REVIEW ARTICLE

Microbial contaminants and risk factors involved in the contamination of different types of cheese samples

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Dairy products, especially cheeses have a great nutritional value and a high consumption level around the world. Cheese is one of the most common foods over the world, and according to the International Dairy Foods Association report, cheese is the major manufactured dairy product, with the developing importance of the dairy industry. Considering a widespread consumption of cheeses, there is a growing concern regarding safety and microbiological quality. Bacterial and fungal growth on cheese represents both a quality and a food safety problem, and poses significant economic losses. Several microbial bacterial genera may destroy cheese; however, normally just some species of the family Enterobacteriaceae and some Gram positive bacteria dominate on specific types of cheese. *Escherichia coli*, *Salmonella* spp., *Listeria monocytogenes* and coagulase positive *Staphylococci* are the main contaminants. Cheese-contamination by bacterial species is dangerous due to toxin production, and some of the toxins have been shown to be stable under normal processing conditions. The main contamination source is the environment in the production facilities. Visible growth on cheese in the plant should be avoided in order to prevent problem of bacteria due to their growth and spreading. Identification of the contamination sources at or below species level is necessary and good hygiene practices could help reduce the microbial load to harmless level.

Key Words: *Listeria*; *Salmonella*; *Escherichia*; *Cheese*; *Dairy products*

INTRODUCTION

Milk is food for people at all ages and is a nutrient rich fluid which is used a brilliant culture medium for a wide range of microorganisms. It had many nutrients like vitamins, proteins, lipids, and carbohydrates, which nutritionally supports the growth of some pathogens [1]. Dairy products contain some important elements and vitamins such as magnesium, calcium, selenium, riboflavin, Vitamin B12, pantothenic acid for bone and muscle health. There is a rising demand for "healthy milk and dairy products [2]. Dairy products are among the first fermented foods that humans consumed and are a good source of dietary proteins and essential amino acids which have enhanced digestibility and bioavailability. Many different types of fermented dairy products under different names exist including cheese and yogurt which contain a wide range of necessary nutrients for human health due to the action of the microorganisms in the fermented dairy products [3-8].

Among fermented dairy products, cheese is a highly appreciated dairy product and is excellent growth medium for many opportunistic food spoilage and disease-causing microorganisms. In developing countries, *Staphylococcus aureus*, *Salmonella* spp., *Listeria monocytogenes* and *Escherichia coli* are the most commonly detected pathogens associated with milk and dairy products that cause the main risks of post processing contaminated cheese [9]. Outbreaks due to consumption of cheese contaminated with pathogenic bacteria and/or their toxins have the most importance of public health and economic consequences. Losses may be due to outbreaks include medications, charges, increased production wastes, loss of business, recall and damage of products, and investigation of the outbreaks.

LITERATURE REVIEW

Importance and protective bacterial cultures in Cheese

Cheese microbiota is divided into starter Lactic Acid Bacteria (LAB), adjunct microbial cultures, and secondary microorganisms. The first group is involved in acid production during manufacture and contributes to the ripening process, while the last two groups play an important role during ripening. The secondary microbiota is formed by spontaneous occurring nonstarter lactic acid bacteria, other bacteria, yeasts and moulds which grow internally or externally in most cheese varieties [10]. LAB is the major component of the starters used in fermentation, especially for dairy products, and some of them are also natural components of the gastrointestinal microbiota [11]. Lactococcus, Leuconostoc and *Lactobacillus* (homo- and hetero- fermentative species) are the most prevalent genera of LAB in cheeses [12]. They are potentially important in autolysis, and associated cheese ripening events [13]. The term “starter culture” refers to one or more selected strains of food grade microorganisms used to produce fermented foods with some desirable characteristics in appearance, body, texture and flavor [14]. The most often starter bacteria present in cheeses are from the genera Lactococcus, *Lactobacillus*, Streptococcus, Leuconostoc and Enterococcus [10]. Secondary cheese microbiota probably has its origins in raw material, post-pasteurization airborne contamination, cheese making equipment or ingredients, or pasteurization survival. In Irish cheddar cheese the main nonstarter LAB isolated were *L. paracasei*, *L. plantaram*, L. curvatus and *L. brevis* [15]. Screening of different LAB for availability as protective cultures to inhibit the growth of foodborne isolates; bacteria, yeasts, and fungi were needed.

Lactic acid bacteria to preserve milk and cheese

Lactic Acid Bacteria is a Gram-positive bacterium which have the potential to ferment milk and enriched it with nutrients which improve the safety of food, as well as provide therapeutic benefits [16].These bacteria is employed to extend the product life span through fermentation of milk. This also aids in the preservation of milk’s nutritious constituents. The use of LAB in milk fermentation results in high-quality dairy products with excellent organoleptic characteristics. Lactic acid bacteria variants from the *Lactobacillus* genus are widely used for health-promoting bacteria [17]. Several *Lactobacillus* variants are thought to have immunological mediator, antihypertensive, anticancer properties and calcium binding, LAB fermentation, as per some research, protects against diarrheal disorders by altering the structure of microbes in the gut. Bacteriocins, which are protein antibacterial compounds, are also produced by this LAB [18].

Traditional cheeses from raw milk

Traditional cheeses which are normally produced from raw milk are very popular due to their intense, unique taste and aroma, as well as high nutritional value. However, several diseases related to the consumption of raw milk cheese have reported in the United States and other parts of

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the world. For example, many cases of death have been reported in raw milk cheeses contaminated with *E. coli* in the United States, Canada, and Europe. Therefore, raw milk cheese is one of the risky products [19]. Various foodborne bacteria including *Brucella melitensis*, *Campylobacter* spp., *Coxiella burnetii*, *E. coli* O157:H7, *L. monocytogenes*, *Mycobacterium bovis*, *Salmonella*, *Staphylococcus aureus* and *Streptococcus* spp. are associated with raw milk cheese [19,20]. The growth of pathogens is a hazardous point in the safety and quality control of raw milk cheese, which mainly depends on the kind of cheese and its manufacturing technology. The high microbial contamination of raw milk due to manual milking and secondary contamination may lead to many diseases in humans. Lighvan is a traditional Iranian cheese that is made from raw milk which may produces serious health problems, due to the limited ripening period of this type of cheese. Thus, different cheese prepared with pasteurized milk was compared with raw milk cheese. The results showed a reduction in the microbial population over the ripening time in both types of cheeses. However, coliforms and *E. coli* were seen in raw milk cheeses until the last day of ripening. Moreover, the raw milk cheeses indicated a higher lipolysis index than the pasteurized ones. According to the obtained results from the sensory evaluation, the raw milk cheese indicated higher acceptability compared with the pasteurized milk cheese. However, since the presence of *E. coli* makes the cheese inedible, it seems that the pasteurization of milk is mandatory for the production of this type of cheese.

Microorganism's contaminated milk and cheese product

Bacteria are one of the major spoilage microorganisms in milk and cheese which provides an excellent growth medium for microorganisms like gram-negative bacteria, coli forms, aerobic psychrotrophic, spore-forming bacteria, yeast, and molds. Pathogenic bacteria are transmissible to humans through milk and milk products. Microorganisms occurring in milk and milk-based products are *B. cereus*, *C. jejuni*, *Brucella* spp., *Coxiella burnetii*, Shiga toxin producing *E. coli* (particularly *E. coli* O157:H7), *Mycobacterium paratuberculosis*, *L. monocytogenes*, *Yersinia enterocolitica*, *Salmonella* spp., and *S. aureus* which have been linked to milk-borne outbreaks [21]. Listeria, *Salmonella*, Campylobacter and Vero toxigenic *E. coli* seem to be the four leading bacteria causative for milk-borne illnesses. Moreover, *B. cereus*, *S. aureus* and *M. paratuberculosi* are developing microorganisms that are important in dairy contamination. One of the most important sickness for human and animal and sometimes death is Salmonellosis which is an illness caused by *Salmonella* spp. [22]. In addition, several strains of *Bacillus cereus* are well known as toxin producers causing food poisoning. Hemolytic Uremic Syndrome (HUS) is a condition that can occur when the small blood vessels in your kidneys become damaged which lead to kidney failure in young children. In many cases, HUS is caused by infection with certain strains of *E. coli* which is potentially a lethal condition, especially in children. It was reported that many foodborne outbreaks were linked to cheese consumption, and the most common causative agents were Shiga Toxin-producing *E. coli* (STEC) and bacterial toxin [23].

There is well documented and represents concern about *E. coli* 0157-H7 since up to 10% of people infected with these bacteria which develop haemolytic syndrome. *Salmonella* and *L. monocytogenes* were detected in cheeses made from raw milk or milk that has undergone a lower heat treatment than pasteurization. Big attention is focused on milk, cheese and ice cream contaminated with pathogenic bacteria like *L. monocytogenes*, *Yersinia enterocolitica*, *Campylobacter jejuni* and enteropathogenic *E. coli*. According to Reguillo et al., [24] saprophytic bacteria with glycolytic, lipolytic and proteolytic activities are found in raw milk. The presence and multiplication of these organisms can be constrained by storing fresh milk below 7°C, which favors the selection of psychrotrophic species such as *Pseudomonas* spp., *Micrococcus*, *B. cereus*, *Alcaligenes* spp., and *Lactobacillus*, Streptococcus, as well as Enterobacteriaceae. High bacterial counts in raw milk are caused by inadequate cleanliness and biofilm formation in storage tanks and milking equipment [25,26]. All of them contribute to the growth of microflora in milk. The normal souring process of milk is advantageous as it leads to the formation of an acid that is beneficial during the processing of butter but pathogens are also unable to grow in low pH environments, but molds can grow.

Psychrotrophic microorganisms

Psychrotrophic microbes are cold loving microorganisms like Pseudomonas spp. in the cold raw milk which can produce heat stable proteases. These enzymes may spoil pasteurized milk at 72-75°C for15-20 Sec. and ultra-high temperature-treated milk at 130-150°C for 2 to 4 Sec. in dairy products [27]. Besides exhibiting spoilage features, some psychrotrophs possess an opportunistic pathogenicity and are inherently toxin producers in addition to they were resistant to antibiotics. Regarding quality, this bacterial group has become a key problem for today's dairy industry as causing of spoilage and significant economic problems [28]. They represent a substantial percentage of the bacteria in raw milk and they hydrolyze and use large molecules of proteins and lipids for growth. For cottage cheese, the typical pH is favorable for the growth of Gram-negative psychrotrophic bacteria with the pH ranging from 4.5-4.7 and insufficient NaCl to limit the growth of contaminating bacteria; therefore, psychrotrophs are the bacteria that normally limit the shelf life of cottage cheese [29]. The psychrotrophic bacteria such as Achromobacter, Alcaligenes, Flavobacterium and Pseudomonas are of concern in cheese [30]. The presence and multiplication of these organisms can be constrained by storing fresh milk below 7°C, which favors the selection of psychrotrophic species which can form lipolytic or proteolytic enzymes, which break down raw milk constituents, resulting in flavor and texture changes [25,31].

Coliforms in milk and cheese products

Coliforms belong to the Enterobacteriaceae family and are gram-negative bacteria that are facultative in nature and have a rod-shaped structure. *Coli*forms are not a natural part of raw milk's microbiota, but they can get into it through the environment, milking machinery, or the udder [32]. They also can break down the diacetyl content of sour cream and lactose, which helps in the production of a yogurt-like flavor. Citrobacter, Klebsiella, *E. coli* and *E. aerogenes* are some of the species which are also commonly found in milk. Coliforms can produce gas in cheese when the starter culture produces lactic acid slowly due to their short generation time. When the pH of the soft, mold ripened cheese rises, it creates an ideal environment for the coliform to grow. They do not indicate a serious illness, but their presence confirms the presence of pathogens of fecal origin [33]. In cheese production, slow lactic acid production by starter cultures favors the growth and production of gas by coliform bacteria, with coliforms having short generation times under such conditions. In soft, mold ripened cheeses, the pH increases during ripening, which increase the growth potential of coliform bacteria [29].

***Escherichia coli* in milk and cheese products**

E. coli are Gram-negative bacteria that are typically found in the small intestines of warm-blooded animals, such as humans. In the human upper gastrointestinal tract, *E. coli* is usually innocuous; nevertheless, it can be hazardous to persons who have a weak immune system. Pathogenic *E. coli* infections can cause urinary tract *Escherichia coli* 0157:H7 is recognized as an emerging pathogen. Several outbreaks involving *E. coli* 0157:H7. The organism causes hemorrhagic colitis or bloody diarrhea. This infection is usually characterized by severe abdominal cramps followed by watery and grossly bloody stools [34,35,20]. The illness occasionally involves Hemolytic Uremic Syndrome (HUS), which is characterized by serious kidney dysfunction with urea in the blood. The significance of *E. coli* 0157:H7 as a food base pathogen is not fully known. Investigations are underway to develop effective and sensitive methods for its detection and isolation. The organism is heat labile and is readily inactivated through cooking and proper pasteurization. Enterotoxins E. coil 027:H20 is another strain of *E. coli* that caused gastroenteritis associated with eating imported Brie cheese [36,37]. Several similar outbreaks occurred in the United States and one in the Netherlands associated with consumption of cheeses from France [35,38].

There are four types of *E. coli* strains that cause diarrhea: (1) EIEC (Enteroinvasive *E. coli*), (2) STEC/EHEC (Shiga toxin-producing *E. coli*), (3) ETEC (Enterotoxigenic *E. coli*), and (4) EPEC (Enteropathogenic *E. coli*). STEC are primarily found in raw milk or other milk products and have also been linked to a number of food-borne outbreaks [39]. Shiga-like toxins (stx) are produced by STEC, with stx1 and stx2 being the most important pathogenic factors for toxicity. It can grow at 37°C which is the appropriate temperature for STEC development. However, research revealed that it might flourish at temperatures ranging from 10 to 50°C and in an acidic (pH 4.5) or low-water activity conditions. STEC infection is most common in raw milk and cheese samples. *E. coli* O157:H7 was detected in lactic cheese, cream cheese and feta cheese at pH 4.5 [40].

***Salmonella* in milk and cheese products**

Salmonella is a Gram negative facultative anaerobic bacilli, motile belong to the Enterobacteriaceae, forming non endospore, oxidase and catalase negative, form no pigmented colonies. It use citrate as a sole source of carbon and energy, and develop unfavorable effects when oxidatively deaminate phenylalanine & tryptophan [41].It could transform nitrite to nitrate, generate gas from glucose, manufacture hydrogen sulphide on triple-sugar iron agar andis one of the most prevalent pathogens in the food industry. Studies about this microorganism date up to 100 years and have been the causative agent on several outbreaks of foodborne diseases particularly in dairy products like cheese [42]. It can be detected in the intestinal tract of healthy animals and contamination of milk mainly occurs during milking operations [43]. Two species *S. typhi* and *S. paratyphi* are 2 of the most well-known species for inducing typhoid fever, which is more severe and also has a 10% death rate. Typhoid fever typically takes 1-3 weeks to develop, as well as the main symptoms include a high temperature, headache, gastrointestinal aches and diarrhea. Septicemia or a persistent infection might also happen. Non typhoidal salmonellosis is a disease that is caused by serotypes other than *S. typhi* and *S. paratyphi*. Although salmonellosis can be fatal in certain situations, it is usually more self-limiting and far less dangerous than typhoid fever. Non typhoidal salmonellosis manifests itself 6-72 hrs of exposure, with symptoms including nausea, stomach cramps, vomiting, fever, diarrhoea, and headache. All *S. dublin*, *S. heidelberg*, *S. saint* paul, *S. typhimurium*, *S. newport*, *S. Kottbus*, as well as *S. newbrunswick* are among the *Salmonella* serotypes linked to milk infections in the United States [44]. Raw milk and cheese have been the main sources of infection in these outbreaks, with the exception of two outbreaks induced by pasteurized milk. The milk outbreak resulted in a lot of S. enteritidis isolates. Nonetheless, since 2000, one of the most common species linked with the milk outbreak seems to have been S. typhi, as well as the potentially contaminated carriers have largely been cheese or cheese-related goods.

Listeria monocytogenes in milk and cheese products

Listeria monocytogenes is a facultative anaerobic, gram-positive, non-spore-forming, rod-shaped bacterium with coccoid or diphtheroid morphology and catalase positive. It is one of the principal genera concerned with human infection [45] and is psychrotrophic bacteria which can grow at temperatures from 3 to 45°C and the optimum temperature was between 30-37°C. Unlike numerous other infectious infections, *L. monocytogenes* can survive and flourish in a broad range of temperatures (4°C to 50°C) and pH conditions [46]. This bacteria forms bluish-green colonies on trypticase soy agar (oblique illumination) and was motile when grown in trypticase soy broth at 25°C. *L. monocytogenes* is heat-sensitive, inactivated by pasteurization, weakly hemolytic on medium containing blood and has a pH range from 4.8 to 9.6. Due to its high mortality rate specially for pregnant women and immuno compromised patients, it is one of the main concerns for ready to eat food including cheeses [47]. EFSA [23] reported 2480 cases of listeriosis in soft and semisoft cheeses. Listeriosis can manifest a variety of symptoms in humans, including meningitis, infectious abortion and perinatal septicemia [48].

Listeria monocytogenes is an emerging foodborne zoonotic pathogen of public health significance. Pal and Awel [49] study the public health significance of *L. monocytogenes* in milk and milk products. It is mentioned in European report that noncompliance of *L. monocytogenes* primarily occurred in soft and semi-soft cheeses made from raw or low heat-treated cow's milk [50]. There has been some discussion regarding the potential for *L. monocytogenes* in milk that survive pasteurization. The organism has been shown to be capable of significantly more rapid growth in pasteurized milk than in raw milk at 7°C and is also capable of growth at 4°C in pasteurized milk. It has been shown to grow slowly in butter made from contaminated cream at 4 or 13°C, and to survive for several months in frozen butter without any appreciable decrease in number [30].

Raw milk and soft cheese are both the major sources of *L. monocytogenes* where numerous investigations showed that certain variants of *L. monocytogenes* may develop in dairy with variable fat content at different temperatures, and cell numbers have increased in the existence of cane sugar. In general, invasive & non-invasive diseases have been associated with outbreaks of dairy products, with a fatality rate of 20-30%. Listeriosis, a gastrointestinal condition characterized by diarrhea, mild fever, muscle pains, nausea and vomiting, is generally associated with non-invasive *L. monocytogenes* diseases. Invasive diseases, which include meningitis and septicemia, have more severe symptoms. Listeriosis can be treated from a few hours to two to three days for healthy persons while in pregnant ladies, neonates, the elderly, or persons with a weakened immune system, the disease can continue for up to 3 months causing nervous system damage [51].

Staphylococcus aureus in milk and cheese products

Staphylococcus aureus is a Gram-positive cocci, oxidase negative and catalase positive. It forms clusters showing pigmented colonies when grown in nutrient agar. It often forms characteristic clumps resembling bunches of grapes [52]. Staphylococcal intoxication (staphylococcal food poisoning) results from ingestion of enterotoxins, synthesized during growth of *S. aureus* in foods. Enterotoxin production is most common amongst *S. aureus* isolates of human origin and there is a strong correlation with production of the enzyme coagulase [53]. It cause a typical food poisoning which is characterized by nausea, vomiting, and diarrhea and it is linked to consumption of food contaminated by one or more preformed enterotoxins that are produced when the bacterial load exceeds 10^5 CFU/g and Staphylococcal food poisoning in soft cheese made from raw milk has been reported [54-57].

Spore forming bacteria in milk and cheese products

Raw milk is the usual source of spore-forming bacteria in finished dairy products. Their numbers before pasteurization seldom exceed 5,000/ml. They can also contaminate milk after processing. The most common spore-forming bacteria found in dairy products are *Bacillus cereus*, *B. licheniformis*, *B. megaterium*, *B. mycoides* and *B. subtilis* [29]. Bacterial spores resistant to heat treatment that was present in the raw milk. The improper packaging of milk after heat treatment allows the entrance of microbe's recontamination whereas the spores of *B. cereus* can form biofilm [58].

In milk products, the commonly found spore formers are *Bacillus licheniformis*, *B. mycoides*, *B. subtilis*, *B. megaterium* and *B. cereus*, where raw milk acts as a medium. They are also present if the packaging is not done properly even after heat treatment. Many studies have shown that many spores become activated after pasteurization and develop when they reach the appropriate temperature [59]. Some microorganisms can also with stand the ultra-high temperature of milk, such as B. stearothermophilus [60]. They cause "flat sour" in canned milk products because they produce acid but not gas [61,20].

Molds and yeasts are important contaminants of dairy product

Molds and yeasts are important contaminants of dairy product as a favorable niche for their growth. They are causes of visible or non-visible defects, such as odorand flavor, leading to cheeses waste and loss. Molds are most commonly seen growing on the surface of cheese when there is an abundance of oxygen, but some can also grow in low oxygen tension. They are seen to be growing in cheeses that are vacuum packed, especially *Penicillium* and *Cladosporium* and degradation of sorbic acid and potassium sorbate occurs in sorbet-containing cheese. Therefore, it leads to the formation of trans-1, 3-pentadiene and develops an off odor, and that flavor is termed as kerosene [62].

Yeasts are eukaryotic microorganisms classified as fungi and play an essential role in the preparation of certain fermented dairy products such as kefir, kefir-derived raw milk cheeses and in the ripening of many cheeses. Yeast is one of the primary causes of yogurt and fermented milk spoilage because they thrive in low pH environments. To have a shelf life of 3 to 4 weeks at 5°C, a good quality yogurt ought to have not over than ten cells. Because they can grow at a lower pH than any cultured product, like, sour cream or buttermilk, they develop an off flavor, which is then referred to as yeasty or fermented. They reduce the diacetyl, resulting in a yogurt-like flavor and the high numbers of yeasts in some acidified dairy products may be attributed to their ability to tolerate low pH values and low water activities; and to grow at low temperatures, to assimilate lactose and organic acids such as succinic, lactic, and citric acid; and to tolerate high salt concentrations. The predominance and growth of some yeast species has been related to the ripening process due to their ability to produce extracellular proteases and lipases and grow well at 5°C. Because yeasts are recovered at the end of the fermentation stage, it is suggested that they play a secondary role in the aroma development. The occurrence of yeasts in yogurt (up to 10^6 to10^7 CFU/g) can cause spoilage [63,64]. *Candida famata*, *C. diffluens*, *Kluyveromyces marxianus* and *Rhodotorula glutinis* were the most molds in milk and cheese products. Geotrichum candidum is the most commonly found yeast that causes cottage cheese spoilage. When the yeast count in yogurt reaches 10^5-10^6 CFU/g, it results in the production of off-flavors as well as gas production. Cheese has a high nutritional value as well as a low pH, surface moisture, lactic acid, amino acids, and peptides. All of this promotes yeast growth, which results in the production of alcohol and CO_2. If yeast is present, it causes the swelling of cheese packets packed in a vacuum or modified atmosphere. *Candida* spp., *G. candidum*, *Pichia* spp., *Kmarxianus* and *Debaryomyces hansenii* are the most commonly found yeasts that cause contamination. Recently, consumers have an increasing awareness of their health risks caused by the utilization of cheese from local market or contained preservatives. So, this is a growing need in the dairy industry to extend product shelf-life and prevent spoilage by natural preservatives and/or new methods of conservation.

Food borne illness linked to cheese consumption

Foodborne illnesses related to cheese consumption have occurred in many countries. *S. aureus* infections have been linked to the use of unpasteurized milk or to contamination due to improper handling because the pathogen at more than 5 Log CFU/mL produces heat-resistant enterotoxin [65,66]. Intake of 20 to 1,000 ng of the enterotoxin can cause typical symptoms of *S. aureus* infection [67]. In England, consumption of *S. aureus* enterotoxin contaminated Stilton cheese (internally mold ripened semisoft blue cheese produced by using *Penicillium roqueforti*, caused 155 illnesses [68,69]. In the USA, cheese has also been a food vector for *S. aureus* food borne outbreaks [70]. In 1981, improper cheese pasteurization led to 16 cases of *S. aureus* food borne illness in USA. In 1999, Minas cheese (soft cheese) made from raw bovine milk caused two outbreaks in Brazil affecting 378 individuals [71]. Lately, six *S. aureus* outbreaks occurred in France due to consumption of enterotoxin type E in soft cheese that was processed during weeks 40 and 41 [72]. Cheese-related *L. monocytogenes* outbreaks have had a relatively high fatality rate (15% to 30%), leading to increased public awareness of this infection [73]. Clinically, *L. monocytogenes* causes sepsis and meningitis in immuno compromised individuals (e.g., transplant patients, elderly patients receiving chemotherapy, individuals with diabetes or liver disease), and around 25% of the invasive listeriosis cases occur in pregnant women [74]. Of 13 *L. monocytogenes* serotypes, three were account for more than 95% of human illnesses [75]. In 2005, 10 cases of *L. monocytogenes* induced listeriosis occurred in a small northwestern Swiss town with three fatalities due to bacteraemia. Eight of these cases were in older immuno compromised patients, while two cases occurred in pregnant women leading to abortion. This outbreak occurred after these patients had Tomme (soft cheese) contaminated with *L. monocytogenes* serotype 1/2a [76]. Fretz et al., [77] reported that Quargel (acid curd cheese) contaminated with *L. monocytogenes* serotype 1/2a caused four deaths out of 14 cases in Austria and Germany in 2009. In the USA, the outbreaks were caused by the intake of Mexican-style cheese made from pasteurized milk [78]. In addition, there was another Mexican-style cheese-related outbreak of listeriosis in North Carolina in the USA [79]. Even Japan, which has a relatively low cheese consumption compared to the western countries, also had *L. monocytogenes* serotype 1/2b outbreaks due to cheese consumption resulting in 86 cases [73]. *Salmonella* is a well-recognized foodborne pathogen. All *Salmonella* strains are gastroenteritis-inducing pathogens and various *Salmonella* serotypes have been involved in cheese-borne outbreaks [80]. In cheese, Shiga Toxin-producing *E. coli* (STEC) strains are important food borne pathogens [81]. In 2003, *E. coli* O157:H7 was linked to an unpasteurized Gouda cheese-related outbreak involving 13 cases in Canada [82].

A microbial risk assessment is considered a scientific and methodical tool for preventing, regulating, and understanding the risk caused by hazardous microorganisms [38]. The microbial risk assessment consists of four components: hazard identification, exposure assessment, hazard characterization, and risk characterization [83]. This methodology has been used in many countries to prevent the occurrence of foodborne illnesses, to reduce the risk of foodborne illness outbreaks, and to establish microbial criteria. As discussed in previous sections, a number of cheese-related foodborne illness outbreaks have occurred. Additionally, cheese consumption has increased despite these foodborne illness outbreaks, which raised the necessity of microbial risk assessments on cheese [70].

Prevention of cheese contamination

Milk and dairy products play a key role in healthy human nutrition and development throughout life [83,84]. Dairy products such as pasteurized milk, butter, cheese, cream, ice cream, and yoghurt are all susceptible to microbial spoilage because of their chemical composition [85]. Ready-to-eat food products including cheeses are intended for consumption without any treatment between final production and consumption [86]. As the quality of raw milk improved, pasteurized milk the pathogenic bacteria likely to be of significance in milk as well as most of the spoilage bacteria will be killed. The addition of carbon dioxide to milk and milk products reduces the rates of growth of many bacteria [29]. Maintenance of the proper hygienic conditions during the processing of milk can reduce the prevalence of bacteria, which spoil the milk product [85]. Good hygiene practices during milking and subsequent handling of milk are essential to reduce the risk of contamination on the farm and in the milk processing plant [87]. Worldwide standardized pasteurization practices would be an effective first step in eliminating or reducing the levels of many spoilage microorganisms [29]. In order to minimize the health risks of milk and dairy products at the point of consumption, all food-chain operators, including the dairy farmer, processor, distributor, retailer and consumer, need to take necessary actions to maintain food safety [84]. It is recommended that microbiological monitoring of the milk and dairy products should be conducted to keep the products safe to the consumers [88,89].

DISCUSSION AND CONCLUSION

Milk and dairy products has an outstanding nutritional quality but is also an excellent medium for bacterial growth. Contamination of milk products by spoilage producing microbes results into a great financial loss to the dairy sector. Pathogenic bacteria can contaminate raw milk in two ways. The first way is an endogenous contamination where the milk is contaminated by a direct transfer from the blood due to systemic infection to the milk and by exogenous contamination where the milk is contaminated during or after milking by the feces, the exterior of the udder and teats, the skin, and environment. After milking, contaminating microorganisms from equipments and utensils, from environment and even from the employees responsible for obtaining and handling milk, are the most important sources of contamination under the technological point of view, since it may cause undesirable alterations in the product. Psychrotrophic bacteria are notorious contaminants of milk in the refrigerated dairy food chain. Measures should be taken to prevent the spread of zoonotic diseases among animals and from animals to milk include controlling the infection from feed and fodder, improving hygiene of animals sheds and the environment, safe waste management and good and easy access to veterinary service. It is emphasized that god manufacturing practice, good hygienic practice and hazard analysis and critical control point should be implement in dairy industry to prevent the contamination of dairy products. Although cheese is considered microbiologically safe, cheese-borne outbreaks have still occurred, especially in soft cheese. Predictive models have been used to determine the factors related to bacterial growth such as pH and Aw, and the results from predictive models are used in microbial risk assessment in many countries to uncover cheese that might pose risks to consumers. The microbial risk assessment then showed that microbial risk increased in cheese with high moisture content, especially for raw milk cheese. However, applications of pre- and post-harvest preventions can reduce the microbial risk. Even though microbial risk assessments were conducted for cheese, in some studies limited data were used for predictive models, temperature profile for distribution and consumption patterns as well as dose-response model, especially for susceptible group such as elderly, pregnant women, and immuno compromised individuals.

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