acidified milk with lemon juice(T3) had a highest elasticity (springiness) than other treatments and control.

Table (3): Texture profile of imitated process cheese samples.

Parameters	Treatments*			
	C	T1	T2	T3
Hardness (kg)	0.580	0.42	0.40	0.37
Chewiness (kg/mm)	4.224	2.83	3.08	3.02
Springiness (mm)	13.20	15.12	14.55	12.52
Cohesiveness	0.55[a]	0.48[b]	0.57[a]	0.54[a]

* See table (1)

The elasticity of cheese has an inverse relationship to Ca^{++} content. Pre-acidified cheese, probablyaffecteda more efficient removal of $Ca^{++}$from the curd than slow reduction of pH after relation asin 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 (Guinee and Fenelon, 2000).

Cohesiveness is defined as the quantity simulating the strength of the internal bonds making up the body of the product El-Zeny (1991). . The same table (3) shows that (T2) process cheese made by acidified with citric acid had the highest cohesiveness followed by control Mozzarella Cheese and (T3). From the same table (3) it could notice that chewiness (kg/mm) in control was a highest than all treatments followed by (T2) and (T3). Finally process cheese made by acidified milk with lactic acid (T1) was a highest gumminess (kg) than control followed by (T1) and (T3) finally (T2).

Organoleptic properties

Table (4) illustrates the organoleptic properties of new imitated Mozzarella cheese. It is clear that control and (T3) gained thehighest scores for color and aroma followed by (T1) and (T2). Control cheese samples hadthe highest scores of consistency than al treatments followed by (T2). On the other hand (T3) had lowest consistency scoresthan all treatment and control. Whereas oiling off was lowest in (T3) and (T2) than other treatments followed by (T1) and control. Treatment (3) gained the highest degree for taste followed by control and finally (T2) and (T1). Control gained alsothe highest total score followed by (T3), (T1) finally (T2).

Table (4): Sensory evaluation of f imitated process cheese samples:

Sensory characteristics	Control (10 Degree)	T1 (10 Degree)	T2 (10 Degree)	T3 (10 Degree)
Color	9	8	7	9
Aroma	8	7	6	9
Consistency	8	6	7	5
Taste	8	7	6	9
Total	39^{a}	31^{b}	28.5^{b}	36^{a}

C = Control (Mozzarella cheese); T1: cheese made by acidified milk with diluted lactic acid. T2: cheese made by acidified milk with diluted citric acid; T3: cheese made by acidified milk with lemon juice.
*No significant difference between means with the same super script

Table (5) shows the sensory evaluation of pizza samples which topped by the new imitated Mozzarella cheese. It is clear that control gained the highest score for appearance followed by (T3) and (T2) finally (T1).Control had the highest score for color followed by (T3) and (T2) finally (T1) also.While (T3) gained the highest total score followed by control and (T2) finally (T1). However (T3) gained the highest scores for aroma followed by control, (T2) and finally (T1). Taste took the same trend of aroma where (T3) had gained the highest scores for texture followed by (T2) and control finally (T1).

Table (5): Sensory evaluation of Pizza pastry samples topped by imitated process cheese.

Sensory characteristics	Possible scores	Treatments			
		C	T1	T2	T3
Appearance	1-20	18^{a}	15^{b}	16^{ab}	17^{a}
Aroma	1-20	15^{a}	10^{b}	15^{a}	17^{a}
Taste	1-20	17^{a}	9^{b}	17^{a}	18^{a}
Texture	1-20	18^{a}	10^{b}	19^{a}	20^{a}
Color	1-20	19^{a}	11^{b}	17^{a}	18^{a}
Total score	100	$87a^{b}$	55^{b}	84^{ab}	90^{a}

C = Control (Mozzarella cheese); T1: cheese made by acidified milk with diluted lactic acid. T2: cheese made by acidified milk with diluted citric acid; T3: cheese made by acidified milk with lemon juice.
*No significant difference between means with the same super script.

Conclusion:

Production of new imitated processed cheese using direct acidified milk was success when used % 2 lemon juice until pH 5.3. This treatment produced acceptable samples to resemble Mozzarella cheese properties for topping pizza and other related bakery products.

This type of cheese which prepared by cooking the acidic curd with emulsifying salt; can be used for topping pizza. . It could be save time, effort, precautions and refrigerating conditions for storage of Mozzarella cheese. Pizza samples topped by T3-cheese gained the high total scores for appearance, aroma, taste, texture and color.

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