

# A review on Determination of Bacterial Contamination in Raw Milk.

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**Abstract:**-Milk is an important source of nutrients to human and animals, but due to its high water activity and nutritional value it serves as an excellent medium for growth of a number of microorganisms under suitable conditions. The present inferences of review were conducted that, to identify microbial quality of raw cow milk and determines that milk-borne bacterial infections or contamination from natural sources. The demand of consumers for safe and high-quality milk has placed a significant responsibility on dairy producers, retailers, market safe milk, and milk products. Researches showed that, The number and types of microorganisms in milk immediately after milking are affected from different factors which include poor hygienic condition of the milking environment, absence of cooling system, poor sanitary condition of the milk containers, poor udder and teats cleaning practice, failure of washing and drying cow's udder, the absence of usage of detergent for cleaning vending environments, and the poor personal hygiene of the milkers. Additionally, condition of storage, manner of transport, use of plastic containers for milk collection, mixing of milk obtained from different cows and the presence of further contamination at the milk selling sites.

Key Words:-Microorganisms, Raw milk, Contamination

## Introduction

Milk is an important source of nutrients to human and animal diet. As a result, it's for the primary and the best food for the offspring of mammals as almost nearly entire food (Pandey and Voskuil, 2011). It's mainly made from water, which a huge range of nutrients consisting of vitamins, proteins, fat, and carbohydrates are suspended. Those excessive nutritional contents, the production, and processing procedures in commercial milk production render it susceptible to contamination via a host of pathogenic microbes that might cause diseases in people. Milk has a complex biochemical composition and its excessive water activity and nutritional value serves as an excellent medium for growth and multiplication of many forms of microorganisms while appropriate situations exist (Parekh and Subhash, 2008). Consequently, milk is known to be an efficient vehicle for the transmission of disease-causing agents to peoples (Garedew, *et. al.*, 2012). In spite of fact that, the conditions for contamination of raw milk were distinctive critical points are due to less hygienic practices in pre-milking, udder practice, sub-most beneficial hygiene of milk handlers and bad sanitation practices associated with milking and storage equipment's (Garedew *et al.*, 2012, Reta, *et. al.*, 2016).

Milk is a complex biological fluid and broadly consumed as nutrient food and it's an excellent medium for the growth of microorganism's balanced diet. Due to the specific production, it's considerably impossible to keep away from contamination of milk with microorganisms. Contemporary, the microbial content material of milk is primarily a major feature for determining its high-quality. Bacterial contamination of raw milk can originate from distinct or different sources: air, milking system, feed, soil and skin or hair of the animals or utensils (Janstova and Drackova, 2006). In addition to this the number and forms of microorganisms in milk immediately after milking are suffering from factors which include animal health and equipment cleanliness, season, and from the feed. Pasteurization can't assure the absence of microorganisms, while they may be found in massive numbers in raw milk or because of post-pasteurization contamination (Santana, *et. al.*, 2004; Uma Maheswari and Sabitha, 2015).

The demand of consumers for safe and high-quality milk has placed a significant responsibility on dairy producers, sellers, market safe milk, and milk products (Adesiyun, *et. al.*, 1995; Mennane *et al.* 2007). Milk and milk products have an important role in feeding the rural and urban population of the world owing to its high nutritional value. It is produced daily, sold for cash or readily processed. It is a cash crop in the milk shed areas permits families to shop for other foodstuffs and significantly contributing to the family food security (Abebe *et al.*, 2012; Reta, *et. al.*, 2016).

Lack of refrigeration facilities at the farm and household level in developing countries of tropical regions with excessive ambient temperature means that raw milk will easily be spoiled during storage and transportation (Godefay and Molla, 2000). Milk and milk products can carry toxic metabolites of different pathogenic organisms developing in it. Ingestion of such products contaminated with those metabolites causes food poisoning or toxicity for consumers. On the other hand, the ingestion of viable pathogenic bacteria along with the food product leads to foodborne infection (Aneja, *et. al.*, 2002).

According to author Javaid *et al.* (2009), the overall health and welfare of people depends in large part on meeting basic tremendous of nutritional needs. Milk and fermented milk products have formed an important part of each daily nutrition and the type of products produced from milk has increased dramatically through the years, as modern food processing technology has advanced. Also, an increase in the global population coupled with the increasing demands for milk as an economic food and as an industrial raw food product has necessitated an increase in production with the aid of dairy farms.

The assessment of microbial load at various levels of processing may function a beneficial tool for quality evaluation and improvement which could be bring about longer shelf life that's an applicable market requirement. The possessing of fresh milk at

an elevated temperature together with unhygienic practices in the milking process might also result in microbiologically inferior quality. However, common place practices for small-scale farmers who produce fresh milk and sell it to local consumers or milk collection centers (Chye, *et. al.*, 2004).

The safety of dairy products with respect to food-borne diseases is a great challenge around the world. This is especially true in developing countries where the production of milk and numerous milk products takes place under unsanitary conditions and poor production practices (Mogessie, 1990). The microbial content material of milk is a primary feature in determining its quality. The paradox of milk shows the hygienic level exercised during milk production and cleanliness of the milking utensils, condition of storage, manner of transport, and cleanliness of the udder of the individual animal (Coorevits, *et. al.*, 2008). The number and types of micro-organisms in milk immediately after milking are affected from factors which include lack of knowledge about clean milk production, use of unclean milking gadget or equipment and absence of potable water for cleansing purposes contributing to the poor hygienic high-quality of raw milk (Bekele and Bayileyegn, 2000). Milk from a healthful udder contains few microorganisms but it alternatives up many microorganisms from the time it leaves the teat of the cow until it's far used for consumption or further processing. These micro-organisms are indicators of both the manner of

handling milk from milking until consumption and the quality of the milk (Abera and Angaw, 2015).

The tackle of microbial contamination in milk can be considered as a key point in the quality and safety control of dairy products. Many microorganisms that contaminate milk are pathogenic, posing as hazards for humans. Other microbial groups are relevant to indicate the hygienic conditions of milk production and storage, allowing producers, industries, and food inspectors to assess the production conditions and predict the spoilage and usage of milk for dairy products. Finally, other microbial groups can present positive and desirable features, leading their usage as probiotics or beneficial bacteria and also as starter cultures for fermented foods. As (P. Kavitha.,*et.al.*,2016) reported Probiotic microorganisms are found in many food products, especially in the fermented foods. Therefore, the probiotic lactic acid bacteria can be isolated from the fermented milk products. All these microbial groups can contaminate milk at different steps of production, highlighting their relevance for the dairy chain (Luana, *et. al.*, 2019).

Milk is a good indicator host for pathogenic microorganisms due to that exerts high water content, nearly neutral pH, and variety of available essential nutrients. However, bacteria, yeasts, and molds are the common contaminants of milk with their rapid growth, particularly at high ambient temperature can cause marked deterioration in the quality of the milk and dairy products (FAO, 1989). The microbial load and bacterial contamination found in raw milk can influenced from different sources such as air, milking equipment, storage, feed, soil, feces, sick animals and ambient temperature (Torkar and Teger, 2008). Daily production and eventual marketing of milk require special consideration to ensure its delivery to the market in hygienic and acceptable condition (Kivari, *et. al.*, 2006 and Gemechu, *et. al.*, 2014).

Though in less developed areas especially in hot tropics, the production of products of safe and high quality is important, the prevailing situation is far from the ideal condition (DeGraaf *et al.*, 1997). Poor hygiene, practiced by handlers of milk and milk products, may lead to the introduction of pathogenic micro-organisms into the products. Since they do not undergo further processing before consumption, these foods may pose risk to the consumers. Since, the provision of milk and milk products of good hygienic quality is desirable from the consumer health point of view (Zelalem, 2010). In addition, there is no formal quality control system in place to monitor and control the quality of milk produced and sold in the town (Abera and Angaw, 2015). The main objective of this review to detect and determine bacterial contamination from different source of milk and identifying microorganisms present in it.

## 2. Definition and Composition of Milk

Milk is a yellowish-white non-transparent liquid secreted by using the mammary glands of all mammals. It is the primary source of nutrition and sole food for offspring of mammals before they are able to eat and digest different forms of food (Pandey and Voskuil, 2011). It incorporates in a balanced form of all the important and digestible factors for building and retaining the human and animal body (Pandey and Voskuil, 2011). The main composition of milk is water (87 –88%); the remaining component is total milk solids which include carbohydrates, fat, proteins and ash or minerals. In addition to pathogenic microbial flora, cattle raw milk also encompass important inorganic mineral elements in trace amounts like P, Ca, K, Mg, Na, Cl and trace elements including Cu, Fe, Cr, Cd and Ni. These minerals are required in plants and animals for completion of their life cycles and enzymatic reactions (Hamid Iqbal,*et.al.*,2016).The compositions of raw milk differ by species, but significant amounts of saturated fats, calcium and protein as well as vitamin C is there in milk. Cow's milk is slightly acidic with pH ranging from 6.4 to 6.8 (Ayub M, *et.al.*,2007). Every so often the composition may even trade from each day, relying on feeding and climate, but additionally for the duration of milking the first milk differs from the final milk drops (Pandey and Voskuil, 2011). Moreover, milk is an excellent source of protein, vitamins, minerals which includes calcium and phosphorus. Fresh milk has a pleasantly smooth and sweet taste and contains hardly any smell (Happy Brown Kanyeka, 2014). However, the high content of a variety of nutrients and water in milk as well as its near neutral pH make milk a good growth medium for some microorganisms (Quigley, *et. al.*, 2013).

The bacterial infection of milk from affected cows render it undeserving for human consumption and offer a mechanism of spread of diseases like tuberculosis, sore-throat, Q-fever, Brucellosis, Leptospirosis, and many others zoonotic importance (Sharif, *et. al.*, 2009). More than 135 different kind pathogenic microorganisms were recognized as causative agents of bovine mastitis. Microorganism, fungi, and yeasts may also all play a function but of those, bacteria have via some distance the most important

element. Staphylococci, streptococci, and contributors of the Enterobacteriaceae are responsible for most people of infections (Quinn, *et. al.*, 1994).

E.coli and Staphylococcus were most contaminations: pathogenic bacteria could be important factors of gastrointestinal infection including food poisoning and foodborne illness. Raw and unpasteurized milk obtained from cow and buffalo by hand milking may contain bacteria from cow's udders, flies, manure, etc. indigenous sweet based products like khoa, gulab jamun, rasgulla are highly susceptible to a variety of microorganism because of high nutritive value and complex chemical composition (Soomro, 2003; Kumar and Prasad, 2010). Raw milk represents an ideal growth medium for microorganism (Haridy, 1992). Among all microorganisms, E.coli is frequently contaminating organism and is a reliable indicator of fecal pollution (Kumar and Prasad, 2010).

### 3. Microbiology of Raw Milk

The microorganisms determined in raw milk may additionally come from numerous sources: the milk as it's far excreted from the teat, the environment (water, soil, and so on.), milking equipment, milk strains, farm bulk tank, the tank (transfer) truck, strains and pumps on the processing plant, and the raw storage silo tanks at the processing plant. Cows with mastitis can shed excessive numbers of bacteria and somatic cells. An elevated somatic cell count number (SCC) can adversely impact milk quality. According to the *International Commission on Microbiological Specifications for Food* (ICMSF; 2000), "the normal flora of the udder includes streptococci, staphylococci, and micrococci (normally >50%) followed by *Corynebacterium* spp., *Escherichia coli*, and others. In reflecting on the various microorganisms in raw milk, the ones with which we are most concerned are those bacteria that can survive the pasteurization process, that is, the thermotolerant, and those bacteria that can grow, although slowly, at refrigeration temperatures, that is, the psychrotrophs (White, 2011).

Hence gram staining of bacteria segregates bacteria into two categories based on cell wall composition. The cell wall of Gram-positive bacteria consists of a cytoplasmic membrane, many polymeric layers of peptidoglycan connected by amino acid bridges, and a variable outer layer called the capsule (Jawets, *et. al.*, 1987). Gram-negative bacteria possess a bilayer outer membrane, a thin peptidoglycan layer, and a bilayer plasma membrane (Beveridge *et. al.*, 1991). Although, Gram-Negative Pathogens including (*Escherichia coli*, *Salmonella*, *Y. enterocolitica*, *Campylobacter jejuni*, *Aeromonas hydrophila* (another psychotropic pathogen), on another hand those Gram-positive Pathogens are *Bacillus cereus*, *Bacillus anthracis*, and *Clostridium perfringens* are predominant (Donnelly, 1990).

The fundamental contaminants of processed dairy products have their origins in the raw milk delivered while identifying that there are numerous pathogens that is probably linked to raw milk, a few specific foodborne ailments attributed to dairy products are as a result of *Salmonella*, *Listeria monocytogenes*, and *Yersinia enterocolitica*. A crucial issue with *Listeria* and *Yersinia* is that they are both psychotropic pathogens. The main outbreak have been caused by those microorganisms, resulting in many who they are each and even deaths. despite the fact that the milk is pasteurized before consumption, the capacity for cross-contamination inside the dairy processing plant stresses the significance of the raw milk being of high quality, to minimize those occurrences and the excessive numbers of pathogens. a number of the pathogens related to raw milk (Donnelly, 1990). The meager research scholar state are psychrotrophs are introduced into the milk, there is evidence that typically within two to three days of the transfer of the milk from transport tankers, the microform of the milk is dominated by way of psychrotrophs, while the thermotolerant microform does not increase and modifications little in composition (Critine cerva 2016)

Microorganisms found in milk favorable situations for their multiplication. The raw milk microbial could be very numerous and can be composed of spoilage, pathogenic microorganisms or even microorganism with high technological significance (Montel, *et. al.*, 2014; Perin, *et. al.*, 2017). Moreover, microbial species is without delay inspired by using the sanitary situations of the dairy herd, environmental hygiene, milking equipment used, and factors related to the raw milk handling, storage, cooling, and processing (Murphy *et. al.*, 2016). Milk remains free from microorganisms within the udder, however for the duration of milking, the contamination is once the product is exposed to air remain in contact with equipment surfaces. in view that environmental (e.g., water quality, hygiene of milking vicinity) and operational conditions (poorly sanitized equipment, poor handling, storage temperature above recommended) are not managed, milk is infected and the bacterial population find optimal conditions for development and multiplication, changing its physical and chemical characteristics and can become a source of contamination by microorganisms of importance in public health (Raza and Kim, 2018).

#### 3.1. Mesophilic and Psychrotrophic Bacteria

Mesophilic microorganisms grow higher in mild temperatures with most optimal increase temperatures starting from 25°C to 40°C; which might be most desirable situations also for each spoilage and pathogenic microorganism. The main genera of mesophilic bacteria that may be found in milk are *Micrococcus*, *Staphylococcus*, *Enterococcus*, *Escherichia*, *Serratia*, *Acinetobacter*, *Flavobacterium*, *Pseudomonas*, *Mycobacterium*, *Bacillus*, *Lactococcus*, *Lactobacillus*, among others (Jay, 2012). Mesophilic bacteria are considered the universal hygiene indicators and their presence in milk are unavoidable since most of these genera are present within the animal's udder, milkers' hands, device surfaces, water, and air. for this reason, mesophilic counts are directly inspired via the conditions that milk is submitted after milking (Jay, 2012; Angelidis, 2014). Within a common sense, milk with counts of mesophilic bacteria higher than 5.0 log CFU/ mL indicates poor hygienic quality during milking and production, while counts lower than 3.0 log CFU/mL indicate good production practices. In general, good production practices, adequate cooling, and refrigeration are effective measures to control mesophilic bacteria in milk (Luana, *et. al.*, 2019).



Milk cooling should be performed immediately after milking and the temperature have to faster attain 4°C to control the multiplication of the microbiota present until thermic treatment. The raw milk refrigeration immediately after milking constitutes the primary tool of conservation of the product. but, this practice should not be carried out alone, due to the fact that, many microorganisms that contaminate raw milk within the preliminary stages of production have the potential to multiply even when submitted to refrigeration temperatures, specifically within the marginal temperatures, above ideal and below 10°C (Perin, *et. al.*, 2012).

The presence of psychrotrophs in milk is of relevance due to their spoilage activity and consequently one of the principal limitations of the shelf-lifestyles of milk (Cousin, 1982). Under suitable conditions of milking and preservation, this group generally represents 10% of the microbial of raw milk, but when milk is received under poor hygiene situations, it can represent approximately seventy five% of the full microbial of raw milk (Cousin, 1982).Pseudomonas is considered a classic psychrotrophic, due to its intense metabolic activity in the range of temperature 47°C (Jay,2012).Nevertheless, Bacillus predominates in marginal cooling temperatures, from 8°C to 10°C. Even if an appropriate temperature for the multiplication of psychrotrophs is among 4°C and 10°C, in raw milk with preliminary mesophilic counts of 5.0 log CFU/mL (which might be considered the most restrict for this group) and stored at temperatures of 4°C or much less, psychrotrophic counts boom substantially after 24 h (Scatamburlo, *et. al.*, 2015; Yamazi, *et. al.*, 2013). Consequently, hygienic conditions of milking and cooling at 4°C or less are ways to prevent undesirable changes resulting from the presence of this group in raw milk (Luana , *et. al.*, 2019).

#### 4. Detection of microbial quality analysis of raw milk

##### 4.1. Bacterial count methods

Currently, a variety of microbiological count methods, including the total bacterial count (TBC) and coliform count (CC),and most important for available monitoring of the hygienic quality of raw milk (Jayarao,*et.al.*, 2004).Among from those strategies TBC is the most common method used for evaluating the hygienic quality of raw milk, which estimates the total number of bacteria present in milk (Ruegg& Reinemann,2002).The Colony Count(CC) measures the number of coliform bacteria in milk primarily originating from cow's environment. The elevation of CC in milk is an indicator of poor sanitary practices in farm area (Reinemann,*et.al.*, 2000 and Ye Wint Naing, *et.al.*,2018). In addition analysis for microbial quality of raw milk involved count of aerobic mesophilic bacteria, total coliform, yeast and moulds and common milk-borne pathogens namely Shiga toxin producing Escherichia coli (STEC), Salmonella spp., Staphylococcus aureus , Streptococci , Brucella spp. and Mycobacteria (Mohamed *et al.*, J Food Microbiol Saf Hyg 2017).

Microbiological evaluation of raw milk would be essential to find the degree of contamination with the expressions and enumeration of indicator organisms (Chatterjee S. N.*et.al.*,2006).Total bacterial count (TBC) has a prime factor in determining its hygienic quality (Khan *et al* 2008). It shows udder infection and the extent of cleanliness of udders, milkers and milking equipment during milk production. It also suggests condition under which milk is stored and transported (Karikari *et al* 1998). Raw milk with excessive microbial load has poor keeping quality and products manufactured from it are of inferior quality and have a reduced shelf life (Hayes, *et. al.*, *et al* 2011). Microorganisms are almost always present in raw milk but with suitable strategies of production their number can be kept low (Boor, *et. al.*, 1998). The presence of the microorganisms in milk and milk products is an indication of unsanitary production and wrong managing of either milk or milk utensils and is normally associated with fecal contamination (Boor *et al* 1998 & Kagkli. *et. al.*, 2006) displayed that besides to the faecal contamination, different factors including milking wet udders, insufficient cooling of milk and udder contamination are the main sources of coliforms in milk. Escherichia coli (*E. coli*) are the most usually isolated coliform from milk within the clinical laboratory (Ahmed and Salam 1991).

##### 4.2. Biochemical isolation of raw milk

Besides to the gram staining, various biochemical tests were also performed for identification of bacteria (Cappuccino and Sherman, 1996). These tests included oxidase test (OX), catalase test (CT), indole production test (Cowans and Steel, 1993), methyl red test (MR), carbohydrate fermentation test and motility test (Mot) were qualitatively estimated (Kannan, 1996 & Garbutt, 1997). On the basis of these tests bacteria were clearly identified (Chatterjee S. N.*et.al.*,2006 & Abbas.M.,*et.al.*,2013).Milk samples from both clinical and sub clinical quarters were bacteriologically examined according to the procedures employed by (Quinn *et al.* 1999). Prior to further biochemical tests, the isolated bacteria on blood agar were sub cultured into nutrient agar. Each culture was subjected to gram staining to determine their shape, and gram reaction (Melesse Etifu, 2012)

Therefore in order to protect the public health, microbiological assessments have an important role to play in the dairy industry. This also reduce economic losses by the early detection of insufficient processing, packaging or refrigeration (Sourav.K,*et.al.*,2014).

#### 5. Prevention and Control of Microbial Contamination in Milk

Prevention and control of the microbial quality of milk is through the elimination of organisms from human carriers by general improvements in water supplies, public health education, non-public and environmental hygiene. The raw milk was early properly for boiling or pasteurization than processing and consumption. Pathogenic organisms from the lactating animals can be controlled through improvements in animal husbandry management and renovation of proper animal practices, and people from the environments and equipment may be prevented with the aid of adhering to general hygienic practices and protection of environmental sanitation (Mosalagae, *et. al.*, 2011; Kanyeke, 2014). Generally, microbial contamination in milk can be minimized

through obedience to effective good hygienic practices at farm level and in order to protect the public against milk-borne infections, it is important to screen milk which is informally taken to the market for demand consumption (Mosalagae, *et. al.*, 2011; Kanyeka, 2014).

## Conclusions

Milk is an important source of nutrients to human and animal diet. As a result, it's the primary and the best food for the offspring of mammals as almost nearly entire food. The existence of massive range of microbial counts and the incidence of pathogens in all prospect to affect the quality and safety of raw milk including products provided from it. The presence of pathogenic microorganisms which include E.coli, salmonella and Listeria spp.in raw milk is of public health anxiety since drinking raw milk nevertheless taken into consideration in most of rural population of undeveloped countries. This documented as accurately, adersed the cause of contaminations that, poor hygienic milking environment, absence of cooling system, poor sanitary condition of the milk containers, poor udder and teats cleaning practice, lack of washing and drying cow's udder, absence of usage of detergent for cleaning vending environments, and the poor personal hygiene of the milkers. Additionally, use of plastic containers for milking and milk collection or transportation and mixing of milk obtained from different cows and the presence of further contamination at the milk selling sites. Therefore to advocate the problems training and guidance should be given to farms, owners including their workers responsible for milking; emphasize the need for the hygienic practices at the farmers while information on health hazard associated with contaminated raw milk should be extended to the public.

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