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## Research Article

# Microbiological Quality of Some Dairy Products with Special Reference to the Incidence of Some Biological Hazards

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## Abstract

**Background and Objective:** Ingestion of contaminated dairy products causes foodborne diseases (FBD) which pose a significant but often under recognized threat to public health, worldwide, consequently this study was aimed to throw light on the microbiological quality of some dairy products sold in Egyptian markets. **Materials and Methods:** Ninety random samples of ice cream, Ras cheese and fresh white soft cheese (30 of each) collected from dairy shops and stores in Port-Said and Giza governorates, Egypt and examined microbiologically. **Results:** The mean titratable acidity of ice cream, Ras cheese and fresh white soft cheese samples were (0.13, 0.43 and 0.14%), microbiological examination proved contamination of the examined samples with high numbers of aerobic mesophilic microorganisms, coliform, staphylococci, yeast, mold and anaerobic spore formers. *E. coli* was isolated from Ras and white fresh soft cheese with 2.9 and 4.4%, respectively. *Listeria monocytogenes* couldn't be isolated from all of the examined samples, while *Yersinia enterocolitica* was isolated from 23.3% of ice cream samples. **Conclusion:** Inadequate hygienic conditions of production and storage mandate the promotion of hygienic regulations and guarantee of safety from production until receiving to consumer.

**Key words:** Ice cream, cheese, listeria monocytogenes, *E. coli*, *Yersinia enterocolitica*, dairy products, foodborne diseases

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**Competing Interest:** The authors have declared that no competing interest exists.

**Data Availability:** All relevant data are within the paper and its supporting information files.

## INTRODUCTION

Foodborne diseases are considered to be among the foremost economic and public health concerns particularly in susceptible people, such as infants, pregnant women, children and elderly throughout the world<sup>1</sup>.

Dairy products among them ice cream which is a frozen dairy product is made by freezing a pasteurized mixture of milk, cream milk solids other than sugar, emulsifiers, stabilizers, flavoring and coloring agents and cheese that is made by coagulating the milk for 40-60 min by rennet are highly at risk to microbial contamination<sup>2</sup>.

Annually, hundreds millions of people all over the world are get poisoned with these products because of contamination either of raw materials, or equipment or final products in the production process, transportation and distribution and non-observance of good manufacturing practices (GMP) by workers<sup>3</sup>.

Pathogens causing foodborne outbreaks that associated with the consumption of milk and dairy products include *E. coli* O157:H7, *Staphylococcus aureus*, *Cl. Botulinum*, *Bacillus cereus*, *Yersinia enterocolitica* and *Listeria monocytogenes*, they represent a major public health hazard, especially for persons who still drink raw milk and raw milk products<sup>4,5</sup>.

A useful indicator for assessing the overall quality and safety of dairy products and monitoring the sanitary conditions applied during the production, collection and handling is the Standard Plate Count<sup>6,7</sup>.

Microbiological quality of dairy products and post heat treatment contamination can be also determined using coliform count, among these group *E. coli* which is considered the most common pathogen causing milk and dairy products borne outbreaks<sup>8</sup>.

Spoilage by fungi (yeast and mold) was regarded as a quality concern rather than a food safety issue since 50 years ago, later many common mold species were discovered to be dangerously toxicogenic due to mycotoxin production which represent a threat to public health<sup>7</sup>.

Presence of anaerobes in dairy products may be indicative of manure and soil contamination because of most of the anaerobic organisms are saprophytes and normally grow in soil and water and the others are commensals of the animal and human intestine<sup>9,10</sup>.

Microbial contamination and growth rate in many dairy foods can be reduced using one or more of the subsequent treatments: pH reduction by lactose fermentation into lactic acid, adding acids or other approved preservatives, restriction the growth of undesirable microorganisms using desirable

microflora, sugar or salt addition to reduce the water activity ( $a_w$ ), packaging to limit available oxygen and freezing<sup>11</sup>.

Because of all mentioned before, this survey study was made to throw light to what extent some of the dairy products (ice cream, Ras cheese and fresh white soft cheese) are in acceptance with the Egyptian standard and hygienically safe for the Egyptian consumers and what are the most common contaminating microorganisms especially those cause a biological hazard to search for a suitable control for these microorganisms in the future study.

## MATERIAL AND METHODS

**Samples collection:** Ninety random samples of ice cream (Vanilla type), Hard cheese (Ras cheese) and fresh white soft cheese (Tallaga cheese) (30 of each) were collected from dairy shops and stores in Port-Said and Giza governorates, Egypt in the period from September, 2018 to June, 2019s. Samples were transmitted to the laboratory in an insulating ice-box as soon as possible for examination.

### Chemical examination:

- Determination of titratable acidity percentage of ice cream was determined according to AOAC<sup>12</sup> and cheese samples according to APHA<sup>13</sup>

### Microbiological examination:

- Preparation of decimal dilutions of the examined samples according to APHA<sup>13</sup>
- Total aerobic mesophilic count was applied according to ISO<sup>14</sup>
- Coliform content (MPN/g) was assessed according to APHA13 with identification of the isolated Coliform according to DeVos *et al.*<sup>15</sup>. Molecular identification of isolated *E. coli* by polymerase chain reaction (PCR) for *E. coli* 16S rRNA gene by ECO-1 and ECO-2 primers (Table 1) according to the standard procedure designated by Schippa *et al.*<sup>16</sup>
- Total Staphylococci count with Identification of the suspected *S. aureus* was determined according to APHA<sup>13</sup>
- Total Yeast and Mold count was assessed according to ISO<sup>17</sup>
- Anaerobic spore formers count was determined according to APHA<sup>13</sup>
- Incidence of *Yersinia enterocolitica* and *Listeria monocytogenes* were assessed according to APHA<sup>13</sup> and BAM online<sup>18</sup>

**Statistical analysis:** Results were calculated in the form of mean+standard deviation using the program Statistical Package for Social Science (SPSS), version<sup>17</sup>.

## RESULTS

**Titrateable acidity:** On studying titrateable acidity (%) of the examined samples, data presented in Table 2 showed that the maximum values were 0.27, 0.70 and 0.58 for the examined ice cream, Ras cheese and white fresh soft cheese samples with mean values of  $0.13 \pm 0.01$ ,  $0.43 \pm 0.02$  and  $0.14 \pm 0.02$ , respectively.

**Total aerobic mesophilic count (CFU g<sup>-1</sup>):** Results revealed that all examined samples of ice cream, 96.7% of Ras cheese and 80% of white fresh soft cheese samples were contaminated with microorganisms (Fig. 1), with mean count of  $15 \times 10^5$ ,  $3.2 \times 10^9$  and  $5.9 \times 10^6$  CFU g<sup>-1</sup>, respectively (Table 2).

**Coliform content (MPN g<sup>-1</sup>):** Regarding the results presented in Fig. 1, it is evident that coliforms were detected in 96.7%, 66.7 and 76.7% of the examined ice cream, Ras cheese and

white fresh soft cheese samples, respectively, with a mean value of  $1.9 \times 10^4$ ,  $7.3 \times 10^4$  and  $3.2 \times 10^4$  MPN g<sup>-1</sup>, respectively (Table 2).

**Isolated coliforms:** The biochemical identification of coliform organisms revealed that *Citrobacter diversus* was the most frequent one (53.6%) in ice cream followed by *Citrobacter freundii* (17.9%), *Enterobacter intermedius* (12.5%), *Serratia fonticola* (8.9%) and *Klebsiella oxytoca* (3.5%). On Ras cheese, results revealed that *Citrobacter diversus* (34.3%), *Citrobacter freundii* (22.9%) and *Serratia fonticola* (17%) were the most frequent one followed by *Enterobacter intermedius* (14.3%), *E. coli* could be detected with (2.9%). *Citrobacter diversus* (39.1) and *Citrobacter freundii* (21.7%) had the highest incidence, then *Enterobacter intermedius* (17.4%) and *Klebsiella oxytoca* (17.4%) in the examined white fresh soft cheese, whereas *E. coli* could be detected with 4.4% in these cheese samples (Fig. 2). PCR confirmed the presence of pathogenic *E. coli* as it showed positive amplification for *E. coli* 16s rRNA gene at 585 bp (Fig. 3).

Table 1: Primers used in this study

Primers name	Sequence	bp	Target gene
ECO-1	ACCTCGGTTTAGTTCACAGA	585	16s rRNA
ECO-2	ACACGCTGACGCTGACCA		

Table 2: Statistical analytical results of the determined parameters in the examined samples

Total number of samples	Minimum	Maximum	Mean $\pm$ SEM
<b>Chemical analysis</b>			
<b>Titrateable acidity (%)</b>			
Ice cream	0.03	0.27	$0.13 \pm 0.01$
Ras cheese	0.14	0.70	$0.43 \pm 0.02$
Fresh soft cheese	0.04	0.58	$0.14 \pm 0.02$
<b>Microbiological examination</b>			
<b>Total aerobic mesophilic count (CFU g<sup>-1</sup>)</b>			
Ice cream	$4 \times 10^3$	$1.2 \times 10^7$	$15 \times 10^5 \pm 6.2 \times 10^5$
Ras cheese	$1 \times 10^5$	$5.7 \times 10^{10}$	$3.2 \times 10^9 \pm 21 \times 10^9$
Fresh soft cheese	$3 \times 10^4$	$3 \times 10^7$	$5.9 \times 10^6 \pm 1.5 \times 10^6$
<b>Coliform count (MPN g<sup>-1</sup>)</b>			
Ice cream	40	$2.4 \times 10^5$	$1.9 \times 10^4 \pm 0.9 \times 10^4$
Ras cheese	$4 \times 10^3$	$< 1.1 \times 10^6$	$7.3 \times 10^4 \pm 3.9 \times 10^4$
Fresh soft cheese	$4 \times 10^2$	$4.6 \times 10^6$	$3.2 \times 10^4 \pm 1.6 \times 10^4$
<b>Total Staphylococci count (CFU g<sup>-1</sup>)</b>			
Ice cream	$10^3$	$1.6 \times 10^8$	$5.8 \times 10^6 \pm 5.4 \times 10^6$
Ras cheese	$10^3$	$1.1 \times 10^8$	$5.9 \times 10^6 \pm 3.6 \times 10^6$
Fresh soft cheese	$10^4$	$1.9 \times 10^7$	$3.4 \times 10^6 \pm 0.99 \times 10^6$
<b>Total yeast count (CFU g<sup>-1</sup>)</b>			
Ice cream	$2 \times 10^2$	$8.92 \times 10^5$	$17 \times 10^4 \pm 4.5 \times 10^4$
Ras cheese	$4 \times 10^5$	$1.53 \times 10^{10}$	$7.7 \times 10^8 \pm 5.09 \times 10^8$
Fresh soft cheese	$2 \times 10^3$	$1.85 \times 10^7$	$14 \times 10^5 \pm 8.1 \times 10^5$
<b>Total mold count (CFU g<sup>-1</sup>)</b>			
Ice cream	10	$10^5$	$4.5 \times 10^3 \pm 3.3 \times 10^3$
Ras cheese	$10^3$	$2 \times 10^5$	$7.1 \times 10^4 \pm 6.7 \times 10^4$
Fresh soft cheese	$10^4$	$10^4$	$3.3 \times 10^2 \pm 3.3 \times 10^2$
<b>Anaerobic spore formers count (CFU g<sup>-1</sup>)</b>			
Ras cheese	$10^2$	$7.5 \times 10^4$	$4.2 \times 10^3 \pm 2.6 \times 10^3$

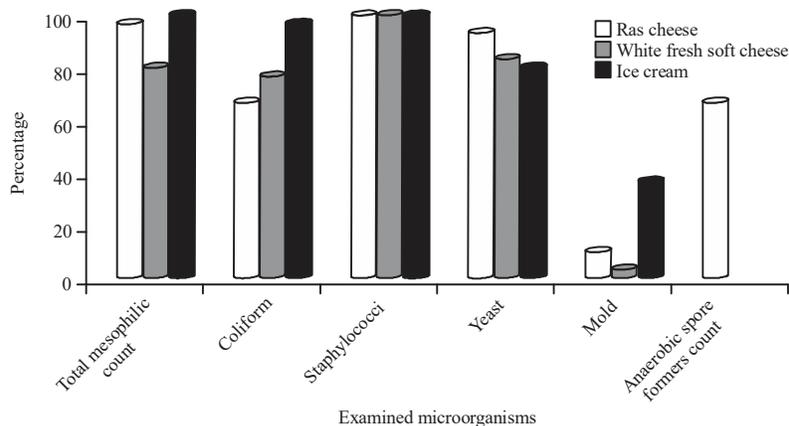


Fig. 1: Incidence of the examined microbiological parameters in the examined samples

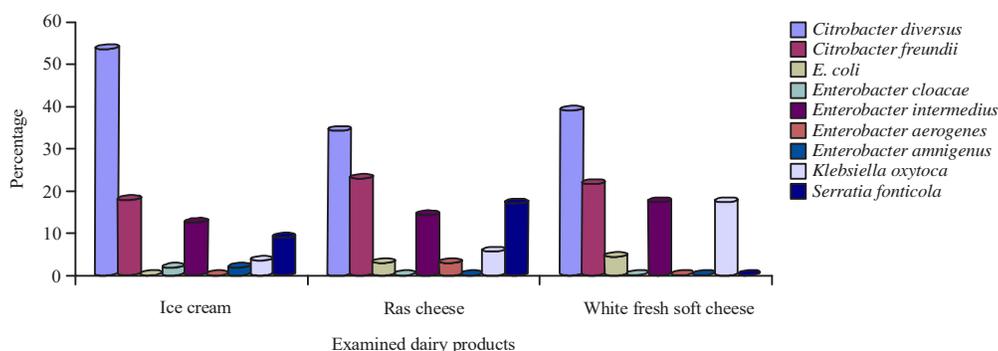


Fig. 2: Incidence of the isolated coliform from the examined samples

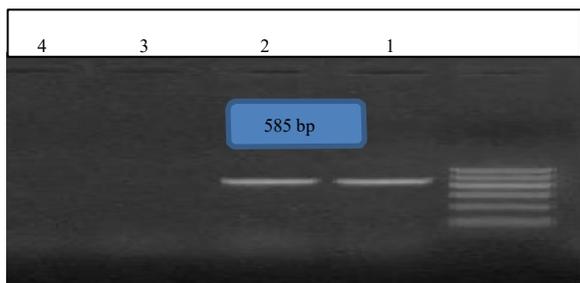


Fig. 3: Agarose gel electrophoresis showing positive amplification for *E. coli* 16s rRNA gene at 585 bp  
Lane 1 and 2: Positive isolates

**Total staphylococcal count:** Results indicated that staphylococci was presented in all of the examined samples of ice cream, Ras cheese and white fresh soft cheese (Fig. 1), with mean count of  $5.8 \times 10^6$ ,  $5.9 \times 10^6$  and  $3.4 \times 10^6$  CFU  $g^{-1}$ , respectively (Table 2).

**Confirmed *Staphylococcus aureus*.** It was obvious that the incidence of *S. aureus* in the examined samples of ice cream, Ras cheese and white fresh soft cheese depending on the

results of coagulase test were 0.0, 8.3 and 19.0%, while depending on the results of TNase test were 35.3, 66.7 and 76.2%, respectively. whereas depending on both tests were 8.3 and 9.5% in Ras cheese and white fresh soft cheese samples, respectively (Fig. 4).

**Total yeast count:** Data depicted in (Fig. 1) revealed that contaminated yeast was detected in 24 (80%), 28(93.3%) and 25(83.3%) of ice cream, Ras cheese and white fresh soft cheese, respectively, with mean values of  $17 \times 10^4$ ,  $7.7 \times 10^8$  and  $14 \times 10^5$  CFU  $g^{-1}$ , respectively (Table 2).

**Total mold count:** Regarding the data recorded in Fig. 1, Mold was presented in 36.7, 10 and 3.3% of the examined ice cream, Ras cheese and white fresh soft cheese with mean values of  $4.5 \times 10^3$ ,  $7.1 \times 10^4$  and  $3.3 \times 10^2$  CFU  $g^{-1}$ , respectively (Table 2).

**Anaerobic spore formers count in Ras cheese samples:** Data depicted in Fig.1 showed the incidence of anaerobic spore-formers in the examined Ras cheese samples. Anaerobes were detected in 66.7% of the examined samples with a mean value of  $4.2 \times 10^3 \pm 2.6 \times 10^3$  CFU  $g^{-1}$  (Table 2).

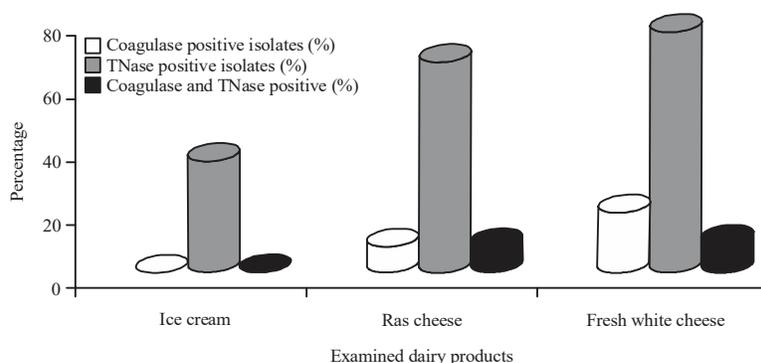


Fig. 4: *Staphylococcus aureus* prevalence depending on the outcomes of coagulase and TNase test

Table 3: Incidence of some pathogenic microorganisms

Type of products	<i>Listeria monocytogenes</i>	<i>Yersinia enterocolitica</i>			
		Number of positive samples		Total number of isolates (35 isolates)	
		Number	Percentage	Number	Percentage
Ice cream	Not detected	7	23.3	12	34.3
White fresh soft cheese	Not detected	-	-	-	-

**Incidence of some pathogenic microorganisms:** Tabulated data in Table 3 revealed that 35 yersinia strains were isolated from 23.3 % of the examined ice cream samples from which 12 strains (34.3%) were confirmed as *Y. enterocolitica* through the biochemical tests, while *Listeria monocytogenes* could not be detected in all of the examined samples of ice cream and white fresh soft cheese (Tallaga cheese).

## DISCUSSION

Dairy products freshness and bacterial activity in milk, bacterial contamination and the temperature at which the dairy products are kept are the main factors affecting the acid formation. Lowering milk pH from 6.8 to <4.6 protect the fermented dairy products such as cheese against the risk of contamination by pathogens and render them hygienically safe<sup>19</sup>.

From the previously mentioned results and according to the Egyptian Standards (1185-1/2005)<sup>20</sup>, all of the examined ice cream samples agree with the standard. The obtained results of ice cream were nearly similar to those obtained by Barman *et al.*<sup>21</sup> who found that the titratable acidity of the examined samples ranged from 0.235-0.275% lactic acid.

Ripened cheese is normally considered to be safe against food borne diseases because of its low pH, beside some of the lactic acid bacterial starter cultures used in fermentation produce antimicrobial compounds such as bacteriocins, hydrogen peroxide, formic acid, acetate and diacetyl<sup>22</sup>. Data

reported for Ras cheese was lower than those obtained by El-Leboudy *et al.*<sup>23</sup>, who reported that the mean titratable acidity (%) was  $1.97 \pm 0.0578$  in the examined samples and Abdelmagid and Hamid<sup>24</sup>. While nearly similar results of white fresh soft cheese were recorded by Sengul and Ertugay<sup>25</sup>.

Higher results were obtained by Kavaz *et al.*<sup>26</sup>, who examined Çökelek (cottage cheese like) cheese samples and found that the Titratable acidity was 0.28% and Haddad and Yamani<sup>27</sup>.

Standard plate count is one of the most common techniques used for assessing the overall quality and safety all over the world during the production, collection and handling of milk and dairy products<sup>6,7</sup>. High count of aerobic mesophilic bacteria in the examined samples considered a bad signal indicating low hygiene and poor quality products. Higher initial microbial load of raw milk used, resistance of spore forming organisms to the heat treatment, post heat treatment contamination, bad storage and/or bad handling might be the causes of these high counts<sup>28</sup>. According to the mentioned results of ice cream and the Egyptian Standards (1185-1/2005)<sup>20</sup>, which recommends that Total aerobic mesophilic count shouldn't exceed  $5 \times 10^4$  CFU g<sup>-1</sup>, 60% of the examined samples disagree with the Egyptian standard. The obtained results of Ice cream were lower than that recorded by Jadhav and Raut<sup>29</sup> and Barman *et al.*<sup>21</sup>, while higher than those reported by Saber<sup>30</sup> and Abou El Khair *et al.*<sup>31</sup>. On the other hand, data reported for Ras cheese was nearly similar to those obtained by

Abd El-Raheem<sup>32</sup>, while lower than those reported by Karima<sup>33</sup>, who found that the mean total colony count was  $2.39 \times 10^{10}$  CFU g<sup>-1</sup> in the examined Ras cheese samples. Results obtained for white fresh soft cheese were lower than Ghada *et al.*<sup>34</sup> and Moraes *et al.*<sup>35</sup> and higher than those reported by Senbetu<sup>36</sup> and Heikal *et al.*<sup>37</sup>.

Coliform count is a traditional indicator of possible faecal contamination, microbial quality and wholesomeness and reflect the hygienic standards adopted in the food operation and as coliform organisms can be easily killed by heat, these bacteria can also be used as an indicator of heat treatment failure as well as post heat treatment contamination<sup>38,8</sup>. High incidence of coliforms in the examined samples indicated the neglected sanitary measures, faulty heat processing or post pasteurization contamination by handlers<sup>39,40</sup>. Moreover, these high numbers resulted in 'Early blowing' defect which characterized by presence large gas holes and a spongy texture in cheese causing economic losses. It is clear from the obtained results that 96.66, 66.66 and 76.66% of the examined ice cream, Ras cheese and fresh white soft cheese (Tallaga) contained high numbers of coliform and disagree with the Egyptian Standards (1185/2005-1007/2005-1008/2005)<sup>20,41,42</sup> which recommend that coliform count should be less than 10 cells/g in the product. High incidence of coliform rendered them of inferior quality and became unmarketable during storage or even unfit for human consumption causing economic losses. The public health importance of coliform bacteria is its implication in gastrointestinal illness as gastroenteritis, epidemic diarrhea in children and cases of food poisoning<sup>43</sup>.

Results reported for ice cream were nearly similar to those given by Al-Gendi<sup>44</sup> while higher figures were recorded by Aya<sup>45</sup>. Lower results were reported by Abou El Khair<sup>31</sup>, Jadhav and Raut<sup>29</sup> and Barman *et al.*<sup>21</sup>. Higher results of Ras cheese were reported by Aya<sup>45</sup> and El-Leboudy *et al.*<sup>23</sup>, while lower results were obtained by Abdelmagid and Hamid<sup>24</sup>. While higher results of Tallaga cheese were reported by Moraes *et al.*<sup>35</sup> and Aya<sup>45</sup>. Lower results were obtained by Trmčić *et al.*<sup>46</sup> and Abdalla and Omer<sup>47</sup>.

It is clear that ice cream samples were in accordance with the Egyptian Standards, as they were free from *E. coli*. While Ras and white fresh soft cheese samples disagreed with this standard and contained *E. coli* with percentages of 2.9 and 4.4 (Fig. 3), its presence in the examined samples is indicator of fecal contamination and suggests that other food borne pathogens of fecal origin may also be present<sup>48</sup>. Most strains of *E. coli* are harmless commensals, some caused human gastrointestinal disease with mild to severe symptoms that may progress to long-term sequel or fatal outcomes in

high-risk individuals. *E. coli* also was found to be responsible for cases of cystitis, pyelitis, pyelonephritis as well as appendicitis and peritonitis<sup>49,50</sup>.

All examined samples contained high numbers of *Staphylococci*, which reflected the poor sanitary conditions applied during production, processing and distribution<sup>51,52</sup>. And this could be occurred due to the dominance of the genus on parts of the human body such as, nose, hands and skin. Higher results of ice cream were recorded by Aya<sup>45</sup>, while lower results were reported by Garbaj<sup>53</sup> and Barman *et al.*<sup>21</sup>. Lower data of Ras cheese was reported by Aya<sup>45</sup>. Whereas lower results of white fresh soft cheese were obtained by Aya<sup>45</sup> and Senbetu<sup>36</sup>.

*Staphylococci* can be divided into 2 groups according to the production of coagulase enzyme, which is capable of coagulating blood plasma. In routine laboratory practice, production of coagulase is frequently used as a sole criterion to distinguish *S. aureus* from other *Staphylococci* that do not synthesize this enzyme which referred to as Coagulase negative Staphylococci (CNS), Coagulase positive *Staphylococcus aureus* (CPS) is considered the most important species of *Staphylococci* due to its pathogenicity and enterotoxin production causing food intoxication<sup>54</sup>, all enterotoxigenic strains are coagulase producers but not all of them are TNase producers. Garbaj<sup>53</sup> reported that about 50.0% of the enterotoxigenic *Staphylococcus aureus* isolates were positive for both coagulase and TNase production. In general staphylococci that produce enterotoxins are coagulase and/or thermonuclease positive<sup>55</sup>. Ingestion of food contaminated with staphylococcal enterotoxins cause *S. aureus* food poisoning intoxication, which is emetic, pyrogenic and mitogenic and suppresses the immunoglobulin production what rendered it one of the most common types of food borne diseases worldwide. *S. aureus* is a common cause of boils, abscesses and more serious infections including osteomyelitis, endocarditis, enterocolitis, toxic shock and scalded skin syndrome<sup>56</sup>.

Results obtained for ice cream were in accordance with those recorded by Kokkinakis *et al.*<sup>57</sup>. While higher results were reported by Saber<sup>30</sup> and Jadhav and Raut<sup>29</sup>. Higher results of Ras cheese were recorded by Abdelmagid and Hamid<sup>24</sup>. The recorded results of white fresh soft cheese were nearly similar to those obtained by Araujo *et al.*<sup>58</sup>, higher outcomes were reported by Mirzaei *et al.*<sup>59</sup>, while lower results were obtained by Heikal *et al.*<sup>37</sup> and Trmčić *et al.*<sup>46</sup>.

Yeast contaminates the dairy products causing economic losses and undesirable changes such as frothy consistency and yeasty flavor. Moreover, some species of yeast constitute public hazard such as gastrointestinal disturbance,

endocarditis and occasionally fatal systemic diseases<sup>60</sup>. According to the Egyptian standards, total viable yeast count should not exceed 100 CFU g<sup>-1</sup> in hard cheese, whereas in Fresh soft cheese should not exceed 400 CFU g<sup>-1</sup>, only 6.66 and 16.66% of Ras cheese and Fresh white soft cheese samples agree with this standards. The high level of yeast contamination in the examined samples may be attributed to inadequate hygienic measures during production or the use of bad quality raw materials<sup>61</sup>.

Lower obtaining of ice cream was reported by Saber<sup>30</sup> and Abou El Khair *et al.*<sup>31</sup>. Lower findings of Ras cheese were recorded by Elbagory *et al.*<sup>62</sup> and El-Leboudy *et al.*<sup>23</sup>, whereas lower results of white fresh soft cheese were obtained by Elbagory *et al.*<sup>62</sup>, while higher results were recorded by Karima<sup>33</sup>.

Contamination of samples with mold causes serious economic losses because it is associated with visible signs of spoilage such as off flavor and discoloration that resulted in product rejection, with the probability that contaminated samples might be a source of mycotoxins which implicated in human food poisoning outbreaks<sup>63,64</sup>. From the obtained results, it was found that 90 and 96.66% of the examined Ras cheese and Fresh white soft cheese agreed with the Egyptian standards, respectively. The main causes of mold contamination are warm weather and inadequate refrigeration, beside increased species diversity and alteration in microbial flora<sup>65</sup>. The fungi in commercial dairy samples generally corresponded to poor cleaning practices and neglected hygienic measures during production, handling and/or the post process contamination<sup>66</sup>.

Lower results of ice cream were recorded by Saber<sup>30</sup> and Abou El Khair *et al.*<sup>31</sup>. Nearly similar findings of Ras cheese were reported by Barman *et al.*<sup>21</sup>. Lower results were described by Elbagory *et al.*<sup>62</sup>, while higher results were obtained by Karima<sup>33</sup> and Ahmed<sup>67</sup>. Higher obtaining of white fresh soft cheese was recorded by Karima<sup>33</sup> and Elbagory *et al.*<sup>62</sup>, while lower outcomes were reported by Senbetu<sup>36</sup> and Mourad *et al.*<sup>68</sup>.

Some species of the genus *Clostridium* cause food-borne disease, the most well-known food-borne disease is botulism which is caused by *Clostridium botulinum* and food poisoning *Cl. perfringens*. More over *Clostridium* spores are able to persist the pasteurization of milk and cheese-making including ripening process, where they germinate into vegetative cells metabolizing lactate into organic acids, mostly butyric acid and gases such as CO<sub>2</sub> and H<sub>2</sub> with consequential abnormal aroma and cracks which defined as late blowing defect (LBD), a major cause of spoilage in semi hard and hard cheeses<sup>69</sup>. High incidence of anaerobic organisms may be

attributed to neglected hygienic measures of production and handling of milk and dairy products. Most of the anaerobic organisms are saprophytes and normally grow in soil and water, some of them are commensals of the animal and human intestine and therefore, presence of such anaerobes in cheese may be indicative of manure and soil contamination<sup>10,70</sup>. Higher incidence was reported by Katherine<sup>71</sup> and Aya<sup>45</sup>, while lower occurrence was reported by Ahmed<sup>67</sup>.

Drinking raw or contaminated pasteurized milk or dairy products produced from raw milk can transmit the infection by *Yersinia enterocolitica*, which usually lead to mild self-limiting enterocolitis or terminal ileitis in humans, symptoms may include watery or bloody diarrhea and fever<sup>72</sup>. Enhancing the awareness of post-processing contamination in the dairy industry is of a major concern to reduce the incidence of entry of the pathogenic microorganisms<sup>73</sup>. On the other hand, Yaman *et al.*<sup>74</sup> could isolate *Yersinia* from 9.6% of the examined samples, While Zaher *et al.*<sup>75</sup>, Mena<sup>76</sup> and Aya<sup>45</sup> couldn't isolate *Y. enterocolitica* from any of the examined samples.

*Listeria monocytogenes* is a Gram-positive, non-spore-forming, motile, facultative anaerobic, rod-shaped bacterium. It's ubiquitous in nature due to its inherent ability to survive and grow under a wide range of adverse environmental conditions, such as refrigeration temperatures, high acidity and salinity, as well as reduced water activity<sup>77</sup>. This microorganism is of a major concern for the food industry, since it is the causal agent of listeriosis which is a severe disease with high hospitalization and case-fatality rates (approximately 91 and 30%, respectively)<sup>78</sup>. *Listeria monocytogenes* failed to be detected in all of the examined samples of ice cream and white fresh soft cheese (Tallaga cheese) and this agree with the Egyptian standard which confirmed that these products should be free from pathogenic microorganisms.

Nearly similar results of *Listeria monocytogenes* were reported by Zhou *et al.*<sup>79</sup>. Mena<sup>76</sup> and Aya<sup>45</sup>, in contrary to the obtained results of *L. monocytogenes*, many authors could isolate *L. monocytogenes* as reported by Kahraman and Kolancıyan<sup>80</sup> and Martinez-Rios and Dalgaard<sup>81</sup>.

## CONCLUSION

By examining ninety random samples of ice cream, Ras cheese and fresh white soft cheese (30 of each), some biological hazards such as *E. coli*, *S. aureus* and *Y. enterocolitica* were isolated beside presence high numbers of contaminated aerobic mesophilic

microorganisms, coliforms, *Staphylococci*, anaerobic spore formers, yeast and mold in the examined samples. Consequently, Public awareness about following the strict hygienic control measures should be applied.

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### SIGNIFICANCE STATEMENT

This study noticed the high contamination level of some dairy products with the high risk to affection with some biological hazards such as *E. coli*, *S. aureus* and *Y. enterocolitica*, consequently this study illustrated the major problem facing the dairy industry that need for a solution by the researchers for keeping the dairy products more safe for consumers.

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