Original Research Article
ISSN (Print) 2786-0272 ISSN (Online) 2786-0280

Evaluation of Some Locally Fermented Milk Products in New Valley Governorate

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Submitted 8/31/2022
Accepted 12/24/2022

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Abstract

One hundred and forty random samples of yoghurt and Laban raib, seventy of each, were collected from different localities and villages in El-Kharga city, New Valley Governorate, Egypt, for chemical and bacteriologic examination. The obtained results of yoghurt samples showed that the average values of acidity%, total *coliforms* and fecal *coliform*, *S. Aureus*, yeasts, and molds were 1.13, 6.8×10^4, 5.5×10^4, 1.42×10^4, 7.2×107 and 4.9×10^6 CFU/ml, respectively. On the other hand, in Laban raib samples, they were 1.13, 1.6×10^5, 1.1×10^5, 2.4×10^3, 2.1×107 and 2.8×10^5 CFU/ml, respectively. The prevalence of *E. coli* and anaerobic bacteria in yoghurt samples was 11.4 and 2.8 %, while in Laban raib samples was 8.5 and 4.3%. *Enterococci* couldn't be detected in all examined yoghurt and Laban raib samples. The aim of this research was to investigate the chemical and bacteriological characteristics of locally produced yoghurt and Laban raib in El-Kharga city, New Valley governorate, Egypt. Based on the results, both yoghurt and Laban raib samples showed high levels of total *coliforms*, fecal *coliforms*, and molds. Additionally, the prevalence of *E. coli* was relatively high in both types of samples, which may indicate poor hygiene practices during production or handling. However, the absence of *Enterococci* in all examined samples is a positive finding. These results suggest the need for improved hygiene and quality control measures in the production of these dairy products in the region. In conclusion, this study highlights the importance of monitoring the quality of locally produced dairy products and implementing appropriate measures to ensure their safety and hygiene.

Keywords: Bacteriological examination, Chemical examination, Fermented milk, Laban raib.

Introduction

Fermented milk has attained considerable and increasing popularity during the last few decades. A variety of wholesome, delicious, and palatable forms of fermented milk could be prepared by using various kinds of starters whose activities were controlled by regulating the holding temperature of milk. According to the methods of treatment used different kinds of fermented milk are now well known all over the world.

Yoghurt is one of the most popular fermented dairy products all over the world. It is a mixture of milk (whole, reduced-fat, low fat or nonfat) and cream fermented by a culture of lactic acid-producing bacteria. Laban raib also is important fundamental fermented milk for the Egyptian peasants which is consumed by the farmers or used for manufacturing kareish cheese.

Therefore, in our country, the most popular kinds of fermented milk are yoghurt and Laban raib. The production method of Laban raib and its handling is still primitive and unhygienic, and most yoghurt factories, a fact that may expose these products to serious contamination.

Changes in chemical, physical and microbiological contents of yoghurt determine the storage and shelf life of the product (Sofu and Ekinci, 2007). Some strains of coliforms are the main reason for taints in milk and milk products rendering them unmarketable (Yabaya and Idris, 2012).

The high count of Escherichia coli and coliforms indicates bad methods of handling and processing as well as poor sanitation. E. coli is associated with food poisoning, diarrhea, and gastroenteritis (Forsythe, 2000).

When Enterococci presents many living cells, it causes illness besides their spoilage effect. It has been reported that some strains of Enterococci were salt tolerant and heat resistant (ICMSF,1986).

Staphylococcus aureus has been reported as food poisoning as it can show illness symptoms within four to six hours after eating contaminated food (De-Buyser et al., 2001). these symptoms include nausea, vomiting, headache, and diarrhea without fever (David et al., 1996).

In yoghurt, yeasts and molds are the major contaminants (Nwagu and Amadi, 2010). The presence of yeasts and molds even in a few numbers results in undesirable changes which render food and make it unmarketable (Abdel-Hameed, 2011).

To advocate the hygienic condition under which the Egyptian fermented milk have been produced this investigation was planned to determine the acidity%, total coliform, fecal coliform, Enterococci and S. Aureus count, also detection of E. coli and anaerobes bacteria.

Materials and Methods

Collection of samples

One hundred and forty random samples of yoghurt and Laban raib, seventy of each, were collected from different localities and villages in El-Kharga city, New Valley governorate, Egypt.

Samples of yoghurt in their plastic containers were collected from small dairy shops and villages. While Laban raib samples were collected in sterile sampling jars, from different villages located in the suburbs of New Valley governorate.

The collected samples were transferred directly to the laboratory with a minimum delay where they were prepared for the examination.

Preparation of samples

Each sample was perfectly mixed before being divided into two subsamples. the first subsample was used for the determination of acidity %, while the second part was examined microbiologically.

i. Determination of acidity% (A.O.A.C, 2005).

ii. Microbiological examination

Preparation of serial dilution (A.P.H.A.,2001).

Determination of total coliforms and fecal coliforms (A.P.H.A., 2001):

i. E.coli isolation (Collee et al., 1996)

ii. Identification of E.coli

a. Microscopic examination (Oyeleke et al., 2008):

b. Biochemical reactions (A.P.H.A., 2004): As indole production, methyl red test, Voges-Proskauer test, citrate utilization test and Triple sugar Iron test (TSI):

c. Enumeration of Enterococci (Mossel et al., 1978):

d. Enumeration, isolation and identification of S.aureus (A.P.H.A., 2004):

I. Identification of S.aureus

II. Microscopical examination (A.P.H.A., 2004)

III. Biochemical reactions

a. Catalase test (Quinn et al., 1994)

b. Mannitol fermentation test(Quinn et al., 1994)

c. Coagulase test (A.P.H.A., 2004)

d. Anaerobes detection (Cruickshank et al., 1970)

e. Determination of total yeast and mold count (ISO21527-1:2008)

Results

Table 1: Statistical analytical results of titratable acidity in the examined fermented milk products samples (CFU/ml).

Type of samples	Minimum	Maximum	Average
Yoghurt	0.65	1.53	1.13
Laban raib	0.82	1.62	1.13

Table 2: Statistical analytical results of total coliforms count in the examined fermented milk products samples (CFU/ml)

Type of samples	No. of examined samples	Positive samples		Minimum	Maximum	Average
		No.	%			
Yoghurt	70	47	67.1	1.5x103	9.3x105	6.8x104
Laban raib	70	59	84.3	9.3x103	2.4x106	1.6x105

Table 3: Frequency distribution in the positive examined fermented milk products samples based on their total coliforms count.

Intervals	Yoghurt		Laban raib	
	No./70	%	No./70	%
$<10^4$	31	65.96	5	8.47
$<10^5$	12	25.53	22	37.29
$<10^6$	4	8.51	24	40.68
$<10^7$	0	0	8	13.56
Total	47	100	59	100

Table 4: Statistical analytical results of fecal coliforms count in the examined fermented milk products samples (CFU/ml).

Type of samples	No. of examined samples	Positive samples		Minimum	Maximum	Average
		No.	%			
Yoghurt	70	40	57.14	$2.3x10^2$	$4.2x10^5$	$5.5x10^4$
Laban raib	70	53	75.7	$5x10^3$	$1.2x10^6$	$1.1x10^5$

Table 5: Frequency distribution in the positive examined fermented milk products samples based on their fecal coliforms count.

Intervals	Yoghurt		Laban raib	
	No./70	%	No./70	%
$<10^3$	2	5	0	0
$<10^4$	22	55	3	5.66
$<10^5$	13	32.5	25	47.17
$<10^6$	3	7.5	19	35.85
$<10^7$	0	0	6	11.32
Total	40	100	53	100

Table 6: Occurrence of isolated E. coli in the examined fermented milk products samples.

Samples	No. of examined samples	Positive samples	
		No.	%
Yoghurt	70	8	11.4
Laban raib	70	6	8.5

Table 7: Statistical analytical results of total Enterococci count in the examined fermented milk products samples (CFU/ml).

Type of samples	No. of examined samples	Positive samples		Minimum	Maximum	Average
		No.	%			
Yoghurt	70	0	0	< 10	< 10	< 10
Laban raib	70	0	0	< 10	< 10	< 10

Table 8: Statistical analytical results of S. aureus count in the examined fermented milk products samples (CFU/ml).

Type of samples	No. of examined samples	Positive samples		Minimum	Maximum	Average
		No.	%			
Yoghurt	70	16	22.8	$6x10^2$	$4x10^4$	$1.42x10^4$
Laban raib	70	5	7.1	$2x10^2$	$3x10^3$	$2.4x10^3$

Table 9: Frequency distribution in the positive examined fermented milk products samples based on their S. aureus count.

Intervals	Yoghurt		Laban raib	
	No./70	%	No./70	%
$>10^3$	11	68.75	5	100
$>10^4$	5	31.25	0	0
Total	16	100	5	100

Table 10: occurrence of anaerobes in the examined fermented milk products samples.

Samples	No. of examined samples	Positive samples	
		No.	%
Yoghurt	70	2	2.8
Laban raib	70	3	4.3

Table 11: Statistical analytical results of yeast count in the examined fermented milk products samples (CFU/ml).

Type of samples	No. of examined samples	Positive samples		Minimum	Maximum	Average
		No.	%			
Yoghurt	70	61	87.14	$7x10^2$	$9x10^8$	$7.2x10^7$
Laban raib	70	58	82.86	$1.2x10^3$	$9x10^7$	$2.1x10^7$

Table 12: Frequency distribution in the positive examined fermented milk products samples based on their yeast count.

Intervals	Yoghurt		Laban raib	
	No./70	%	No./70	%
$<10^3$	6	9.8	0	0
$<10^4$	3	4.9	4	6.9
$<10^5$	7	11.5	6	10.4
$<10^6$	8	13.1	13	22.4
$<10^7$	10	16.4	22	37.9
$<10^8$	20	32.8	13	22.4
$<10^9$	7	11.5	0	0
Total	61	100	58	100

Table 13: Statistical analytical results of mold count in the examined fermented milk products samples (CFU/ml).

Type of samples	No. of examined samples	Positive samples		Minimum	Maximum	Average
		No.	%			
Yoghurt	70	18	25.7	$5x10^2$	$3x10^7$	$4.9x10^6$
Laban raib	70	35	50	$5x10^4$	$8.6x10^5$	$2.8x10^5$

Table 14: Frequency distribution in the positive examined fermented milk products samples based on their mold count.

Intervals	Yoghurt		Laban raib	
	No./70	%	No./70	%
$<10^3$	3	16.67	0	0
$<10^4$	2	11.11	0	0
$<10^5$	7	38.89	17	48.6
$<10^6$	3	16.67	11	31.4
$<10^7$	3	16.67	7	20
Total	18	100	35	100

Discussion

In titratable acidity may be attributed to the starter, temperature, and incubation time (Walstra et al., 1999).

Total coliform coun It is evident from results in Table (2) and Table (3) that total coliform could be detected in 47(67.1%) yoghurt samples with an average value of $6.8 x 10^4$, the highest frequency distribution lies within the range of 10^3-10^4 cfu/ml. The lower results showed by El-Kasas (2004), Moustafa (2004), Al-Tahii (2005), Wafy (2006), Zeinhom (2007), El-Diasty and El-Kaseh (2008), Reyhan and Ufuk (2008), Moustafa (2011), Abou El-Makarem (2013), Armanios (2013) and Shawer (2013). But Olasupo et al. (2002), Hassan (2003), Sadik (2009) and Shahin (2015) showed higher results.

In Laban raib, the results in Table (2) showed that total coliform was detected in 59(84.3%) of the examined samples with an average value of 1.6×10^5, Sayed (2012) and Ahmed et al. (2017), showed lower results.

The presence of coliforms in milk and its products ensures unsanitary production, storage, or post-pasteurization contamination (Robinson, 2002).

It's apparent from Table (4) and Table (5) that fecal coliform was detected in 40(57.14%) yoghurt samples with an average value of 5.5×10^4. The highest frequency distribution lies within the range of 10^3-10^4 cfu/ml. Lower results for fecal coliforms in yoghurt samples were detected by Wafy (2006), Zeinhom (2007), Abdel-Rahman (2010), Sayed (2012) and Armanios (2013). On the other hand, Hassan (2003) presented the highest.

In Laban raib samples, statistical analytic results demonstrated that fecal coliform was found in 75.7 %of the examined samples with high-frequency distribution of 47.17 between 10^4-10^5 cfu/ml with an average value of 1.1×105. Ahmed et al (2017) couldn't detect coliforms.

The presence of fecal coliform indicates enteric pathogen pollution, unhygienic measures and improper sanitation which considers public health hazards (Emam, 2008).

The results in Table (6) revealed that E. coli was found in 8(11.4%) of yoghurt samples and 6(8.5%) of Laban raib samples. The summarized results of E.coli in yoghurt samples by El-Bessary (2001), Aid (2002), Hassan (2003), El-Kasas (2004), Zeinhom (2007), Abd El-Aal (2008), Shalaby and Galab (2008), Abdel-Rahman (2010), El-Kholy et al (2014), Sadek et al (2014), Ibrahim et al (2015), Sunday et al (2016), Samuel and Ifeany (2016), Emam et al (2016), Fahim et al (2016)and Lotfy et al (2017) showed higher results. While Reyhan and Ufuk (2008), Moustafa (2011), and Sayed (2012) showed lower results as they couldn't find E. coli in the examined yoghurt samples.

Ahmed et al (2018) showed nearly similar results, while Abd-Allah et al (2020) showed a higher result, in Laban raib samples.

E. coli exists normally in a man's gastrointestinal tract. Therefore, under insufficient sanitation and cleaning, E. coli in dairy products is an indicator of fecal contamination. Also, E. coli has a distinctive role in cases of appendicitis, pyelitis, pyelonephritis, peritonitis, and septicemia. (Fawzi, 1999).

Data recorded in Table (7) proved that Enterococci couldn't be detected in all the examined yoghurt and Laban raib samples. El-Bessary (2001), Hassan (2003), Wafy (2006), Zeinhom (2007), Abd El-Aal (2008), Sadik (2009), Abdel-Rahman (2010), Abou El-Makarem (2013), Armanios (2013) and Shahin (2015) recorded higher results than the obtained results.

Enterococci are widely found in nature. Milk and milk products can be contaminated with Enterococci through the equipment, water supply and unsanitary and unhygienic conditions of production and handling. The evidence concerning their involvement is only circumstantial. (Garg and Mital, 1991)

Table (8) and Table (9) confirmed that S.aureus were found in 16(22.8%) of examined yoghurt samples with an average value of $1.4^2\times10^4$ cfu/ml. The highest frequency distribution lies within the range of 10^2-10^3. The obtained results of S.aureus in yoghurt samples by Olasupo et al. (2002), Hassan (2003), Moustafa (2004), Al-Tahiri (2005), Wafy (2006), Abd El-Aal (2008), Abou El-Makarem (2013), Shawer (2013), and Osman (2015) were lower than our results. Armanios (2013) showed a nearly similar result, while Sadik (2009) and Shahin (2015) showed higher results.

Also, it's confirmed from Table (8) and Table (9) that S.aureus were found in 5(7.1%) of examined Laban raib samples with an average value of 2.4×10^3 cfu/ml, the highest frequency distribution was found within the range of 10^2-10^3. Abd-Allah et al (2020) presented higher results.

S.aurues bacteria cause lots of food poisoning outbreaks. S.aureus produces thermostable enterotoxins which are resistant to digestive enzymes. Le Loir et al., (2003). Ingesting thermally stable S.aureus enterotoxins at a dose of 0.1 to 1.0 mg/kg of body weight makes the patient symptomatic within two to four hours Stewart et al. (2005).

Table (10) cleared that the anaerobes bacteria could be found in 2(2.8%) of the examined yoghurt samples and 3(4.3%) of the examined Laban raib samples. Sayed (2012) presented a lower result as he couldn't detect anaerobes in all the examined yoghurt and Laban raib samples.

Contamination of dairy food by anaerobes can be so dangerous, especially for children. Anaerobes could colonize and produce toxins in the intestine, their spores can also germinate at cooling temperature and produce toxins after their rapid multiplying causing food poisoning outbreaks (Sayed and Abdel-Haleem, 2005).

Results are shown in Table (11) and Table (12) clarified that yeasts were found in 61(87.14%) of the examined yoghurt samples with an average value of 7.2×10^7 and high-frequency distribution lies between 107-108. Lower results of yeast count were detected by El-kasas (2004), Moustafa (2004), Al-Tahiri (2005), Wafy (2006), El-Diasty and El-Kaseh (2008), Tanweer et al. (2008), Sadik (2009), El-Asuoty (2011), Moustafa (2011), Abou El-Makarem (2013), Shawer (2013) and Osman (2015).

In Laban raib samples, yeasts were found in 58(82.86%) of the examined samples with an average value of 2.1×10^7 and high-frequency distribution lies between 10^6-10^7. Ahmed et al. (2017) and Abd-Allah et al (2020) presented lower results.

Yoghurt is a high acidic product by nature so, it provides an environment favoring the growth of yeasts and molds as spoilage microorganisms, they consume the acid resulting in an acid decrease which provides a suitable environment for putrefactive bacteria. Also, yeast and mold presence are an indication for poor sanitation (El-Malt et al., 2013)

Inspection of the results present in Table (13) and Table (14) revealed that molds could be detected in 18(25.7%) yoghurt samples with an average value of 4.9×10^6 and the highest frequency distribution lies between 10^4-10^5.

Lower results of mold count were detected by El-kasas (2004), Moustafa (2004), Al-Tahiri (2005), Wafy (2006), El-Diasty and El-Kaseh (2008), Tanweer et al. (2008), Sadik (2009), El-Asuoty (2011), Moustafa (2011), Abou El-Makarem (2013), Shawer (2013) and Osman (2015).

In Laban raib samples, mold was found in 35(50%) of the examined samples with an average value of 2.8×10^5 and high-frequency distribution lies between 10^4-10^5. Ahmed et al. (2017) and Abd-Allah et al (2020) presented lower results.

Conclusion

Contamination of dairy products by yeasts and molds comes from the air, containers used in the processing and improper storage, resulting in several defects in dairy products.

Conflict of interest

The authors haven't conflict of interest to declare.

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