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(Original Article)

Influence of Potassium Sorbate on Chemical and Microbiological Properties of Ras Cheese During Ripening Periods

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Abstract

The objective of this study to explain the effect of potassium sorbate E 202 (0.1%, 0.2% and 0.3%) on some chemicals properties (Moisture, Acidity, Salt, Fat, pH, Total nitrogen and Soluble nitrogen), microbiological properties (Total Bacteria count, Total count of fungi and yeast and its effect on fungal growth and mycotoxins production) of Ras cheese during five ripening periods (fresh, 15 days, 30 days, 60 days, 90 days) and sensory evaluation for cheese in (30 days, 60 days and 90 days). The results show that moisture and pH decrease in all treatments during ripening period's progress, Acidity, T.N and S.N increases in all treatments as the settlement period progresses, and the highest was in the second treatment (Ras cheese treated with 0.2% potassium sorbate). Results showed a good role of potassium sorbate in inhibiting fungal growth by increasing its concentration and its role in inhibiting the ability of fungi to produce mycotoxins. Cheese treated with potassium sorbate 0.3% had the best sensory acceptance during the ripening period (60 days), with a rate of 96.5% that mean that it can use potassium sorbate as a good antifungal factor to improve Ras cheese quality.

Keywords: *Chemical properties, Hard Cheese, Microbiological properties, Potassium sorbate, Ripening period*

Introduction

Ras cheese is the most widely consumed hard cheese in Egypt and possibly the Arab world. This cheese, comparable to Greek hard Kefalotyri, is produced in small and medium-scale dairy factories around Egypt. This cheese is commercially known as Romi or Torky cheese (Abou-Donia, 2008). It is made from raw cow's milk or a mixture of cow's and buffalo milk without using starter cultures (Awad *et al.,* 2003). Ras cheese's maturing duration makes it ideal for consumption. The Egyptian Organization for Standardization and Quality recommends pasteurized milk for all forms of cheese to maintain its quality and safety during ripening (Awad, 2006 and Singh *et al*., 2003). Ras cheese ripens at temperatures ranging from 9°C to 12°C (Hattem *et al.,* 2012). Microbial contamination in cheese can occur from several sources including starting culture, brine, floor and packing

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materials, cheese cloth, curd cutting knife, cold room, and production room air (Temelli *et al*., 2006). Cheese contaminated with fungi can deteriorate or produce objectionable odors, smells, or metabolic products, making it inappropriate for ingestion (Banjara *et al*., 2015.). The fundamental issue of Ras cheese is that the storage areas where cheese is stored for months to acquire texture during ripening are not cleaned on a regular basis, despite being meant to be. Ras cheese is brined or dusted with salt and pressured by molds, causing salted water to drip onto the wood shelves where the cheese blocks remain for months. This allows wood to absorb and build up microbiological contamination such as bacteria and fungi. As fungi change into cheese blocks, they produce mycotoxins such as aflatoxins, which can cause liver damage and cancer. Mycotoxins are secondary metabolites of mycobiota that can be hazardous to animals and humans. *Aspergillus flavus* and *Aspergillus parasiticus* are the most common species capable of producing aflatoxins. Dairy products can contain mycotoxins from two sources: (a) indirect contamination from ingested feed, such as aflatoxin M1, and (b) intentional or accidental mold growth. Seddek *et al.* (2016) suggest that these naturally occurring chemicals provide a selective advantage to producer strains in complex ecosystems.

Potassium sorbate E 202 is called an organic acid and has been widely utilized as a food preservative agent (Rajapaksha *et al*., 2013). It can be mixed with the food itself or added as part of the packaging process. When used in compliance with appropriate manufacturing or feeding practices. Potassium sorbate increases the shelf life of foods by inhibiting the growth of molds and yeasts. The French discovered it in the 1850s, obtaining it from mountain ash tree berries. Its safety and efficacy as a preservative have been studied for the past fifty years. The US Food and Drug Administration (FDA) consider it as usually safe when used properly. This study aims to study the effect of spraying different solutions of potassium sorbate (0.1%, 0.2% and 0.3%) on the surface of Ras cheese on chemical and microbiological quality of cheese during ripening periods and its effect on mycotoxins production.

Materials and Methods

1. Materials

Cheese milk

Fresh cow's milk used in this study was obtained from El fares Market Seed area, Assiut. Milk has the following chemical composition (fat 3.83, SNF8.89, pH 6.47, salt 0.73%, protein 3.25%).

Starter

Pure cultures of *Streptococcus thermophilus and Lb. delbrueckii* subsp. *bulgaricus*11842 obtained from the Microbial Resources Center, Ain Shams University, Cairo, Egypt.

Rennet

Standard microbial rennet EMCU powder was added to cheese milk in an amount required to coagulate unsalted milk within 40- 45 minutes at 35 ° C was purchased from local store in Cairo (Caglio Star, Espana production).

Salt

Commercial edible grade sodium chloride used for salting Ras cheese obtained from local markets in Assiut.

Wax

A commercial fine food grade plastic wax was obtained from local market in Egypt.

Potassium sorbate

Potassium sorbate pure was obtained from the Egyptian chemical store company, Alexandria, Egypt.

2. Methods

Ras Cheese Manufacture

Ras cheeses were manufactured in triplicate as described by Hofi *et al*. (1973) and addition of 0% control, 0.1%, 0.2% and 0.3%, potassium sorbate by spraying the surface of the cheese wheel. Cheeses were ripened at 12±2°C for 3 months.

Chemical analysis

Samples of Ras cheese were analyzed in triplicate for moisture, fat, acidity, and salt according to AOAC (2020). pH values were determined according to the method described by Hooi *et al*. (2004), Protein was determined by Kjeldahl method and convert the nitrogen to protein using 6.38 factor according to the ISO 8968-1:2014 | IDF 20 – 1:2014.

Microbiological analysis

Preparation of Samples

Ten grams of cheese were weighed and emulsified in sterile mortar 90 ml sodium citrate solution 2%. This 1:10 of cheese used for making serial dilutions required for the microbial analysis (ISO 6887-1:2017).

The total aerobic mesophilic Bacterial counts (TBCs)

A 1 mL aliquot of diluted material was plated on nutrient agar medium and cultured for 48 hours at 37ºC. Each dilution was plated on triplicate plates, and plates with 30-300 colonies were selected (Marshall, 2004).

Yeasts and Molds

One ml of the appropriate dilution was plated on potato dextrose agar medium and incubated at 28-30ºC for 6 days for molds (Smith and Dawson, 1944) and one ml in the appropriate dilution was plated on yeast- malt extract ager medium and incubated at 24-26ºC for 3 days for yeast count (Wickerham, 1951).

Mycotoxins extraction and Detection

Liquid medium was used for mycotoxins production (100 ml) then added 100 ml chloroform and homogenized at 16000rpm for 5 min according to Refaie (2013). Mycotoxins detection by using Thin Layer chromatography technique was adopted according to van Egmoned and Paulsch (1986).

Sensory Evaluation

All cheese samples were evaluated during ripening periods (30 days,60 days and 90days) for the flavor (50 points), body and texture (40 points) appearance (10 points) with total rating (100 points) according to Osman (2012) by the staff members of Dairy Science Department, Faculty of Agriculture, Assiut university, Egypt.

Statistical analysis

Data were statistically analyzed using the analysis of variance (ANOVA) for the F completely randomized design (CRD) as published by Gomez and Gomez (1984) using a computer software package "SAS". The Revised Least significant (RLSD) method was used to test the difference between treatment means at a 5% level of probability as described by Snedecor and Cohran (1980).

Results and Discussion

1. Gross chemical composition of Ras cheese

Data in Table (1) shows the effect of adding potassium sorbate (0.1%, 0.2% and 0.3%) on the chemical composition of Ras cheese during ripening period. The data show a progressive decrease in moisture content across all treatments during the ripening process. According to Conner (1980), this could be owing to water evaporation or protein binding during ripening. Results presented show that the control cheese contained higher moisture content when fresh than other treatments, followed by T3but T1 and T2 were the same in moisture content. In general, the mean content was higher in control with a value of 33.67% than the other treatments. All cheese treatments showed steady increases in titratable acidity peaking at the end of ripening. Cheese's acidity is naturally derived from milk components and develops throughout ripening. Abd El-Monem (2018) found that the degradation of protein and amino acid intermediates, as well as fatty acids from fat hydrolysis, significantly contributes to cheese's acidity. The acidity content of the cheese treatments was 0.62, 0.73, 0.63 and 0.54% for fresh C, T1, T2 and T3, respectively. However, the results were 1.77, 1.79, 1.05 and 1.44% after 90 days of storage period for the same previous treatments, respectively. The T1 cheese that supplanted with potassium sorbate 0.1% had significantly higher acidity than all treatments during the different stages of storage, while the T3 cheese supplemented with potassium sorbate 0.3% presented the lower acidity.

Table 1. Effect of potassium sorbate on the chemical properties of Ras cheese during ripening period

Items	Treat / Age	Fresh	15 days	30 days	60 days	90 days	Mean
Moisture	Control	38.33± 0.88 a	36.33± 0.88 abc	33.17 ±0.60 defg	31.00± 0.58 ghi	29.50 ±0.76 i	33.67± 0.92 A
	T1	36.17± 0.44 abc	34.00 ± 0.76 cdef	32.50± 0.76 efgh	31.33± 0.88 ghi	29.67 ±0.33 i	32.73±0.65 A
	T2	36.17 ±0.60 abc	34.92 ± 0.46 bcde	33.23± 0.72 defg	32.17± 0.60 fgh	31.00 ±0.58 ghi	33.50±0.55 A
	T3	37.17± 0.44 ab	35.50± 0.29 bcd	33.50 ± 1.04 defg	31.83 ±1.36 fghi	30.67 ±1.20 hi	33.73± .73 A
Mean		36.96±0.38 *A*	35.18±0.38 *B*	33.1±0.36 *C*	31.58± 0.41 *D*	30.20±0.39 *E*	
Acidity	Control	0.62±0.01 i	1.21± 0.17 ef	1.52 ±0.05 bcd	1.53± 0.05 bcd	1.77± 0.04 a	1.33±0.11 B
	T1	0.73±0.01 hi	1.5 ±0.03 bcd	1.62± 0.0 1 abc	1.70 ±0.01 ab	1.79± 0.02 a	1.47±0.10 A
	T2	0.63± 0.02 i	0.75± 0.02 hi	0.93 ±0.12 gh	1± 0.12 fg	1.05 ±0.12 fg	0.87±0.05 D
	T3	0.54 ±0.03 i	0.93 ±0.02 gh	1.43 ±0.18 cde	1.36± 0.01 de	1.42 ±0.01 cde	1.14±0.098 C
Mean		0.63±0.02 *D*	1.09± 0.09 *C*	1.38±0.09 *B*	1.40±0.08 *B*	1.51±0.1 *A*	
Fat	Control	28.67± 0.88 hi	30 ±.0.58 fghi	30.17± 0.73 fgh	31± 0.58 defg	33.33± 0.88 abc	30.63 ±0.50 B
	T1	28.5± 0.29 hi	31.33± 0.73 cdef	32.25± 0.52 abcde	32.83± 0.44 abcde	34.17 ±0.60 a	31.82± 0.55 A
	T2	28.5± 0.29 hi	29.17± 0.44 ghi	30.83± 0.44 efg	32.33 ±0.44 abcde	33.17 ±0.60 abc	30.8 ±0.51 B
	T3	28± 0.29 i	29.92 ± 0.22 fghi	31.83±0.60 Bcdef	33± 1 abcd	33.67 ±0.93 ab	31.28± 0.61 AB
Mean		28.42±0.22 *E*	30.10 ±0.32 *D*	31.27±0.35 *C*	32.29±0.37 *B*	33.58±0.35 *A*	
Salt	Control	1.7 ±0.17 i	2.58 ±0.03 gh	2.56 ±0.03 gh	2.61± 0.03 fgh	2.84± 0.09 e	2.46 ±0.11 C
	T1	1.63± 0.03 i	2.41± 0.01 h	2.7± 0.09 efg	2.77± 0.08 efg	3.2 ±0.11 cd	2.54± 0.14 C
	T2	1.83 ±0.01 i	2.82± 0.04 ef	3.32± 0.01c	3.57 ±0.04 b	3.68± 0.01 ab	3.04± 0.18 B
	T3	1.83 ±0.07 i	3.1± 0.06 d	3.33± 0.09 c	3.63± 0.09 ab	3.83± 0.09 a	3.15±0.19 A
Mean		1.75±0.05 *E*	2.72± 0.08 *D*	2.98±0.12 *C*	3.15± 0.14 *B*	3.39±0.12 *A*	
pH	Control	5.55± 0.003 a	5.42±0.01 a	5.33±0.01 a	5.30±0.003 ab	5.27±0.01 ab	5.37 ±0.03 AB
	T1	5.63 ±0.01 a	5.32±0.01 ab	4.27±0.1 bc	5.23±0.01 abc	5.2±0.006 abc	5.13± 0.20 B
	T2	5.78± 0.01 a	5.71±0.01 a	5.63±0.01 a	5.58±0.01a	5.52±0.01 a	5.64 ±0.02 A
	T3	5.25± 0.01 ab	5.22±0.01 abc	4.20±1.01 c	5.17±0.01 abc	5.12±0.01 abc	4.99 ±0.20 B
Mean		5.55±0.06 *A*	5.42±0.06 *A*	4.86±0.36 *B*	5.32±0.05 *AB*	5.28±0.05 *AB*	
TN	Control	2.63± 0.01 o	3.18±0.01 n	3.41±0.02 im	3.51±0.02 ijk	3.56±0.02 ghi	3.25± 0.09 D
	T1	3.22± 0.01 n	3.39±0.01 m	3.46±0.01 ki	3.55±0.01 hij	3.62±0.01 def	3.45± 0.04 C
	T2	3.49 ±0.01 jk	3.54±0.01 hij	3.58±0.01 fgh	3.66±0.02 cde	3.70±0.02 bc	3.59± 0.02 B
	T3	3.61± 0.02 efg	3.68±0.03 cd	3.72±0.03 bc	3.74±0.02 ab	3.78±0.01 a	3.71 ±0.01 A
Mean		3.24±0.11 *E*	3.45±0.06 *D*	3.54±0.04 *C*	3.61±0.03 *B*	3.67±0.03 *A*	
S.N.	Control	0.28 ±0.01 f	0.35 ±0.01 e	0.45± 0.01 cd	0.57 ±0.02 b	0.72 ±0.04 a	0.48 ±0.04 A
	T1	0.07 ±0.0 1 h	0.16± 0.01 g	0.24 ±0.01 f	0.36± 0.01e	0.42 ±0.01 d	0.25 ±0.03 C
	T2	0.06± 0.0 h	0.13± 0.01 g	0.24 ±0.02 f	0.36± 0.02 e	0.48±0.01 c	0.25 ±0.04 C
Mean		0.14±0.02 *E*	0.23±0.03 *D*	0.35±0.03 *C*	0.47±0.03 *B*	0.58±0.04 *A*	

Mean of treatments (Capital), Mean of age (*Capital Italic*), the interaction between treatments and age (small). Averages having the same letter are not significant at the 5% level according to Duncan's multiple range test. T1 Ras cheese with potassium sorbate 0.1%, T2 Ras cheese with potassium sorbate 0.2%, T3 Ras cheese with potassium sorbate 0.3%.

According to Abd El-Monem (2018) the fat content of cheese has a crucial role in its palatability and texture. Ras cheese had a modest rise in fat content, likely due to moisture loss during ripening and decreased non-fat ingredients due to microbe growth in all treatments (Kamaly, 1978; Ezzat, 1990). The results show that T1 was the highest in fat content during all ripening period followed by T3, T2 and control with mean values 31.82, 31.28, 30.8 and 30.63 respectively.

The salt contents of all cheese treatments decreased gradually that may be due to the loss of water caused by evaporation (El-Etriby *et al*., 1998) and Osmosis because of absorbing more of the sprinkled salt during the salting process. T 3 was the highest salt during all ripening period followed by T2, T1 and control, respectively.

The pH values of all cheese treatments decreased gradually and reached the minimum values at the end of the ripening process. The cheese supplanted with potassium sorbate 0.3% was the lowest pH values than the others.

The total nitrogen of all treatments increased reached the maximum values at the end of the ripening period. T3 was the highest value of T.N followed by T2, T1 and control with mean value of 3.71, 3.59, 3.45 and 3.25, respectively.

Control Ras cheese was the highest in soluble nitrogen value with mean value 0.48 followed by T3 with mean value 0.43, but T1 and T2 were the same mean value recorded 0.25.

2-Sensory Evolution of Ras cheese

Data in Table 2 and Fig 1 showed that the highest total point of mentioned organoleptic properties were given to cheese supplemented with potassium sorbate 0.3% during repining period (60 days) 96.5% points followed by cheese supplemented with potassium sorbate 0.2% in the same period with 91.5% points, while cheese supplemented with potassium sorbate 0.1% was the lowest in compression to others (0.2% and 0.3%) recorded 75.5% in the same ripening period.

Table 2. organoleptic properties of control Ras cheese and Ras cheese supplemented with three concentrations of potassium sorbate (0.1%, 0.2% and 0.3%)

Treatments	Ripening period (days)	Flavor (50)	Body & Texture (40)	Appearance (10)	Total (100)
Control	30 days	36	31.5	7	74.5
	60 days	34	31	7	72
	90 days	38.5	29.5	7	75
Potassium sorbet 0.1%	30 days	38.5	31	7.5	77
	60 days	34.5	33	8	75.5
	90 days	33	34	7.5	74.5
Potassium sorbet 0.2%	30 days	35	29	7.5	71.5
	60 days	44	37.5	9.5	91.5
	90 days	44.5	36.5	8	89
Potassium sorbet 0.3%	30 days	42.5	35	8.5	86
	60 days	48.5	39	9	96.5
	90 days	44.5	37	8.5	90

Control Ras cheese was the lowest points in all ripening period (74.5% in 30 days, 72% in 60 days and 75% in 90 days that may be affected by the increase in loss of moisture values may be play an important role in delaying the sensory properties in control cheese compared with other treatments that coated or supplemented with potassium sorbate. Our results agree with (Abolila *et al.,* 2017).

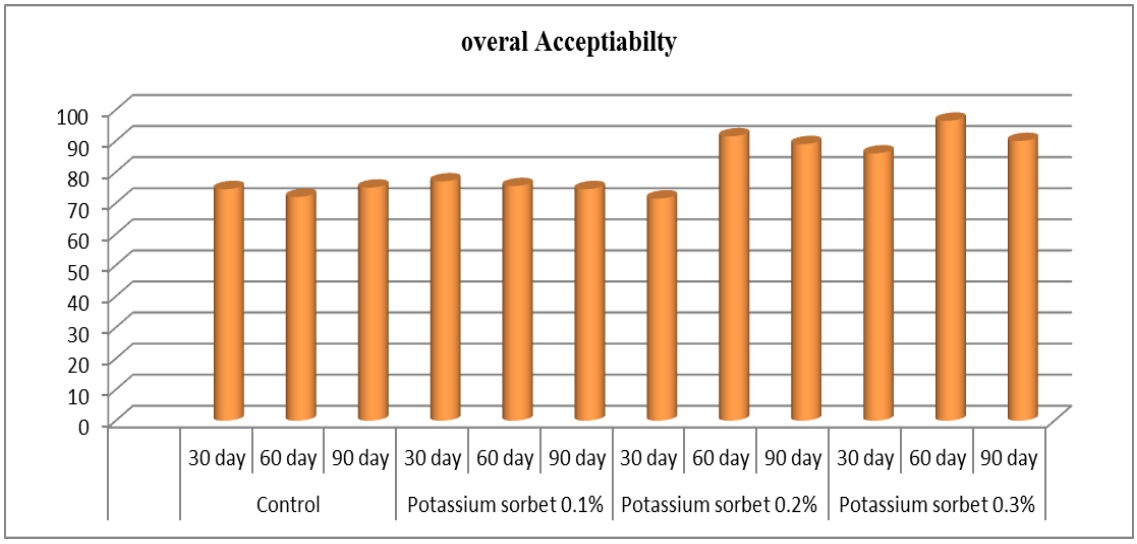

Fig.1. Organoleptic properties of Ras cheese supplemented with different concentrations of potassium sorbate

3- Fungi isolated from different treatments of Ras cheese

Data in Table (3) revealed that 9 species belonging to 4 genera of fungi were isolated from control Ras cheese and Ras cheese supplemented with potassium sorbate with different concentrations (0.1% - 0.2% and 0.3%) on potato Dextrose agar medium at 28± 1ºC. The results showed that control Ras cheese was the richest sample in fungal population giving rise to 13616 CFU/gm, followed by Ras cheese supplemented with potassium sorbate 0.1% giving 4850 CFU/gm. Ras cheese supplemented with potassium sorbate 0.2% recorded 2520 CFU/gm, while the lowest total counts involved in Ras cheese supplemented with potassium sorbate 0.3% by 1617 CFU/gm.

Aspergillus was the most prevalent genus isolated in all samples of Ras cheese including 6 species (*A. flavus*, *A. fumigatus*, *A. japonicus*, *A. niger*, *A. tammarii* and *A. terreus).* The highest population of *Aspergillus* was recovered from control Ras cheese giving 10966 CFU/gm Ras cheese followed by Ras cheese supplemented with pot. sorbate 0.1% giving 4000 CFU/gm, Ras cheese supplemented with pot. sorbate 0.2% contain 2270 CFU/gm from three species (*A. flavus, A. niger* and *A. terreus*), while the lowest population of *Aspergillus* was recovered from Ras cheese supplemented with pot. sorbate 0.3% giving 1517 CFU/gm from three species (*A. flavus, A. Niger* and *A. terreus*).

Cladosporium cladosporides isolated in high occurrence in all samples of cheese. It recorded in control cheese in all ripening period but involved in Ras cheese with pot. sorbate 0.1% and pot. sorbate 0.2% only in fresh and 15 days period. The

lowest occurrence was in Ras cheese with pot. sorbate 0.3% only in fresh time giving 100 CFU/gm.

Table 3. Total counts and percentage of total counts of Ras cheese contaminated fungi isolated on potato dextrose agar medium at 28±1°C

Fungal species	Ripening period	Control		Pot. sorbate 0.1%		Pot. sorbate 0.2%		Pot. sorbate 0.3%	
		T.C	% T.C	T.C	% T.C	T.C	% T.C	T.C	% T.C
A. flavus	Fresh	700	5.1	400	8.2	350	13.8	100	6.2
	15 days	500	3.7	250	5.2	200	8	100	6.2
	30 days	500	3.7	0	0.0	0	0.0	0	0.0
	60 days	150	1.1	0	0.0	0	0.0	0	0.0
	90 days	100	0.7	0	0.0	0	0.0	0	0.0
A. fumigatus	Fresh	100	0.7	100	2.1	0	0.0	0	0.0
	15 days	0	0.0	0	0.0	0	0.0	0	0.0
	30 days	0	0.0	0	0.0	0	0.0	0	0.0
	60 days	0	0.0	0	0.0	0	0.0	0	0.0
	90 days	0	0.0	0	0.0	0	0.0	0	0.0
A. japonicas	Fresh	500	3.7	300	6.2	0	0.0	0	0.0
	15 days	450	3.3	150	3.1	0	0.0	0	0.0
	30 days	400	2.9	100	2.1	0	0.0	0	0.0
	60 days	300	2.2	0	0.0	0	0.0	0	0.0
	90 days	250	1.8	0	0.0	0	0.0	0	0.0
A. niger	Fresh	1000	7.3	400	8.2	300	11.9	267	16.5
	15 days	800	5.9	400	8.2	300	11.9	250	15.5
	30 days	600	4.4	375	7.7	270	10.7	225	13.9
	60 days	533	3.9	325	6.7	200	8	150	9.3
	90 days	433	3.2	200	4.1	150	5.9	100	6.2
A. tamarii	Fresh	200	1.5	200	4.1	0	0.0	0	0.0
	15 days	100	0.7	100	2.1	0	0.0	0	0.0
	30 days	0	0.0	0	0.0	0	0.0	0	0.0
	60 days	0	0.0	0	0.0	0	0.0	0	0.0
	90 days	0	0.0	0	0.0	0	0.0	0	0.0
A. terreus	Fresh	800	5.9	300	6.2	300	11.9	175	10.8
	15 days	750	5.5	300	6.2	200	8	150	9.2
	30 days	700	5.1	100	2.1	0	0.0	0	0.0
	60 days	600	4.4	0	0.0	0	0.0	0	0.0
	90 days	500	3.7	0	0.0	0	0.0	0	0.0
Cladosporium cladosporioides	Fresh	600	4.4	400	8.2	150	5.9	100	6.2
	15 days	400	3	200	4.1	100	4	0	0.0
	30 days	350	2.6	0	0.0	0	0.0	0	0.0
	60 days	300	2.2	0	0.0	0	0.0	0	0.0
	90 days	300	2.2	0	0.0	0	0.0	0	0.0
Mucor hiemalis	Fresh	300	2.2	0	0.0	0	0.0	0	0.0
	15 days	200	1.5	0	0.0	0	0.0	0	0.0
	30 days	0	0.0	0	0.0	0	0.0	0	0.0
	60 days	0	0.0	0	0.0	0	0.0	0	0.0
	90 days	0	0.0	0	0.0	0	0.0	0	0.0
Trichoderma harzianum	Fresh	200	1.5	150	3.1	0	0.0	0	0.0
	15 days	0	0.0	100	2.1	0	0.0	0	0.0
	30 days	0	0.0	0	0.0	0	0.0	0	0.0
	60 days	0	0.0	0	0.0	0	0.0	0	0.0
	90 days	0	0.0	0	0.0	0	0.0	0	0.0
Total counts		**13616**		**4850**		**2520**		**1617**	
No. of genera		**4**		**3**		**2**		**2**	
No. of species & varieties		**9**		**8**		**4**		**4**	

TC= Total count, % TC= percentage of total count

Mucor hiemalis isolated in control Ras cheese in fresh and 15 days period but not detected in all treatments in all ripening period.

Trichoderma harzianum isolated in moderate occurrence from control Ras cheese and cheese supplemented with pot. sorbate 0.1% during ripening period fresh giving 200 CFU/gm and 150 CFU/gm of cheese, respectively.

In agreement with our results, *Geotrichum candidum*, *Aspergillus ochraceus*, *A. alliaceus*, *A. oryzae*, *A. niger*, *A. nidulans*, *Emericella nidulans*, *A. flavus*, *A. glaucus*, *A. flavipes*, *Penicillium* sp., *Mucor* sp. and *Rhizopus stolonifer* were isolated from Ras cheese (El-Fadaly *et al.,* 2015). *Aspergillus* was the most predominant and represented by four species namely *A. flavus, A. niger, A. ustus, and A. fumigatus* on surface of Egyptian Ras Cheese collected from different locations in Assiut City, Egypt (Seddek *et al*., 2016). *A. niger, A. fumgatus, Rhizopus stolonifera* and *Alternaria chlamydospora* were isolated from3 samples of Ras cheese collected from three locations in Assiut city according to (Zain El-din *et al*.,2022).

Data in Table 4 revealed that isolates of different *Aspergilus sp*, *Cladosporium*, *Mucar* and *Trichoderma* are isolated from different treatments of Ras cheese on potato dextrose ager at 28±1ºC were tested for aflatoxins (B1, B2, G1 and G2). Isolates of *A. flavus* showed positive aflatoxins (B1, B2, G1 and G2) in all treatments. It also shows positive Kojic acid production and free from Citrinin and Ochratoxin in all treatments. Isolates of A. *fumigatus* and *A. japonicas* show negative results for production of all types of mycotoxins (aflatoxins, Kojic acid Citrinin and Ochratoxin). Isolates of *A. niger show positive* aflatoxins B1production in all treatments but negative in the other types of aflatoxins. It also free from Citrinin and Kojic acid, while Isolates of *A. niger* can produce Ochratoxin in all treatments. *A. tamarii has the ability to produce aflatoxin G1and kojic acid in* Control and Ras cheese with pot. sorbate 0.1%, while Ras cheese with pot. sorbate 0.2% and 0.3% were free from all types of aflatoxins and other mycotoxins. *A. terreus* isolates free from all types of aflatoxins, kojic acid and Ochratoxin in all treatments, but show positive citrnin production in all treatments. All isolates of *Cladosporium cladosporioides, Mucor hiemalis and Trichoderma harzianum* were free from all types of mycotoxins in control, Ras cheese with 0.1%, 0.2% and 0.3% pot. sorbate .in agreement with our results, Shehab *et al.,* 2019indicated that the incidence of AFM1 in Ras cheese was (26.7%) and AFM2 in Ras cheese (13.3%) from all samples. Aflatoxins M1, M2, B1, B2, G1, G2 and Ochratoxin A were detected in the surface of examined pooled Ras cheese samples with an incidence rate of 41.66, 25, 33.3, 25, 16.6, 8.3 and 16.6%, respectively (Elramly *et al*., 2019).

Table 4. Mycotoxins produced by different fungal groups isolated from different treatments of Ras cheese on potato dextrose agar (PDA) at 28±10ºC

Fungal species	Treatments	Aflatoxins				Citrinin	Kojic acid	Ochratoxin
		B1	B2	G 1	G2			
A. flavus	Control	+ve	+ve	+ve	+ve	-ve	+ve	-ve
	Pot. 0.1%	+ve	+ve	+ve	+ve	-ve	+ve	-ve
	Pot.0.2%	+ve	+ve	+ve	+ve	-ve	+ve	-ve
	Pot. 0.3%	+ve	+ve	+ve	+ve	-ve	+ve	-ve
A. fumigatus	Control	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.1%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot.0.2%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.3%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
A. japonicus	Control	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.1%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot.0.2%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.3%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
A. Niger	Control	+ve	-ve	-ve	-ve	-ve	-ve	+ve
	Pot. 0.1%	+ve	-ve	-ve	-ve	-ve	-ve	+ve
	Pot.0.2%	+ve	-ve	-ve	-ve	-ve	-ve	+ve
	Pot. 0.3%	+ve	-ve	-ve	-ve	-ve	-ve	+ve
A. tamarii	Control	-ve	-ve	+ve	-ve	-ve	+ve	-ve
	Pot. 0.1%	-ve	-ve	+ve	-ve	-ve	+ve	-ve
	Pot.0.2%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.3%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
A. terreus	Control	-ve	-ve	-ve	-ve	+ve	-ve	-ve
	Pot. 0.1%	-ve	-ve	-ve	-ve	+ve	-ve	-ve
	Pot.0.2%	-ve	-ve	-ve	-ve	+ve	-ve	-ve
	Pot. 0.3%	-ve	-ve	-ve	-ve	+ve	-ve	-ve
Cladosporium cladosporioides	Control	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.1%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot.0.2%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.3%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Mucor hiemalis	Control	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.1%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot.0.2%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.3%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Trichoderma harzianum	Control	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.1%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot.0.2%	-ve	-ve	-ve	-ve	-ve	-ve	-ve
	Pot. 0.3%	-ve	-ve	-ve	-ve	-ve	-ve	-ve

Data in fig 2 showed the counts of yeasts (× 10^1,10^2,10^3 CFU/g cheese) in control and treated Ras cheese during the experimental ripening period. Yeast found in few numbers in cheese supplemented with potassium sorbate and these counts decrease in the mentioned cheese with the progress of ripening period and concertation. No yeasts were found in cheese supplemented with potassium sorbate 0.3% during ripening period (90 days). This indicates the effect of potassium sorbate on the number of yeasts, and this effect increases with increasing the concentration of potassium sorbate until it completely disappears in cheese treated with potassium sorbate0.3% at all dilutions during the storage period of 3 months. Our results agree with Egyptian standards (ES 2005) that refer to the count of yeasts in Ras cheese as not exceeding 2 log CFU/g (100 CFU/g).

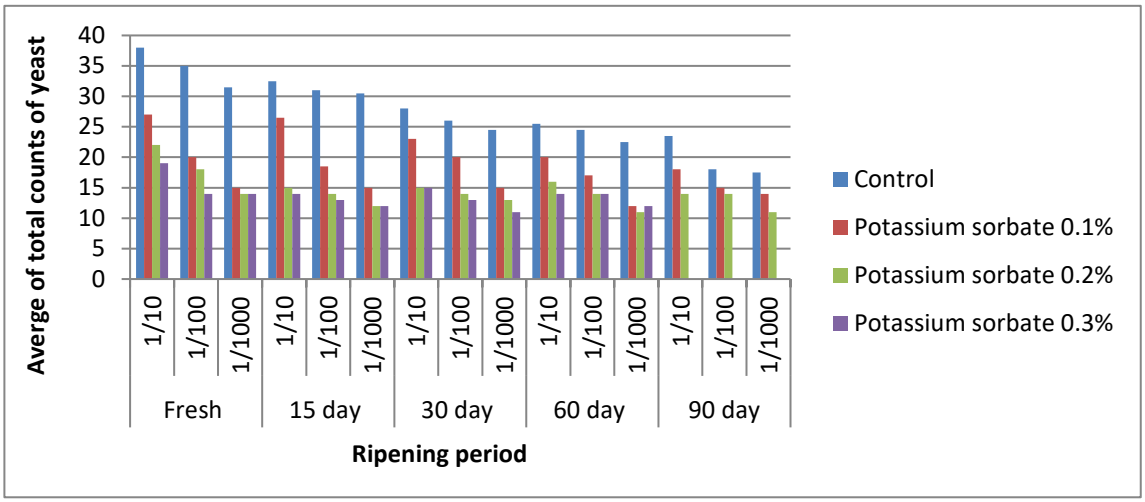

Fig. 2. changes of the average yeast count (× 10^1,10^2,10^3 CFU/g cheese) of control Ras cheese and cheese supplemented with potassium sorbate (0.1%, 0.2% and 0.3%) during ripening period

Data in fig 3 presents the average of total bacteria count of control Ras cheese and cheese with potassium sorbate (0.1%, 0.2% and 0.3%) during ripening period. Results indicated that control Ras cheese has the highest count of bacteria compared to cheese treated with potassium sorbate. Also, the treatment with potassium sorbate 0.3% was the best as it reduced the presence of bacteria during the ripening periods, which explains the role of potassium sorbate as an antimicrobial. Our results agree with the results of Nassib *et al.* (2010) who showed that Ras cheese treated with potassium sorbate led to a reduction in bacterial numbers and an improvement in the quality of the cheese compared to control Ras cheese.

Conclusion

Potassium sorbate is an efficient antifungal preservative for yeasts and molds at low concentrations. Ras cheese treatment with potassium sorbate can lengthen its shelf-life during refrigeration, and we can control of mycotoxins production which is beneficial for both manufacturers and customers. The use of potassium sorbate also led to an improvement in the chemical and sensory

properties, with the highest treatment in the sensory evaluation being the Ras cheese treated with potassium sorbate 0.3% recorded 96.5% for 60 days period.

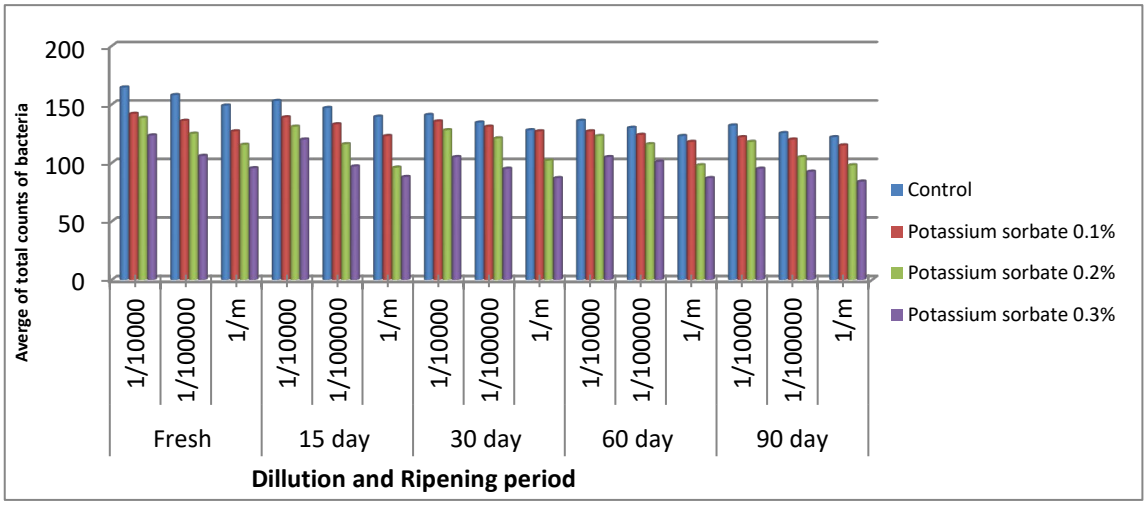

Fig. 3. Changes of the average Bacteria count (× 10^4, 10^5, 10^6 CFU/g cheese) of control Ras cheese and cheese supplemented with potassium sorbate (0.1%, 0.2% and 0.3%) during ripening period.

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تأثير سوربات البوتاسيوم على الخواص الكيميائية والميكروبيولوجية للجبن الراس خلال فترات التسوية

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الملخص

الهدف من هذه الدراسة توضيح دور سوربات البوتاسيوم E 202 (0.1%، 0.2% و0.3%) عن طريق رشه في صورة محلول علي سطح الجبن الراس على بعض الخواص الكيميائية (الرطوبة، الحموضة، الملح، الدهون، الرقم الهيدروجيني، النيتروجين الكلي والنيتروجين الذائب)، الخواص الميكروبيولوجية (العدد الكلي للبكتيريا، العدد الكلي للفطريات والخمائر وتأثيرها على نمو الفطريات وإنتاج الأفلاتوكسين) للجبن الراس خلال خمس فترات تسوية (طازج، 15 يوم، 30 يوم، 60 يوم، 90 يوم) والتقييم الحسي للجبن في 30 يوم، 60 يوم و90 يوم. أظهرت النتائج انخفاض في مستوي الرطوبة وpH في كل المعاملات بتقدم فترات التسوية، زادت كل من الحموضة وT.N وS.N في كل المعاملات بتقدم فتره التسوية وكانت اعلاهم المعاملة الثانية (جبن راس معامل بسوربات البوتاسيوم 0.2%). كما أظهرت دوراً جيداً لسوربات البوتاسيوم في تثبيط نمو الفطريات والخمائر مع زيادة تركيزه وبتقدم فترات التسوية ودوره في تثبيط قدرة الفطريات في انتاج السموم الفطرية .وكان الجبن المعامل بسوربات البوتاسيوم 0.3% افضل قبول في التقييم الحسي خلال فترة تسوية 60 يوم بمعدل 96.5% وهذا يعني أنه يمكننا استخدام سوربات البوتاسيوم كعامل مضاد للفطريات جيد لتحسين جودة الجبن الراس.

***الكلمات المفتاحية**: الجبن الجاف الخواص الكميائية، الخواص الميكروبيولوجية، سوربات البوتاسيوم، فترات التسوية.*