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# Microbial Contamination in Milk Quality and Health Risk of the Consumers of Raw Milk and Dairy Products

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## Abstract

The dairy products industry is going toward safe milk and its products in the food market. Milk quality and food safety concern in the consumers' health and nutrition in public health surveillance prevent food-borne diseases, food poisoning, and zoonosis risk by raw milk and fresh dairy products. The aim of this work is focused on milk microbial contamination and its impacts on milk production and dairy industry with their implications in milk product quality, food-borne diseases from raw milk, and unpasteurized milk by food-borne pathogen microbial contamination and milk and dairy product spoilage. The microbial milk contamination source comes from herd hygiene and health status, mastitis prevalence, production environment, and milking parlor and milk conserving practices in dairy farm. Moreover, these facts are implicated in milk quality and milk spoilage and unsafe dairy products. The milk production system and the dairy plant operations keep track in pasteurized milk and fresh dairy products reviewing the traceability in field situational diagnosis report.

**Keywords:** cow milk production, food safety, food-borne disease, milk contamination, milk spoilage

## 1. Introduction

The objective of the dairy industry is to maintain productivity and competitiveness in a growing milk commerce, which is demanding a large volume of milk and a wide range of dairy products in the food market and the preferences of the final food consumer with remarkable differences according to patterns of consumer behavior by demographic categories, culture, and socioeconomic variations in the human population in the food market [1, 2]. The consumers prefer a safe and healthy milk product selection, with a great variety and availability

in the market. This fact affects the health and nutrition consumer's information about the milk products made with raw milk [3, 4]. Milk is also an important source of bacterial infection for human health, when milk is consumed without pasteurization [5–7]. Milk is a basic food in the human diet with great value as a nutritious healthy food; in the first years of human life, milk and dairy products are an important nutritional fact in the diet of the adult population [8]. According to the sustainable production system, their main priorities are contributing to the regional social and economic development, land resource preservation, and animal welfare quality in dairy cattle husbandry maintaining a productive healthy cow herd to produce high milk quality [9]. The global responsibilities of the milk industry and big dairy farm and small holder producers are offering high-quality milk and safe dairy products in the commerce preventing food-borne diseases to spread in the population [10, 11].

## **2. The source of milk contamination**

The milk market requires and offers safe and high-quality products, preventing a contamination source by good hygiene practices to reduce a possible exposure of food-borne pathogens and chemical milk residues. The mammary gland participates in the excretion of numerous xenobiotic substances from veterinary drug milk residues and contaminants originated from milk and other chemical residues to environmental pollutants on the grasslands, animal feedstuffs, and the field crops [12]. The presence of residual concentrations of milk contaminants and pathogens is an indicator of milk quality in cow dairy farms. In evaluating the raw milk bulk tank at the dairy farms, quick information about udder health status, environmental pathogens, milk chemical residues, and antibiotics is obtained [13–16]. The relationship among dairy cow production and milk safety and dairy product quality is considered in different subjects: raw and pasteurized milk contamination and microbial aspects of the quality of milk and dairy products, cow husbandry in animal welfare influence, feeding conditions, and herd hygiene practices and milk composition. Also the environmental pollutants, and chemicals from agriculture, pesticides residues, drug veterinary residues and management in dairy production. Those relationships that exist in milk production are auditable and selectively regulated to prevent milk contaminants. The contaminants agents are tracking and monitored at milk parlor, in refrigerated milk tank and the milk bulk tank on platform by the application of proper sampling methods required in the Control Analytical Methods for milk quality in Dairy Industry Management assurance the food safety [17]. Are affecting milk production and dairy products related to food safety and milk quality [18]. In the phenomenon of the climatic change, the zoonosis and food-borne diseases are priorities in the public health programs in many countries, ones of the surveillance task is the diseases transmitted by raw milk, and unpasteurized fresh dairy products [19, 20]. The aflatoxin M1 contamination levels in milk appear to be a serious health hazard derivate from hepatotoxic and carcinogen effects of aflatoxin M1, which show a high risk on milk food safety. The milk contamination risk is established through the forages, corn and concentrated feeds; those are contaminated by aflatoxin B1 (AFB1). There is an aim to watch over the limit exposure to aflatoxins in dairy by imposing regulatory limits [21]. The presence of biotics from grazing cows and conserved pastures and feeding grains, like aflatoxins AFB1 and AFM2, has been usually monitored in milk [22]. In dairy production, an important practice is oriented to reduce environment fungal contamination and the proper conserving methods of silages, forages, and grains for animal feed [23]. The controlled grazing land is a relevant characteristic of the milk

produced at grazing, was its richness in beta-carotene, lutein, vitamin E and sesquiterpenes among winter seasonal period monitored farms. These conditions should have a great influence on the physicochemical milk profile of raw milk bulk tank at dairy farm, in comparison with the milk of the producers with herds fed with diets rich in concentrate, corn silage, and pasture [24]. The silage is a significant source of contamination of raw milk with spores compared with grass and maize silage. Preventive management of outgrowth of aerobic spores in silage by the application of acid lactic bacteria or chemical additives can improve the silage fermentation; it will contribute to reduce the total spore load of raw milk for dairy process [25]. The microbial contamination of milk could be produced from sources of bacteria and fungi are identified in grassland and other feedstuffs. The health herd status will be implicated in specific zoonosis produced by animal carriers of *Salmonella* spp., *Mycobacterium bovis*, *Mycobacterium avium* subsp. *paratuberculosis*, and Brucellosis and *Escherichia coli* 0157H7 are focused by sanitary conditions and the health risk [26, 27]. The health level status is an important issue in milk production; the maintenance of herd hygiene, disease control programs, and preventive management is oriented to reduce the prevalence of contagious diseases in dairy cattle [28]. The others sources of milk contamination may be present in the herd management, poor hygiene milk practices, mastitis, infectious pathogens in infected cows and the presence of environmental pathogens by poor animal hygiene [7]. The good hygiene practices in the herd cow is an important fact for to reduce contamination from production environment, feces, slurry, soil and mud those are microbial sources for the udder contamination. The poor hygiene practices could occurs microbial milk contamination, pathogens dissemination, and udder contamination may be occurred at milking time between cows, hands of milker man and milk machine from others [29]. The microbial analysis of raw milk are influenced by microorganisms present in the teat canal and the surface of teat skin [30]. The bad hygiene practices and poor cleanness procedure equipment, the surrounding air in the milk parlor, as well as other environmental factors including housing conditions, water supply, and during feeding have an important effect on the milk contamination [31, 32]. Other microbial contamination of milk possibility may occur during the long milk storage, under low insufficient temperature [33]. Usually contaminated environments are a potential source of food-borne pathogens and spoilage bacteria present in raw milk bulk tank in the dairy farm, which are affecting the milk quality and emerging public health risk [34–36]. The cow herd should be monitored for preventing possible food-borne pathogens and food intoxications, which is a preventive strategy for health risks and to diminish the poor dairy product quality. The variation of the milk components of bulk milk among herds, could give an approach of the grassland interaction among the dairy cows, the environmental pollutants, and the environment health status have a potential public health risk [37]. In dairy farms the milk tank study is widely used for monitoring the herd udder health status as an indicator of quality for milk producers used by the dairy industry [38]. Through a microbiological study, it is possible to know the possible bacterial contamination source for modifying hygiene practices and to recall critical bacterial contamination in milk traceability for preventing the milk spoilage on the quality of the pasteurized milk and dairy products, affecting the consumer's acceptability. When milk food-borne disease outbreaks occur in the human population, there are other many reasons to trace back and investigate; fresh cheeses are elaborated with non-pasteurized milk or elaborated without proper hygiene conditions using pasteurized and unpasteurized milk [3, 28, 39]. Outbreaks of milk food-borne diseases (**Table 1**) have been associated with diseases due to infected foods and contaminated dairy products after pasteurization [40]. Another consideration of food-borne pathogens in raw milk into dairy food processing plants can persist in



Mammary gland health status	Cow herd health status	Production environment	Production land water source
<i>S. aureus</i>	<i>Mycobacterium bovis</i>	<i>Listeria monocytogenes</i>	Hepatitis A virus*
<i>Streptococcus agalactiae</i>	<i>Mycobacterium avium</i>	<i>Salmonella</i> ssp.	<i>Leptospira</i> spp.*
<i>Streptococcus</i> spp.	subsp. <i>paratuberculosis</i>	<i>E. coli</i> O 157:H7	* <i>Bacillus</i>
<i>Streptococcus pyogenes</i>	<i>Brucella</i> ssp.	<i>E. coli</i> (STEC)	<i>licheniformis</i>
<i>Streptococcus zooepidemicus</i>	<i>S. aureus</i> MRSA-LA	<i>E. coli</i> (EHEC)	* <i>Bacillus subtilis</i>
(B-hemolytic)	<i>Salmonella</i>	<i>Yersinia enterocolitica</i>	* <i>Pseudomonas</i>
<i>Streptococcus</i> Lancefield C group)	<i>typhimurium</i> phage type 561 (STM DT7)	<i>Enterobacter sakazakii</i>	<i>aeruginosa</i>
<i>Corynebacterium ulcerans</i>		<i>Campylobacter jejuni</i>	* <i>Clostridium</i>
		<i>Enterococcus faecalis</i>	<i>disporicum</i>
		<i>Citrobacter freundii</i>	<i>Aspergillus</i> spp.
		<i>Bacillus cereus</i>	Aflatoxin M1
		* <i>Cryptosporidium parvum</i>	Mycotoxin B1
		<i>Coxiella burnetii</i> *	
		<i>Toxoplasma gondii</i> *	

\*Occasionally  
+Involved in enteric diseases

**Table 1.**  
*Food-borne pathogens and food poisoning in milk production source.*

biofilms, with subsequent contamination of processed milk products. Inadequate milk pasteurization allows survival of food-borne pathogens in milk and dairy products [41, 42]. The health educational program for the human population should be oriented to reduce the risk of exposure for food borne diseases by the information in the end of the food chain, by adequate handling of milk and dairy products at home for prevention of the risk of food-borne diseases thought the consumption of non-pasteurized raw milk and dairy products prepared with unsafe hygiene practices in dairy food process [42–44].

3. Udder health and the milk quality

The infectious bovine mastitis in milk production is considered a disease with high economic impact reducing milk yield and the industrial dairy process and food safety. *S. aureus* and *Streptococcus agalactiae* are the most prevalent contagious pathogens in bovine mastitis from dairy herds around the world. The intramammary infection in dairy cows is relationship with infections by contagious pathogens and environmental pathogens as coliforms bacteria and *Streptococcus uberis* mostly are occurring in the dry period and the lactation in clinical cases regularly [45]. In the dairy herd with low prevalence of subclinical mastitis, the milk losses could be estimated between 3 and 5 % of the milk yield production, comparing to a herd average within milk somatic cell counts about 200,000 cells/mL [46]. The change in milk yield and composition depends of the severity and duration of the mammary gland infection and somatic cells counts. In an uninfected mammary gland that contains <100,000 somatic cells/mL, >200,000/mL, somatic cells counts suggest an incipient mammary gland inflammatory response [47, 48]. The bovine mastitis in dairy herds affects milk composition and somatic cells counts, serum protein, and proteolytic enzymes. Other undesirable milk mastitis conditions are bacterial toxins and abnormal proteins derived from inflammatory tissular response, which influence milk flavor and taste as well as milk product stability in the dairy process [46]. A wide variety of environmental pathogen exposure routes have been documented during the last decade; at present new pathogens and transmission routes are emerging. The main food-borne disease outbreaks comes from notified from the consumption of milk food products. The accidental

ingestion of fresh dairy products contaminated with *E. coli* and other food-borne pathogens were originated from the soil or water provokes mainly enteric diseases. The knowledge regarding to biotic and abiotic factors involved in the survival or enhance of the agents, and their potential dissemination in the environment, and exposure routes of the main food borne pathogens, are considered now in the investigation of the public health risks from dairy farms [49]. In the prevention and control strategies applied to mastitis in dairy herds, can be included in the program by a situational diagnosis previous provide information about of herd somatic cell counts, microbial agents and mastitis in different clinical stages. This wide study is done to provide strategic information of herd hygiene status, milking hygiene practices, and milk machines' regular maintenance for proper functioning. The monitoring of somatic cell counts from the milk tank, and the lactating dairy cows, and dry cow period. To bring information about of the mammary gland health status in the surveillance program [50–52]. Antibiotic milk residues, are commonly associated with mastitis treatment in lactating cows; non observe the legal restriction and milk discard period in medicated cows; in those cases are expected more mastitis incidence of the cows, with lower milk somatic cells counts herd-year SCC, with mean values of >500,000 somatic cells/mL, are indicating an increases of mastitis cases in the cow population during the lactation period. It will have potential detection of antibiotic residues in the tank milk; these situations illustrate the importance of the maintenance of udder health and milk hygiene practices and cow selection genetic programs [45, 53]. Pre- and post-milking disinfectant routines help to reduce dramatically the infection, while udder hygiene in the milking routine directly dismisses mastitis cow pathogen transmission [50]. The prudent antibiotic use in cow herd medication schemes will help efficacy in clinical mastitis cases and dry cow infections. In contrast an increase in the incidence of mastitis in lactating cows will increase the potential risk for antibiotic residues of milk and antibiotic bacterial resistance in herd [54]. The development of antibiotic resistance in bacterial pathogens from dairy herds, is considered an emerging public health risk as many countries derived from dairy herds and the development of antibiotic resistance in bacterial pathogens [55]. *S. aureus* resistant strains (ORSA/MRSA), which are subject of surveillance programs of bacterial antibiotic resistance in human health [56, 57]. The use of antibiotics in animal food is incriminated as to be partly responsible for emergence of antibiotic-resistant bacteria with an importance in human medicine. The methicillin-resistant *S. aureus* (MRSA) strain was identified in animal companion and small dairy herds. The MRSA in humans is wildly studied in nosocomial infections and home care patients [58, 59]. The regulations of antibiotic and veterinary drug administration surveillance in animal food should be observed by agriculture department authorities [60]. The bacterial growth inhibitor test is to be performed by different conventional methods, such as the agar diffusion test with *Bacillus stearothermophilus* variety *calidolactis*, sensitive to  $\beta$ -lactamic antibiotics. The test is less effective in the detection of spiramycin, sulfonamide, or chloramphenicol milk residues. When the test of inhibitors of bacterial growth is testing with *Streptococcus thermophilus*, *Bacillus subtilis*, *Bacillus megaterium*, *Bacillus cereus*, and *Micrococcus luteus*, the sensitivity of the test for antibiotic macrolides and sulfonamides will increase slightly [61].

#### 4. Food-borne pathogens from milk

The surveillance of food-borne disease in primary purpose in the herd is to characterize potential pathogens which are recovered from animal, milk tank, milk pipelines, and milking equipment, including the man milkers and the production environments. The monitoring programs have been designed to determine the milk

production process' critical points, the health herd level, and control of animal risk for food-borne pathogens; a survey is oriented to cut the chain of disease and exposure routes to humans preventing milk and dairy product contamination [62, 63]. The surveillance of food-borne diseases usually is difficult to research an area for population monitoring. An outbreak survey of human gastrointestinal disease could be an epidemiological indicator of food-borne disease, which may be originated from drinking unpasteurized milk; *Salmonella* species can be found in ice cream and fresh cheeses as well as *Brucella melitensis* in non-pasteurized milk and homemade dairy products mostly goat cheese [42, 64]. The main zoonotic pathogens identified in raw milk were *Brucella* ssp., mainly *Brucella melitensis*, *Listeria monocytogenes*, *Salmonella* ssp., *Mycobacterium bovis*, *Yersinia enterocolitica*, *Streptococcus pyogenes*, and *Streptococcus agalactiae*, and *Escherichia coli* O157:H7 and *Enterobacter sakazakii* are recently reported [5, 65]. New emerging pathogens causing milk food-borne diseases are considered: hepatitis A virus, *Mycobacterium avium* subsp. *paratuberculosis*, *Streptococcus zooepidemicus* (B-hemolytic *Streptococcus* Lancefield C group), *Campylobacter jejuni*, *Citrobacter freundii*, *Corynebacterium ulcerans*, and *Cryptosporidium parvum* [66]. The food-borne pathogens predominantly have been involved in human disease and have originated often in many food animals as well as animal in active carrier states such as in *Salmonella* species, *E. coli* O157:H7, *Campylobacter* species, *Yersinia enterocolitica*, *Listeria* species, *Aeromonas hydrophila*, *Leptospira interrogans*, and *Mycobacterium* species. In contrast, *Coxiella burnetii*, *Cryptosporidium parvum*, and *Toxoplasma gondii* may be infrequent [67]. The *Campylobacter* infections are expected seasonally as many cases are reported to public health services [4]. Food-borne outbreaks are incriminated occurring with contaminated fresh milk and dairy products provoking acute infections and food intoxications, occurring in family's at small villages, in declared official cases confirmed with consumption of homemade fresh dairy products elaborated with non-pasteurized milk [66, 68]. The outbreaks of food-borne disease could occur after milk pasteurization and manipulating dairy products. Outbreak cases were investigated and tested by laboratory microbial probe tests for rapid detection of microbial contamination and toxins produced by *Salmonella* species, *Listeria* species, and *S. aureus* enterotoxins [39, 67]. The enzyme-linked assays for microbial and toxins and the DNA probe test are very useful for screening food samples before processing and traceability in food conserving microbial testing [49, 68]. The dairy cattle are also known reservoirs for *Salmonella* species; the animal carrier is often asymptomatic and difficult to identify because *Salmonella* prevalence is fluctuating in the seasonal period from fecal samples, tank milk, filters milk, and water. Other areas in the farm were also positives in samples from production environments like high-animal-traffic areas [34]. The dairy cow farms suspected to *Salmonella typhimurium* phage type 561 (STM DT7 international typing scheme), are strongly investigated in environment and microbiological carrier cows prompt agent detection, for triggering control measures and herd hygiene for to cut off contamination level in dairy products [69]. The *Klebsiella* species, *Enterobacter* ssp., and *Salmonella* ssp. might be present in raw milk depending of hygienic practices in the herd [1]. *Yersinia enterocolitica* O: 8 outbreaks resulted from post-pasteurization contamination. In other cases no deficiencies in pasteurization procedures or equipment were detected. *Y. enterocolitica* O: 8 were isolated from raw-milk sample and a fecal sample, and from a fecal sample and is a such as milk bottles rinsing with untreated water prior to filling milk [70]. *E. coli* infection may occur among small residents of a community, closely related to a possible common source of infection. The epidemiologic evidence of the *E. coli* infection is evidence are supported from raw milk is the cause of infections by the number of ill persons that drank raw milk. The O157:H7 is



isolated from raw milk samples and environmental samples [71]. The *E. coli*'s ability to persist in cattle production environments contributes to the contamination and recontamination cycle of dairy cattle as well as human infection. *Escherichia coli* (STEC) are the most important emergent food-borne pathogens. Shiga toxin-producing STEC are common as colonizers in the intestine of healthy cattle and easily spread into the environment by fecal shedding by the surface application of farm effluent on soil. The bacteria can be transmitted to humans through food, such as ground beef inadequately cooked or unpasteurized milk. The prevalence of Shiga-like toxin produced by *E. coli* (STEC) in raw milk cheeses, including soft, hard, unripened, and blue mold cheeses, was mainly related to effective control strategies and must be considered on cattle farms in order to limit entry of STEC strains into the production environment [72]. The prevalence of Shiga-like toxin produced by *E. coli* (STEC) in raw milk cheeses was mainly related to serotypes O6, O174, O175, O176, O109, O76, and O162 and in minor frequency O22 serogroup [44]. Enterohemorrhagic *E. coli* (EHEC), EHEC O26: H-, has emerged as a significant cause of hemolytic-uremic syndrome in human (HUS). The source of the vehicle of contamination with EHEC O26 is not often identified; fecal samples were taken from cows of the farm that produced the incriminating milk. *E. coli* O26 infection illustrates the hazards associated with the consumption of raw milk [73, 74]. *Brucella* spp. is identified in ewes' milk cheese as an important human infection source, and it has been an important public health risk. The isolation of *Brucella* species on raw milk, goats' cheese, and ewes' cheese has been reported; *B. melitensis* was isolated from cheese samples [63]. *Bacillus* spp. contamination of raw milk from the environment of production might be originated from different sources, air, milking equipment, feed, soil, and feces, and grass differences in feeding and housing strategies of cows may influence the microbial quality of milk. *Bacillus licheniformis*, *Bacillus pumilus*, *Bacillus circulans* and *Bacillus subtilis* and strain types of the species belonging to the *Bacillus cereus* group. Higher numbers of thermotolerant sporulated organisms in milk were found from conventional dairy farms compared to organic farms [75]. Contamination of milk by bacterial spores occurred during grazing season, the spore content of milk by *Bacillus cereus* psychotropic, affects post-pasteurized milk by spore total number; this was attributed to the nipple teat contamination with soil due by low evaporation of soil water and dirty farm access this was attributed to the nipple contamination with wet soil, due by low evaporation of soil water and farm access dirty [76]. The assurance of microbial quality in dairy product, requires monitoring and identifying bacteria associated with food safety concerns in raw milk and traditional cheeses in local industry, semi hard cheese could preserve microbial contamination for a few months; *Staphylococcus sciuri*, *Staphylococcus epidermidis*, *Staphylococcus saprophyticus*, *Staphylococcus aureus* was not detected in old cheese. *Staphylococcus agnetis*, *S. chromogenes*, *S. devriesei*, *Staphylococcus equorum*, *S. haemolyticus*, *Staphylococcus lentus*, *S. sciuri*, *Staphylococcus vitulinus* and *S. xylosus*. The probability of finding *S. chromogenes* and *S. agnetis* on the teat and inguinal region increased with age [77].

## 5. Raw and pasteurized milk microbial contamination

In human population raw milk and dairy products are often tangled up in food-borne disease outbreaks; occasionally pasteurized milk may be contaminated and lead to bacteria spoilage of milk and dairy product storage during the dairy processing with a potential health risk for the consumers [5]. The microbiological quality of dairy products reflects good hygienic practices during the dairy milking



process; raw milk contamination may occur in diseased or infected cows with environmental bacteria [1]. In raw milk samples collected from the milk-producing areas tested for *L. monocytogenes* and *Salmonella* spp., the presence and enumeration of mesophilic aerobes and total coliforms is an indicator of *E. coli* contamination and poor microbiological quality of dairy products and causes interference with the native microbiota and other important pathogens [52]. The bacteria acid lactic species (BAL) identified mainly as *Lactococcus lactis* subsp. *lactis* and *Enterococcus faecium* were considered as antagonistic bacteria for the enteric pathogens [36]. The microbial contamination, affects fresh dairy products quality raw milk elaborated. This condition might constitute a potential risk in milk food-borne diseases and public health and dairy food quality [77]. The presence of highly heat-resistant spores of *Bacillus sporothermodurans* in ultrahigh temperature or sterilized milk has emerged as an important item in the dairy industry. The predominant bacterial species isolated at the dairy farm comes from the water, feed-stuffs, and milking equipment, in this aspect *Bacillus licheniformis* and *Bacillus pallidus* acts as entry points for highly heat-resistant spores into the raw milk and the contamination risk level aerobic spore-forming bacteria that could lead to spoilage of milk and dairy products [30]. The milk products are contaminated by *Pseudomonas* spp. in the systems processing milk; it has direct effects on the product shelf life in the dairy industrial plants. The spoilage of milk components has produced by *Pseudomonas fluorescens*, and *Pseudomonas putida* in raw and pasteurized milk provoked by different enzyme systems of the bacteria comes from of protease, lecithinase, and lipase activities. The bacterial contamination source was a derivate from production herd environment and the hygienic farm measures [78, 79]. The hygiene in the production environment in the dairy farm is a very important fact to prevent food-borne diseases and food quality [18]. The incidence of food-borne infections in human population is increasing in the recent years. Oftenly the expose was occurred in private homes and food markets, were prevail the microbial risk contamination in the cases related to the prepared food dairy products, raw milk consumption and eggs, from others food products [80]. Bovine colostrum in human food is considered an excellent source of bioactive proteins, to improve gastrointestinal health and enhance body condition. The consumers are demanding safe and high-quality milk products. There is no influence of herd size and localization in the bacteriological colostrum quality. In milk quality, the animal hygiene and herd health status is considered a goal to warranty the milk as free of *Salmonella* ssp., *S. aureus*, coagulase negative staphylococci, *Streptococcus agalactiae* and streptococci non-agalactiae and coliforms and non-coliforms [36]. In the milk industry, spore-forming bacteria can survive food-processing thermal treatments particularly *Bacillus* and *Clostridium* species to determine the shelf life of a variety of heat-treated milk products, mainly if the level of post-process contamination is low. The management approach of the food production chain, based on raw materials, ingredients and environmental sources, influences the quality of the final product. The strategy on the farm to reduce contamination by foodborne pathogens is to establish hygienic practices on the farm in various components of the milk production chain. Contamination by *Clostridium tyrobutyricum* was consistently found in milk related to farm administration rather than food contamination. Because rottenly *Clostridium disporicum*, identified as an important member of clostridia populations transferred to milk, as a bacteria present in soil, forage, grass silage, maize silage, dry hay. These clostridia may contribute to raw milk contamination by the environmental bacteria as present in soil, forage, grass silage, maize silage, and dry hay [81, 82]. Other virulence factors identified in isolation were assayed: biofilm formation and adhesion to mammalian cells and antibiotic resistance. The genes encoding for virulence factors were present in

*Enterococcus faecalis* more than in *Enterococcus faecalis* and *Enterococcus faecium*. The enterococci were also implicated in vancomycin-resistant strains and the severe multiresistant human nosocomial infection. The recovery of bacteriocinogenic *Enterococcus faecium* isolated with no virulence traits suggests a potential use for biotechnological applications in food animal production [83]. The fecal contamination of the milking equipment is also responsible for the raw milk contamination. Lactobacilli were identified in cows' teats, raw milk, the milking machine, and the milking parlor on one farm. The lactobacilli present in the feces were predominantly *Lactobacillus mucosae* and *Lactobacillus brevis*. The majority of enterococcal isolates from cow feces were identified as *Aerococcus viridans* [36]. *Bacillus cereus* spores are implicated in the herd with environmental microbial contamination; a large number of spores were present in free stalls and bedding material, especially with sawdust beds. A positive relationship was observed between raw milk and the number of spores determined in the feed and feces [84]. *Bacillus licheniformis* and *Bacillus cereus* contamination was present in raw milk, pasteurized milk, and yogurt during dairy processing of dairy milk products as well as other different strains in raw milk and yogurt. The evidence of dairy environmental contamination was attributed to *Bacillus* strains during the technological processing of milk [85]. The enterotoxigenic *Bacillus cereus* and their enterotoxins could be detected in milk products from retail shops. *Bacillus cereus* was isolated from milk products. Enterotoxigenic *B. cereus* hemolytic was identified from milk and milk products and *B. cereus* non-hemolytic enterotoxins just like in milk [86]. The evaluation of the hygienic quality of raw milk is meant to be possible based on the presence of fecal contamination evaluated in raw milk indicated by the coliforms bacteria, and Bifidobacteria species (*Bifidobacterium pseudolongum* subsp. *globosum*), identified isolates compared with bifidobacteria isolated from dung of the cows and the contaminated raw milk samples. The raw milk samples harbored *Bifidobacterium pseudolongum* subsp. *globosum* [87].

## 6. Microbiological quality of milk and dairy products: spoilage bacteria

Milk and dairy product quality is the consequence of all activities developed during the production process, from the farms to the transformation in the dairy industry [88, 89]. Cow's milk contains the nutritional requirements necessary for the growth of the calf, since it is a source rich in lipids, proteins, amino acids, vitamins, and minerals, which added to its high activity of water (aw) and makes it an excellent matrix for the growth of a large number of spoilage microorganisms (Table 2) and pathogens for humans [90, 91]. Not so long ago, it was believed that the milk contained in the mammary gland was sterile and that the microorganisms isolated had their origin from external contamination. Nevertheless, this idea has been questioned due to the development of more sensitive molecular methods which suggests that there is colonization of a wide variety of microorganisms in the healthy mammary gland [92].

The microbial composition of milk is influenced by several different parameters such as, in the case of raw milk, the microorganisms present in the teat canal, on the surface of teat skin, in the surrounding air, and in feed as well as other environmental factors including housing conditions, the quality of the water supply, and equipment hygiene [93–95]. Moreover, the insufficient cold capacity and long storage times can also increase the bacterial count owing to the bacterial growth during milk storage [96]. Therefore, it is not always easy to determine the cause of a high bacterial count in raw milk; there are several parameters that can give an insight of the source of contamination [97].

Kind of defect	Cause	Related microorganism	Reference
Pasteurized, sterilized, and UHT milk			
Precipitation when milk added to hot beverage (bitty cream)	Activity of phospholipases and proteinases and fat destabilization	<i>Bacillus</i> spp.	[130]
Gelation	Thermoresistant proteinases	Psychrotrophic bacteria (Gram-negative and Gram-positive): <i>Pseudomonas</i> spp. ( $10^6$ – $10^8$ cfu mL <sup>-1</sup> )	[131, 132]
Shorter shelf life	Proteolytic and lipolytic activities	<i>Bacillus cereus</i> spp. ( $10^6$ cfu mL <sup>-1</sup> )	[130, 133]
Undesirable flavor: unclean, fruity, bitter, rancid, yeasty	High concentration of free fatty acids due to activity of thermostable lipases; protein hydrolysis due to activity of heat stable proteinases	<i>Pseudomonas fragi</i> <i>P. fluorescens</i>	[130]
Increase of free fatty acids and casein hydrolyses, destabilizing the casein micelles (acid coagulation of milk)	Proteolytic and lipolytic activities	<i>Bacillus</i> spp.	[133]; [134]; [120]
Milk spoiling	Biofilm formation	Consortium of species	[135]
Powder milk			
Shorter shelf life, rancidity, and bitterness	Bacterial proteinases and lipases and increase of free fatty acid	<i>Bacillus</i> spp.	[111]
Cheese			
Destabilization of the natural plasmin system of milk. Affect the quality of cheese, flavor and texture development, and reduce the yield of the curd	Activity of lipases and proteinases remain in curd that ongoing hydrological changes during ripening; cause spoilage of milk and dairy product.	Psychrotrophic spp. ( $>10^3$ cfu mL <sup>-1</sup> )	[114, 136, 137]
Change coagulation time and quality of curd (fragile and less compact)	Higher concentration of free amino acids (bacterial proteinases) which stimulates starter culture which growth.	<i>Bacillus</i> spp. ( $\geq 10^6$ cfu mL <sup>-1</sup> )	[133, 138]
Undesirables flavor: rancid taste in hard cheeses (ripening)	Longer coagulation time: higher concentration of free fatty acids (bacterial lipases) which inhibits starter culture growth		
	Lipases: free fatty acids increase		
Bitterness and off-flavors			[130]
Fermented milks			
Changes of texture and flavor: more firm gel and higher viscosity, more pronounced syneresis		Psychrotrophic	[139]
Lipolytic changes (free fatty acid): atypical flavor as bitter, rancid, unclean, and fruity			[140]

Kind of defect	Cause	Related microorganism	Reference
Creams and butter			
Reduced shelf life Rancidity and off-flavor Fruity, bitterness, soapy	High concentration of lipases and proteinases in milk (cream) High concentration of free fatty acids (C4-C6; C110-C12)	Psychrotrophic <i>Pseudomonas</i> spp. <i>Bacillus</i> spp. <i>Pseudomonas fragi</i> <i>P. fluorescens</i>	[141, 142]

**Table 2.**  
*Principal causes and defects in milk and dairy products caused by spoilage microorganisms.*

Bulk milk analysis is used by dairy industry, veterinarians, and milk producers as an indicator of quality [98]. Through a microbiological profile, it is possible to prevent and modify the possible contamination points. For this reason, the bacterial count of bulk milk is a useful tool for monitoring the environment hygiene, translating high values as negative effects on the quality of the pasteurized milk and milk products, reducing the shelf life and its sensory characteristics [99]. Regarding these indicators, the standard plate count (SPC) in milk represents those bacteria that grow between 30 and 35°C under aerobic conditions and is conformed mainly by bacteria coming from teat skin, feces, milker's hands, equipment, soil, water, etc. [100]. Their importance is given by the fact that they reflect not only the hygienic quality of the raw milk but also the way in which the product was handled. The higher values of SPC indicate raw milk not suitable for consumption, poor handling practices in its elaboration, and an increased risk of the presence of pathogenic microorganisms. Additionally, this parameter reflects the efficiency of cleaning procedures and storage temperatures as well as the hygiene of the udders during milking [100]. With regard to dairy products, this parameter acquires remarkable importance particularly in the elaboration of cheeses, recommending low counts in order to minimize the alteration of the composition of the milk and the final yield obtained [101]. According to the regulations of the European Union, the dairy farms remittent to processing plants of these products must have bacterial counts below 100,000 cfu/mL [102].

In relation to the factors of variation in SPC, there are several studies supporting that the seasonal effect is of great significance in the production of quality milk in terms of hygiene [103]. A work in raw milk from Canada [104] determined that high bacterial counts in summer and spring are related to higher room temperatures that favor the rapid bacterial multiplication. The whole routine of milking, from the pre-sealed and post-sealed to the implementation and maintenance of practices of cleaning and disinfection of dairy equipment, has a great influence in the improvement of milk quality, although for counts below 50,000 cfu/mL, the major factor is hygiene [105].

On the other hand, the rapid cooling of milk and the maintenance of its coldness for prolonged periods stimulate the growth of psychrotrophic bacteria, modifying the native microbiota in favor of Gram-negative ones in approximately more than 90% of the total population [99, 106]. *Psychrotrophic* microorganisms are defined as mesophilic microorganisms which are adapted to grow at refrigeration temperature (7°C or lower), although their optimum temperature of multiplication is higher. They can be widely distributed in the environment: soil, water, and being part of the normal microbiota of animals and man [107]. Numerous psychrotrophic microorganisms have been isolated from raw milk: *Pseudomonas* (*Ps.*), *Enterobacter*, *Flavobacterium*, *Klebsiella*, *Aeromonas*, *Acinetobacter*, *Alcaligenes*, and *Achromobacter* have been reported as the most representative genera [108], while the most frequently isolated species are *Ps. fluorescens*, *Ps. fragi*, *Ps. aeruginosa*, and *Ps. putrefaciens* [109]. In terms of quality, psychrotrophic bacteria have become a problem of special importance for the dairy industry,



being recognized as one of the main agents causing deterioration ending in significant economic losses for the sector [110]. In general, psychrotrophs are capable of producing extracellular or intracellular enzymes (proteases, lipases, and phospholipases), many of which are heat-resistant, which means that they are capable of maintaining their activity after heat treatment (pasteurization or more severe treatment) and also generating big adverse changes in the quality of dairy products [111]. This deterioration in milk quality translates as changes in flavor, undesirable coagulation of proteins, and an increase in the concentration of free fatty acids and amino acids [110].

With regard to other aspects of quality, such as the suitability of milk for the production of dairy products, psychrotrophs have a significant negative effect on yields and in the reduction of their shelf life [112]. When coming from the environment, psychrotrophs are also considered indicators of the hygienic quality of milk [108]. In some countries its count is used as a complement to the bacterial count to determine the quality of the milk and is of special interest when the milk will be subjected to certain technological processes. For example, the regulatory limits for hygienic quality in the Czech Republic are set at  $\leq 100,000$  cfu/mL of bacterial count and  $\leq 50,000$  cfu/mL of psychrotrophs [113]. Furthermore, in the case where milk is used in technological processes, the requirements increase using the limits set by the EU of  $< 30,000$  cfu/mL for bacterial counts and  $< 5000$  cfu/mL for psychrotrophs [102]. In Scotland, an average of 130.000 cfu/mL psychrotrophs in silos of dairy industries from which 70.2% were *Pseudomonas* was found [114].

In terms of food safety, the pasteurizing milk was established as a necessary step for the consumption of fluid milk and other dairy products [115, 116]. In spite of that, this procedure applied in dairy industries for the elimination of pathogenic microorganisms does not completely inactivate all microorganisms, even in the most severe thermal treatments. For instance, some bacteria like thermophilic bacteria resist milk pasteurization. Also, the spores highly resistant to heat can survive the ultrahigh temperature (UHT) process and even to the processes of spray-drying persisting in pasteurized powders [116, 117]. For these reasons, the Food and Drug Administration (FDA) in the USA declared the thermophilic, thermophilic, psychrotrophic, and spore-forming bacteria as the microorganisms with the highest risk of spoilage in dairy products [118]. The thermophilic count is used as an indicator of sanitization of equipment in the industry and establishments [99], being the ideal ranges those between 100 and 200 cfu/mL [119]. Of this group, *Bacillus cereus* is the most commonly found in milk and dairy products [120], and their spores are characterized by having the ability to survive the thermal processing used in the industry [121]. In addition, some species can multiply at refrigeration temperatures, which is why it is also considered a psychrotrophic microorganism [122]. *Bacillus* spp. produces extracellular proteases and lipases and phospholipases (lecithinase) resistant to thermal treatments, comparable with the enzymes produced by *Pseudomonas* [123]. The combination of these characteristics in a microbial species indicates a great deteriorating potential [124]. Raw milk contamination by spores of *B. cereus* has been reported as the main cause of the presence of these groups of microorganisms in processed milks [125]. The spores of thermophilic microorganisms can be found in processed products, such as pasteurized milk and stored cream, decreasing their shelf life [91, 126]. To ensure the shelf life of the pasteurized milk, it is necessary to comply with a maximum spore limit of *B. cereus* of 3 log spores/mL [127]. In dehydrated products, they have a main importance because these products are more prone to thermophilic deterioration because of having a long useful life [116].

Therefore, psychrotrophs and thermophilics are of great importance in the quality of the milk that will be industrialized, mainly due to its effects on the composition. The lipolytic and proteolytic enzymes that they produce cause deterioration during the storage of milk and dairy products [107]. Moreover, studies suggest

that these proteases found in raw milk are produced by psychrotrophic bacteria, especially of the genus *Pseudomonas* [128]. In this regard, a study conducted in fresh milk observed a production of extracellular proteases of psychrotrophs, which cause an increase in plasmin activity, hydrolyzing casein and decreasing its levels (count above  $10^7$  cfu/mL); this increase in plasmin activity could affect the quality of cheeses or other dairy products [129]. In the case of thermodurics, it has been reported that *B. cereus* can also release proteases that degrade casein by damaging the milk. The  $\kappa$ -casein is the protein fraction which is more affected by hydrolysis; after 7 days of storage at 20°C, all  $\kappa$ -casein is converted to para- $\kappa$ -casein, while  $\beta$ -casein is reduced by 70%. Furthermore, as part of the deterioration caused by this microorganism in milk, it has been observed that it releases peptides of low molecular weight causing undesirable flavors [120].

In summary, milk and milk products provide favorable conditions for the growth of various microorganisms. These include groups capable of growing at refrigeration temperatures, withstanding heat treatments and producing heat-resistant enzymes, which are responsible for the deterioration and reduction of the shelf life of milk and by-products. The effectiveness in the control of these microorganisms is a critical challenge for the dairy industry, and its relevance has been discussed in this chapter.

## 7. Conclusions

The paper remarks the importance among the milk production and food safety, closely related in the assurance of the milk quality and the prevention of milk spoilage. The dairy industry management programs as for food safety, the milk quality and the dairy products. Preventing the microbial and chemical contamination. The food-borne diseases in public health programs are a priority in the surveillance of milk food-borne diseases by the monitoring of food-borne pathogens and the microbial contamination in milk products. Actually dairy farms are compromised to reduce the milk contamination source from udder and the dairy cow herd health status and the production environment, by hygiene practices in the cow herd management and good milk conserving in the raw milk bulk tank. The food hygiene protocols are fundament for to reduce the microbial contamination of the raw milk and pasteurized milk, regarding the health risk by the microbial pathogens in the food borne diseases and bacterial spoilage, source of deteriorating dairy products and milk. The microbial quality of foods is required for the traceability in dairy products industry. Consumers education programs and practices of good handling of foods, could be reduce the exposure to food borne pathogens and the consumption of unsafe food products. The traceability of milk and dairy products, from the production-distribution chain food and the consumption is a good policy for to the assurance the quality and to reduce the public health risks.

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## Conflict of interest

The authors declare that there is no conflict of interest in the participation and collaboration in the elaboration of this work and its divulgation.

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