



Consumers' behavior in quantitative microbial risk assessment for pathogens in raw milk: Incorporation of the likelihood of consumption as a function of storage time and temperature

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ABSTRACT

Foodborne disease as a result of raw milk consumption is an increasing concern in Western countries. Quantitative microbial risk assessment models have been used to estimate the risk of illness due to different pathogens in raw milk. In these models, the duration and temperature of storage before consumption have a critical influence in the final outcome of the simulations and are usually described and modeled as independent distributions in the consumer phase module. We hypothesize that this assumption can result in the computation, during simulations, of extreme scenarios that ultimately lead to an overestimation of the risk. In this study, a sensorial analysis was conducted to replicate consumers' behavior. The results of the analysis were used to establish, by means of a logistic model, the relationship between time–temperature combinations and the probability that a serving of raw milk is actually consumed. To assess our hypothesis, 2 recently published quantitative microbial risk assessment models quantifying the risks of listeriosis and salmonellosis related to the consumption of raw milk were implemented. First, the default settings described in the publications were kept; second, the likelihood of consumption as a function of the length and temperature of storage was included. When results were compared, the density of computed extreme scenarios decreased significantly in the modified model; consequently, the probability of illness and the expected number of cases per year also decreased. Reductions of 11.6 and 12.7% in the proportion of computed scenarios in which a contaminated milk serving was consumed were observed for the first and the second study, respectively. Our results confirm that overlooking the time–temperature

dependency may yield to an important overestimation of the risk. Furthermore, we provide estimates of this dependency that could easily be implemented in future quantitative microbial risk assessment models of raw milk pathogens.

Key words: raw milk, quantitative microbial risk assessment, consumer behavior, milk spoilage

INTRODUCTION

Probabilistic modeling is becoming established as one of the main tools to inform risk management decisions with regard to foodborne hazards. Quantitative microbial risk assessment models (QMRA) are increasingly applied to scenarios involving established and emerging food safety hazards as risk analysis becomes standard practice to manage food safety and ensure that regulatory decisions about foods are science based and transparent (FAO, 2006; WHO/FAO, 2010).

One of the most significant examples from the public health perspective in recent years has been the use of QMRA to estimate risks associated with the consumption of unpasteurized milk. Growing interest in raw milk consumption by some groups of consumers and an increasing number of foodborne incidents in which raw milk has been identified as the source have lead agencies such as the UK Food Standards Agency, the European Food Safety Authority, or the US Centers for Disease Control to conduct consultations and issue scientific opinions on the risk posed by milk-borne hazards (CDC, 2014; FSA, 2014; EFSA, 2015).

The public health risk related to consumption of raw milk is a particularly relevant (and debated) topic. Raw milk can contain human pathogens, which can be inactivated by appropriate heat treatment (pasteurization or sterilization). However, the perception of raw milk as a more natural product has led to several consumers opting for raw as opposed to heat-treated milk. In light of this trend, models have been developed in recent

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years to assess probability of exposure or infection by pathogens such as *Salmonella*, *Listeria monocytogenes*, *Campylobacter jejuni*, *Escherichia coli* O157, or *Staphylococcus aureus* as a result of raw milk consumption (Heidinger et al., 2009; Latorre et al., 2011; Giacometti et al., 2012; Giacometti et al., 2015).

Quantitative microbial risk assessment models aimed at assessing the risk from farm to table include a consumer phase module, a stage of the model that occurs at household level, where the food is no longer controlled by professionals and where control of storage conditions or application of sufficient heat treatments cannot be enforced by legislation (Nauta and Christensen, 2011). In QMRA related to unpasteurized or pasteurized (Koutsoumanis et al., 2010) milk, the time and temperature of storage in the consumer phase modules are usually described and modeled as independent distributions. Time and temperature are the most important parameters that regulate microbial growth in milk and are regularly identified in sensitivity analysis as the factors with greatest effect on the model output (Koutsoumanis et al., 2010; Latorre et al., 2011).

When both, storage time and temperature, are modeled as independent probability distributions (most often Triangular or Pert), in some instances during simulations, values from the tails of the distributions are sampled together yielding scenarios with high bacteria concentration at the time of consumption. An implicit assumption underlying the cited models is that 100% of the computed scenarios will result in milk being consumed, whatever the time–temperature combination is. However, in reality some time–temperature combinations are unlikely to result in milk being consumed as it would be perceived by the consumer as unsuitable (raw milk stored at high temperature for extended periods might be spoiled and thus not actually consumed). Therefore, given that in microbial dose–response models the probability of illness is directly dependent on the number of bacteria ingested per serving (i.e., each bacteria has the same probability to generate infection), the amount of simulated scenarios under extreme conditions may have a significant effect on the final output.

This limitation was already highlighted by Latorre et al. (2011), who noted that some correlation between these variables may exist and that without any restriction, the model cannot take into account that some extreme scenarios may not occur or end with milk not being consumed. However, to our knowledge, this limitation and the effect that this assumption may have on model output have never been formally assessed.

Following these considerations, the objectives of this work were to (1) model the dependencies between time

and temperature to express the likelihood for a raw milk serving to be actually consumed for any computed storage time–temperature combination and (2) assess the extent to which this dependency would affect the output of a QMRA model.

To this end, results of a simplified sensorial analysis on raw milk stored for 5 d at different temperatures were used to estimate the probability that at given time–temperature combinations, the milk is spoiled, recognized as such, and thus not consumed. The potential effect of the estimated time–temperature relationship on model output was then evaluated by its inclusion in 2 recently published QMRA of raw milk consumption and comparison of published results with those of the modified model.

MATERIALS AND METHODS

Raw Milk Sample Collection for Sensorial Analysis

A total of 1.5 L of raw milk was collected from 30 automatic vending machines in Lombardy by the public veterinary services, univocally coded, placed in cold boxes at $5^{\circ}\text{C} \pm 3$, and taken to the laboratory within 30 min. Upon arrival, 5 aliquots of 200 mL were obtained from each sample and kept in different isothermal conditions at 3, 5, 8, 12, and 16°C for 5 d (temperatures were chosen to reflect the range of temperatures at which the domestic refrigerators can be expected to operate).

A total of 500 mL from each sample was used to test the samples for pH, SCC, lactic acid bacteria, total mesophilic flora, *Enterobacteriaceae*, and the major pathogens to ensure operator safety. An instrument with automatic temperature compensation (Hanna instrument HI9321, Hanna Instruments Inc., Woonsocket, RI) was used for pH measurement. The SCC was determined by an Optofluorimetric accredited internal method MP02/063 (Fossomatic, Foss Electric, Hillerød, Denmark). The ISO standards ISO4833-2, ISO21528-2, and ISO16649-2 (ISO, 2001, 2004, 2013) were used for surface plate enumeration of total mesophilic flora, *Enterobacteriaceae*, and *E. coli*, and the standards AFNOR BRD 07/10 and AFNOR BRD 07/06 (AFNOR, 2008, 2009) were used for PCR real-time detection of *L. monocytogenes* and *Salmonella*. Enumeration of lactic acid bacteria was performed by the accredited internal method MP01/048 (decimal dilution and plating in MRSA agar plate incubated under microaerophilic condition at $37 \pm 2^{\circ}\text{C}$ for 72 ± 2 h and decimal dilution and plating on M17 agar plate at $37 \pm 2^{\circ}\text{C}$ for 48 ± 2 h for enumeration of mesophilic lactic flora and lactococci, respectively). The accredited internal method

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