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Impact of Raw Materials and Processing Techniques on The Microbiological Quality of Egyptian Domiati Cheese

Shimaa, S Awaad*, Moawad AA, Ayah B, Abdel-Salam and Sallam SS

Department of Food Hygiene and Control, Faculty of Veterinary Medicine, Cairo University, Egypt
***Corresponding author:** Shimaa@cu.edu.eg

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ABSTRACT

Domiati cheese is the most popular brand of cheese ripened in brine in the Middle East in terms of consumed quantities. This study was performed to investigate the impact of the microbiological quality of the used raw materials, the applied traditional processing techniques and ripening period on the quality and safety of the produced cheese. Three hundred random composite samples were collected from three factories at Fayoum Governorate, Egypt. Collected samples represent twenty-five each of: raw milk, table salt, calf rennet, microbial rennet, water, environmental air, whey, fresh cheese, ripened cheese and swabs from: worker hands; cheese molds and utensils; tanks. All samples were examined microbiologically for Standard Plate Count (SPC), coliforms count, *Staphylococcus aureus* (*S. aureus*) count, total yeast and mould count, presence of *E. coli*, *Salmonellae* and *Listeria monocytogenes* (*L. monocytogenes*). The mean value of SPC, coliforms, *S. aureus* and total yeast and mould counts ranged from (79×10^2 CFU/m^3 for air to 13×10^8 CFU/g for fresh cheese), (7×10^2 MPN/ cm^2 for tank swabs to 80×10^6 MPN/ml for raw milk), (9×10^2 CFU/g for salt to 69×10^6 CFU/g for fresh cheese) and (2×10^2 CFU/cm^2 for hand swabs to 60×10^4 CFU/g for fresh cheese), respectively. Whereas, *E. coli*, *Salmonella* and *L. monocytogenes* failed to be detected in all examined samples. There were significant differences in all determined microbiological parameters ($P\leq0.05$) between fresh and ripened cheese which may be attributed to different adverse conditions such as water activity, pH, salt content and temperature carried out to improve the quality of the product.

Key words: Domiati cheese, Ripening period, Raw materials, Processing techniques

INTRODUCTION

White-brined cheeses are produced in border countries of the Mediterranean Sea and some Balkan countries (Atanasova *et al*., 2020). These cheeses are produced under different names and the best-known brands are: Feta cheese (in Greece), Domiati (in Egypt), Beyaz peynir (in Turkey) and Halloumi (in Cyprus). As a consequence of long-term brining, ripened cheeses have a salty, acidic, and sometimes a piquant taste. These cheeses have no rind, no gas holes, and a close texture (McSweeney *et al*., 2017).

In Egypt, Domiati cheese is the most popular soft white pickled cheese variety and is named after the city and governorate of Damietta and it makes up about 75% of the cheese produced and consumed in Egypt (Zhang *et al*., 2003).

Domiati cheese is made from cow or buffalo milk or a mixture of them. It is consumed as fresh or after being ripened in brine for a period of 2-4 months. Two types of Domiati cheese are sold in the market depending upon the ripening temperature. Cheese ripened at room temperature is known as Istamboli cheese, and that ripened at 10°C is called Baramili cheese (El-Baradei *et al*., 2007; McSweeney *et al*., 2017).

Domiati cheese differs chiefly from other pickled cheese varieties by the fact that milk is salted at first step before fermentation and coagulation. The proportion of added salt (10-15%) depends on the season of manufacture and on the temperature of cheese ripening (McSweeney *et al*., 2017). Therefore, the quality of Domiati cheese is the major area of concern for producers and consumers. It depends on the types of microorganisms introduced from raw materials, processing techniques and hygienic practices applied in dairy plant, so handling of milk during cheese making plays an important role in the proliferation of microbial flora and consequently impair its utility and may render the product unfit for human consumption (Aly and Galal, 2002).

Microbiological testing may often be required to verify that raw materials are delivered in agreement with local specifications and as a mean of monitoring for selecting and approving the suppliers (McMeekin, 2003).

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The application of advanced technologies such as ultrafiltration, besides implementation of food safety management systems in large scale cheese processing plants has eliminated several risks associated with the product. However, Domiati cheese is still processed in small and medium-sized processing facilities from warmed but not pasteurized milk. Warming of milk is used to dissolve the salt and for aiding coagulation (El-Baradei *et al.*, 2007). The low pH and high salt content are two factors contributing to the inactivation of bacterial pathogens during the 60 days ripening period of the product (Shehata *et al.*, 2007).

Total aerobic count, coliforms count and total yeast and mould counts are used as indicators for the quality of cheese (McMeekin, 2003). The department of Food Hygiene and Control at the Faculty of Veterinary Medicine, Cairo University, Egypt had received complaints from the owners of three Domiati cheese family business small factories at Fayoum Governorate complaining from low quality cheese result in economic losses to the owners, besides affecting their reputation as Domiati cheese producers since more than 50 years.

In trials to investigate the real cause of the problem, this study was designed to through light on the following topics:

1. Assessment of the impact of the microbiological quality of raw materials, the applied traditional processing techniques and ripening period on the quality of the produced Domiati cheese in the three cheese factories.
2. Depending upon the obtained results, further work will be planned to improve the quality of ripened Domiati cheese.

MATERIALS AND METHODS

The three dairy factories were visited and found to be neighboring family business (not really separate dairy factories). They use the same raw materials supplied by the same suppliers and applied the same traditional processing techniques under the similar environmental conditions. Therefore, the study depends on composite samples from the three factories.

Collection and microbiological examination of samples

Three hundred random composite samples were collected from the three factories. Collected samples represent twenty-five each of: raw milk, table salt, calf rennet, microbial rennet (CHY-MAX® Powder Extra NB), water, environmental air, whey, fresh cheese, ripened cheese and swabs from: worker hands; cheese molds and utensils; tanks.

Collected samples were transferred immediately in insulated ice box to be examined for SPC, coliforms count, *S. aureus* count, yeast count, mould count, total yeast and mould count, and presence of *E. coli*, *Salmonella* and *L. monocytogenes* (APHA, 2004). The obtained results were calculated as CFU/ml, g, cm^2 or m^3 as recommended by (Awad and Mawla, 2012).

Statistical analysis according to SPSS version 25

The obtained results were statistically analyzed for:

Paired T-test: Multiple linear regressions using stepwise selection method.

RESULTS

The results of microbiological examination of 300 random samples revealed that; the mean values of SPC for (raw milk, table salt, calf rennet, microbial rennet (CHY-MAX® Powder Extra NB), water, environmental air, whey, fresh cheese, ripened cheese and swabs from: worker hands; cheese molds and utensils; tanks) were ($41\times10^7\pm14\times10^7$ CFU/ml, $15\times10^4\pm7.5\times10^4$ CFU/g, $60\times10^7\pm16\times10^7$ CFU/ml, $41\times10^4\pm18\times10^4$ CFU/ g, $26\times10^6\pm8.5\times10^6$ CFU/ ml, $79\times10^2\pm 5\times10^2$ CFU/m^3, $11\times10^8\pm3.9\times10^8$ CFU/ml, $13\times10^8\pm2.6\times10^8$ CFU/g, $43\times10^6\pm 14\times10^6$ CFU/g, $10\times10^4\pm 4.7\times10^4$ CFU/ cm^2, $62\times10^4\pm 47\times10^4$ CFU/ cm^2 and $85\times10^3\pm 51\times10^3$ CFU/ cm^2, respectively). While the mean values of the previously mentioned samples of coliforms were ($80\times10^6\pm9.5\times10^6$ MPN/ml, $9\times10^2\pm2\times10^2$ MPN/g, $28\times10^6\pm8.5\times10^6$ MPN/ml, not detected, $21\times10^2\pm12\times10^2$ MPN/ ml, not applicable, $29\times10^6\pm8.5\times10^6$ MPN/ml, $60\times10^6\pm10\times10^6$ MPN/g, not detected, $14\times10^2\pm 4\times10^2$ MPN/ cm^2, $56\times10^2\pm 24\times10^2$ MPN/ cm^2 and $7\times10^2\pm 4\times10^2$ MPN/ cm^2, respectively) (Table 1).

In (Table 2), the mean values of the same mentioned samples for fungi were ($32\times10^4\pm8\times10^4$ CFU/ ml, $39\times10^2\pm14\times10^2$CFU/g, $53\times10^3\pm16\times10^3$ CFU/ml, $7\times10^2\pm1\times10^2$ CFU/ g, $17\times10^4\pm11\times10^4$ CFU/ml, $19\times10^2\pm 3\times10^2$ CFU/m^3, $42\times10^4\pm10\times10^4$ CFU/ml, $60\times10^4\pm14\times10^4$ CFU/g, $3\times10^4\pm 0.9\times10^4$CFU/g, $2\times10^2\pm 0.5\times10^2$ CFU/ cm^2, $24\times10^2\pm 11\times10^2$ CFU/cm^2 and $2.5\times10^2\pm 1.2\times10^2$ CFU/ cm^2, respectively). While, in (Table 3) the mean values for *S. aureus* were ($25\times10^6\pm8\times10^6$ CFU/ ml, $9\times10^2\pm2\times10^2$ CFU/g and $12\times10^6\pm8\times10^6$ CFU/ml, not detected, not detected, not applicable, $10\times10^6\pm5\times10^6$ CFU/ml, $69\times10^6\pm27\times10^6$ CFU/g, $17\times10^2\pm 15\times10^2$ CFU/g, $2\times10^2\pm 1\times10^2$ CFU/ cm^2, $32\times10^2\pm 22\times10^2$ CFU/ cm^2 and 3.5 ± 2.5 CFU/cm^2, respectively).

E. coli, *Salmonella spp* and *L. monocytogenes* failed to be detected in all examined samples (Table 4). There were significant differences ($P\leq0.05$) for SPC, coliforms, yeast, mould, fungi, *S. aureus* between fresh and ripened cheese (Table 6).

DISCUSSION

Due to their high and diverse nutrients content, high water content and almost neutral pH; milk and dairy products are highly nutritious food for human beings, and they also serve as an ideal medium for the growth of many types of microorganisms (Touch and Deeth, 2009).

Traditionally, in countryside Domiati cheese is made from raw milk where cheese makers rely on the natural microflora of such milk for acidification, shortening the ripening time and acquiring the produced cheese its distinctive rich flavor popular to the consumer (Bintsis and Papademas, 2002), which is contrary to the ES: 1008-3/ 2005 that recommend pasteurization of milk or being treated by any heat treatment equivalent to it. Whereas large scale produced Domiati cheese has less distinctive flavor as it is made from heat treated milk fortified with starter culture (McSweeney *et al*., 2017; Barac *et al*., 2019).

In Domiati cheese, table salt is commonly added at a percentage of 10% to enhance the flavor and as a preservative through lowering the water activity and increasing the osmotic pressure (McSweeney, 2007).

Table 1: Statistical analytical results of Standard Plate count and Coliforms for different examined samples (25 each)

Samples results		Raw milk*	Salt**	Calf rennet*	Microbial rennet**	Water*	Hand swabs***	Cheese molds and utensils swabs***	Tank swabs***	Air****	Whey*	Fresh cheese**	Ripened cheese**
Standard Plate Count (CFU)													
Positive samples	No.	25	24	25	25	25	25	25	25	25	25	25	25
	%	100	96	100	100	100	100	100	100	100	100	100	100
Min.		60×10^5	20×10^2	74×10^4	10×10^2	12×10^4	4×10^2	4×10^2	2×10^2	36×10^2	33×10^5	24×10^6	80×10^2
Max.		34×10^8	18×10^5	24×10^8	33×10^5	18×10^7	12×10^5	12×10^6	13×10^5	13×10^3	90×10^8	41×10^8	22×10^7
Mean		41×10^7	15×10^4	60×10^7	41×10^4	26×10^6	10×10^4	62×10^4	85×10^3	79×10^2	11×10^8	13×10^8	43×10^6
±*SEM*		14×10^7	7.5×10^4	16×10^7	18×10^4	8.5×10^6	4.7×10^4	47×10^4	51×10^3	5×10^2	3.9×10^8	2.6×10^8	14×10^6
Coliforms (MPN)													
Positive samples	No.	25	8	25	0	6	24	23	18	N/A	25	25	0
	%	100	32	100	0	24	96	92	72	N/A	100	100	0
Min.		4×10^4	4×10^2	23×10^3	ND	7×10^2	6	12	3	N/A	21×10^4	30×10^4	ND
Max.		11×10^7	46×10^2	11×10^7	ND	30×10^3	75×10^2	48×10^3	11×10^3	N/A	11×10^7	11×10^7	ND
Mean		80×10^6	9×10^2	28×10^6	ND	21×10^2	14×10^2	56×10^2	7×10^2	N/A	29×10^6	60×10^6	ND
±*SEM*		9.5×10^6	2×10^2	8.5×10^6	ND	12×10^2	4×10^2	24×10^2	4×10^2	N/A	8.5×10^6	10×10^6	ND

* CFU or MPN/ml; **CFU or MPN/g; ***CFU or MPN/cm^2; ****CFU or MPN/cm^3; ND=Not detected; N/A=Not Applicable

Table 2: Statistical analysis results of total yeast and mould counts for different examined samples (25 each)

Samples Results		Raw milk*	Salt**	Calf rennet*	Microbial rennet**	Water*	Hand swabs***	Cheese molds and utensils swabs***	Tank swabs***	Air****	Whey*	Fresh cheese**	Ripened cheese**
Yeast count (CFU)													
Positive samples	No.	25	20	21	16	15	20	21	9	25	25	25	25
	%	100	80	84	64	60	80	84	36	100	100	100	100
Min.		30×10^2	1×10^2	20×10^2	1×10^2	20×10^2	2	4	6	50	10×10^2	2×10^3	10×10^2
Max.		16×10^5	25×10^3	29×10^4	33×10^2	18×10^5	9×10^2	23×10^3	15×10^2	70×10^2	21×10^5	24×10^5	19×10^4
Mean		31×10^4	40×10^2	57×10^3	7×10^2	17×10^4	2×10^2	23×10^2	177	14×10^2	42×10^4	60×10^4	33×10^3
±*SEM*		8×10^4	15×10^2	18×10^3	2×10^2	11×10^4	0.5×10^2	12×10^2	112	0.3×10^2	10×10^4	14×10^4	9×10^3
Mould count (CFU)													
Positive samples	No.	13	12	16	14	0	12	14	14	24	12	16	22
	%	52	48	64	56	0	48	56	56	96	48	64	88
Min.		2×10^2	1×10^2	3×10^2	1×10^2	ND	1	4	1	29	1×10^2	100	10
Max.		20×10^3	4×10^2	20×10^3	3×10^2	ND	8	16×10^2	13×10^2	13×10^2	10×10^3	90×10^3	15×10^2
Mean		70×10^2	2×10^2	73×10^2	2×10^2	ND	3	2×10^2	116	514	24×10^2	74×10^2	3×10^2
±*SEM*		18×10^2	0.3×10^2	14×10^2	0.2×10^2	ND	0.8	1×10^2	92	76	10×10^2	3×10^2	0.8×10^2
Total yeast and mould count (CFU)													
Positive samples	No.	25	21	25	20	15	20	22	16	25	25	25	25
	%	100	84	100	80	60	80	88	64	100	100	100	100
Min.		30×10^2	1×10^2	10×10^2	1×10^2	20×10^2	2	20	1	350	50×10^2	3×10^3	10×10^2
Max.		16×10^5	25×10^3	31×10^4	33×10^2	18×10^5	9×10^2	23×10^3	15×10^2	73×10^2	21×10^5	24×10^5	19×10^4
Mean		32×10^4	39×10^2	53×10^3	7×10^2	17×10^4	2×10^2	24×10^2	2.5×10^2	19×10^2	42×10^4	60×10^4	3×10^4
±*SEM*		8×10^4	14×10^2	16×10^3	1×10^2	11×10^4	0.5×10^2	11×10^2	1.2×10^2	3×10^2	10×10^4	14×10^4	0.9×10^4

* CFU /ml; **CFU/ g; ***CFU/ cm^2; ****CFU/ cm^3; ND=Not detected

Table 3: Statistical analytical results of *S. aureus* count in examined samples (25 each)

Samples results		Raw milk*	Salt**	Calf rennet*	Microbial rennet**	Water*	Hand swabs***	Cheese molds and utensils swabs***	Tank swabs***	Air****	Whey*	Fresh cheese**	Ripened cheese**
S. aureus count (CFU)													
Positive samples	No.	11	3	7	0	0	3	4	2	N/A	9	15	2
	%	44	12	28	0	0	12	16	8	N/A	36	60	8
Min.		76×10^4	2×10^2	14×10^2	ND	ND	11	16	1	N/A	20×10^4	30×10^5	2×10^2
Max.		90×10^6	27×10^2	64×10^6	ND	ND	4×10^2	96×10^2	6	N/A	51×10^6	42×10^7	32×10^2
Mean		25×10^6	9×10^2	12×10^6	ND	ND	2×10^2	32×10^2	3.5	N/A	10×10^6	69×10^6	17×10^2
±*SEM*		8×10^6	2×10^2	8×10^6	ND	ND	1×10^2	22×10^2	2.5	N/	5×10^6	27×10^6	15×10^2

* CFU /ml; **CFU/ g; ***CFU/ cm^2; ****CFU/ cm^3; ND=Not detected; N/A=Not Applicable.

Table 4: Statistical analytical results for presence of some pathogens in examined samples (25 each)

Samples Pathogen	Raw milk	Salt	Calf rennet	Microbial rennet	Water	Hand swabs	Cheese molds and utensils swabs	Tank swabs	Air	Whey	Fresh cheese	Ripened cheese
E.coli	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Salmonella spp.	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
L. monocytogenes	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND=Not detected.

Table 5: Degree of acceptability of ripened Domiati cheese samples vs. Egyptian specifications for Domiati cheese (ES: 1008-3/2005).

Parameters	Critical limit	Acceptable		Not acceptable	
		No.	%	No.	%
Coliforms	Not more than 10 MPN/ g	25	100	0	0
Yeast count	Not more than 400 CFU/ g	0	0	25	100
Mould count	Not more than 10 CFU/ g	5	20	20	80
S. aureus	absent/ g	23	92	2	8
E. coli	absent/ g	25	100	0	0
Salmonellae	absent/ 25g	25	100	0	0
Listeria monocytogenes	absent/ g	25	100	0	0

Table 6: Effect of ripening period on the microbiological quality of the produced Domiati cheese

	Fresh cheese (Before ripening)	Ripened cheese (After ripening)	P-value
SPC	$13\times10^{8}\pm2.6\times10^{8}$ (a)	$43\times10^{6}\pm14\times10^{6}$ (b)	<0.0001
Coliforms	$60\times10^{6}\pm10\times10^{6}$ (a)	N.D (b)	<0.0001
S. aureus	$42\times10^{6}\pm17\times10^{6}$ (a)	$1.4\times10^{2}\pm1\times10^{2}$ (b)	0.025
Yeast count	$60\times10^{4}\pm14\times10^{4}$ (a)	$33\times10^{3}\pm9\times10^{3}$ (b)	0.001
Mould count	$74\times10^{2}\pm3\times10^{2}$(a)	$3\times10^{2}\pm0.8\times10^{2}$ (b)	0.001
Total yeast and mould count	$60\times10^{4}\pm14\times10^{4}$ (a)	$3\times10^{4}\pm0.9\times10^{4}$ (b)	0.001

Data presented as mean±SEM. Rows with different superscript letters (a and b) are significantly different at P≤0.05.

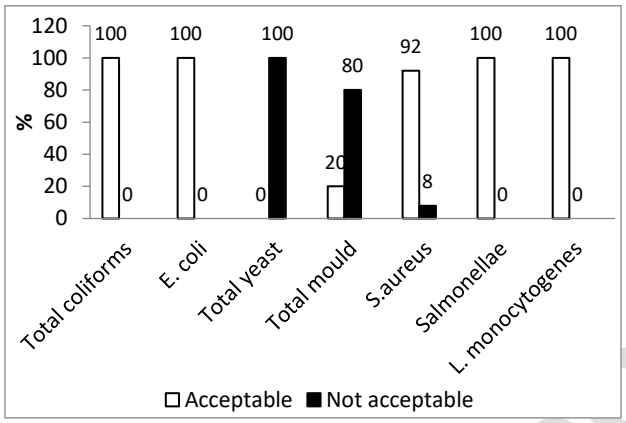

Fig. 1: Degree of acceptability of Domiati cheese samples vs. Egyptian Specifications for Domiati cheese (ES: 1008-3/2005).

Coagulation of milk with rennet is the first main step in the production of different varieties of rennet coagulated cheese (Tamime, 2006). The traditional dairies use rennet paste prepared locally from curdled milk in abomasa of slaughtered suckling calves (Tamime, 2006; Hassan and Aita, 2011). The national legal and statutory requirements prohibit slaughtering of calves before reaching the age of 2 years, which lead to shortage in calf rennet, therefore, the traditional factories use dried microbial rennet (CHY-MAX® Extra) to help calf rennet in coagulation of Domiati cheese.

In cheese industry, water is used for cleaning of hands, food contact surfaces, equipment, housekeeping or as a part of the product. Therefore, the quality of water is of concern during production of cheese to safeguard it from contamination (Habes *et al.*, 2015).

In food production industry, food handlers are considered as a potential source for contamination (Malavi *et al.*, 2018). The health status of the food handlers, their personal hygiene, knowledge and their good hygienic practices play an important role in food quality. It was estimated that 10–20% of foodborne illness outbreaks result from contamination through food handlers (Allam *et al.*, 2016). Cleaning and sanitizing of food contact surfaces in cheese making are proposed as effective measures in preventing cross contamination and prevalence of food borne diseases (Baghapour *et al.*, 2014).

Air quality is a good index of the overall hygienic and sanitary conditions adopted during production (McMeekin, 2003). Whey is a byproduct that is rich with aromatic compounds which support organoleptic quality and survival of microorganisms. (Bouymajane *et al.*, 2018). Fresh Domiati cheese is hold for at least 60 days for ripening in tins of different weights before consumption (Jay *et al.*, 2005). SPC is used as a good indicator for monitoring the sanitary conditions practiced during production, collection, handling and transport of raw materials (Blackburn, 2006).

Results presented in Table (1) show that the SPC of the examined raw milk, table salt, calf rennet and microbial rennet; were higher than those obtained by Sultana *et al.* (2014). Microbial quality specification for raw milk requires SPC less than 300 000 CFU/ml for commingled milk prior to pasteurization (Jay *et al.*, 2005). All the examined raw milk samples used in dairy factories under study exceeded the acceptable value.

While the mean values of SPC of the examined water, hand swabs, cheese molds and utensils swabs, tank swabs and air samples (Table 1) were nearly similar to those reported by Gamal *et al.* (2014). All the examined water samples failed to comply with ES: 190-1/2007 for drinking water which stated that SPC must not exceed 50 CFU/ml for drinking water.

All examined samples (100%) of workers hand swabs, food contact surfaces (cheese mould and utensils swabs and tank swabs) and air exceeded the recommended values given by Lambrechts *et al.* (2014) (<100 CFU/cm^2); Griffiths, 1997 (<100 CFU/cm^2) and APHA, 1992 (90 CFU/m^3), respectively. Whereas the mean values of SPC of the examined whey, fresh cheese and ripened cheese samples (Table 1) were higher than those recorded by Hassan and Gomaa (2016).

It was observed statistically that for each one SPC CFU/m^3 increase in air, a 12×10^6 CFU/g increase in SPC of fresh cheese ($F_{1,24}$ =6.101, P=0.021, R^2=0.21). There was a significant difference (P≤0.05) for SPC between fresh and ripened cheese (Table 6), which may attributed to the high salt content (10%), the low pH (~3.3) and anaerobic condition inside the cheese tins (Fox *et al.,* 2004). The results in Table 1 show that coliforms of the examined raw milk, table salt, calf rennet and microbial rennet were nearly similar to those obtained by Gamal *et al.* (2014).

Jones and Sumner (1999) and Ruegg (2003) stated that a coliform count less than 100 MPN/ml or g is considered acceptable for milk intended to be pasteurized. Therefore, all examined raw milk samples were considered unacceptable according to this level of acceptance.

The obtained results in Table 1 of coliform contamination of water, hand swabs, cheese molds and utensils swabs, tank swabs s and air were nearly similar to those recorded by Shash *et al.* (2010). Of all the samples, 24% of the examined water samples failed to meet the requirements of ES: 190-1/2007 for treated drinking water which must not exceed 1MPN/100ml. All examined ripened cheese samples comply with ES: 1008-3/2005 for coliforms (Table 5; Fig. 1). The results of ripened cheese were lower than those obtained by Hassan and Gomaa (2016).

Fungal contamination is mainly of air borne source, most fungi are tolerant to high salt content of the cheese up to 15% NaCl, can grow in wide range of pH (1.0–10.0) and temperature which may results in their survival in the final product resulting in spoilage of the cheese and may affect consumer health (Garnier *et al.*, 2017).

The results revealed in Table 2 showed that the mean values of yeast and mould of the examined raw milk, table salt, calf rennet and microbial rennet were similar to those recorded by Hassan and Aita (2011). While the obtained results of fungi of the examined water, hand swabs, cheese molds and utensils, tank swabs and air samples were higher than those reported by Vinayananda *et al.* (2018).

The obtained results of fungi of the examined whey, fresh cheese and ripened cheese samples was nearly similar to those recorded by Hassan and Gomaa (2016).

There was statistical analysis reveal a significant difference in fungal count (P≤0.05) between fresh and ripened cheese (Table 6), this may be attributed to that some strains of fungi could not tolerate high salt content and anaerobic condition of ripening (Garnier *et al.*, 2017).

It was observed statistically that for each one CFU/ ml increase in raw milk for total yeasts and moulds count, a 0.768 CFU/g increase in total yeasts and mould of fresh cheese ($F_{1,24}$ =8.232, P=0.002, R^2=0.428), while for each one CFU/ ml increase in whey for total yeasts and moulds count, a 0.714 CFU/g increase in total yeasts and moulds of fresh cheese ($F_{1,24}$ =6.931, P=0.015, R^2=0.232).

Yeasts were detected in 100% of the examined ripened cheese samples. Therefore, 100% of the examined samples were not complying with the ES: 1008-3/2005 (Table 5; Fig. 1). It was observed statistically that for each one CFU/ml increase in whey for yeasts, a 0.721 CFU/g increase in yeasts of fresh cheese was found ($F_{1,24}$=7.043, P=0.014, R^2=0.201).

On the other hand, moulds were detected in 88% of the examined ripened cheese samples. Only 20% of Domiati cheese samples were complying with the ES: 1008-3/2005 (Table 5; Fig. 1). *S. aureus* can tolerate a pH between 4 to 10 and a salt concentration of 0 to 20%. *S. aureus* enterotoxins are being identified as a major source of food-borne toxic infection; these may explain the importance of contamination of Domiati cheese with *S. aureus* during its manufacture. (Ahmed *et al.,* 2019)

Sources of *S. aureus* almost always originated from raw milk, food handlers or contaminated utensils. Thus, neglected personal hygiene and medical supervision may result in contamination of food under preparation with *S. aureus* which may lead to toxic infection (Fox *et al*., 2017).

These results presented in Table 3 show that the mean values of *S. aureus* of the examined raw milk, table salt, calf rennet and microbial rennet were higher than those reported by Ahmed *et al.* (2019).

According to ES: 154-1/2005 for raw milk, *S. aureus* count must not exceed 1×10^2 CFU/ml in raw milk, all positive raw milk samples (44%) failed to meet the national standard. Whereas, *S. aureus* failed to be detected in all examined water samples. So, the obtained results revealed that 100% of the examined water samples comply with ES: 190-1/2007.

The results of *S. aureus* of the examined water, hand swabs, cheese moulds and utensils swabs and tanks swabs were higher than those recorded by Alrabadi (2017). Results for ripened cheese of *S. aureus* were nearly similar to those reported by Sharaf *et al.* (2014). Only 2 samples (8%) of the examined ripened Domiati cheese were not complying with the ES: 1008-3/2005 (Table 5; Fig. 1)

Statistical analysis revealed a significant difference (P≤0.05) between fresh and ripened cheese (Table 6), which may be attributed to the fact that certain strains of *S. aureus* that are evidently sensitive to NaCl and exhibit autolysis even in the presence of a relative low concentration of NaCl (Ochiai, 1999).

E. coli, *Salmonella spp* and *L. monocytogenes* failed to be detected in all examined samples (Table 4). The obtained results were similar to those already reported (Hassan and Gomaa, 2016; Mehmood *et al.,* 2020). The obtained results for *E. coli*, *Salmonella spp* and *L. monocytogenes* indicated compliance of raw milk, water and ripened cheese samples with the ES: 154-1/2005, ES: 190-1/2007 and ES: 1008-3/2005, respectively (Table 5 and Fig. 1).

Conclusion

Regardless the quality of used raw materials and the processing techniques, ripening period seemed to have significant effect on improving the quality of the final product through the prevailed adverse conditions as water activity, pH, salt content, temperature and anaerobic condition within the cheese. The diverse of microorganisms and their counts in some raw materials, food handlers and food contact surfaces, besides, the processing conditions have impact on the quality and safety of the product.

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