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Temperature status of domestic refrigerators and its effect on the risk of listeriosis from ready-to-eat (RTE) cooked meat products

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ABSTRACT

Inadequate domestic refrigeration is frequently cited as a factor that contributes to foodborne poisoning and infection, and consumer behaviour in this regard can vary largely. This study provides insight into the temperature profiles of domestic refrigerators in the Netherlands and the impact on the number of listeriosis cases related to ready-to-eat (RTE) cooked meat products. A survey was conducted among Dutch consumers (n = 1020) to assess their knowledge and behaviour related to refrigerators. Out of these participants, 534 measured their refrigerator's temperature, revealing an average temperature of 5.7 °C (standard deviation (SD) of 2.2 °C) with a maximum of 17 °C. Elderly people (65 years and older) had refrigerators with temperatures that were on average $0.6~^{\circ}\text{C}$ higher than those of younger people (35 years or younger). The 24-hour temperature profiles of an additional set of actively surveyed refrigerators (n = 50) showed that the temperature measured on the upper shelf was significantly higher (mean 7.7 °C, SD 2.7 °C) than the temperature measured on the bottom shelf (5.7 °C, SD 2.1 °C). Quantitative Microbiological Risk Assessment (QMRA) predicted that the primary factors contributing to the risk of listeriosis were the initial concentration and the time and temperature during household storage. Scenario analysis revealed that storing opened RTE cooked meat products at home for either <7 days or at temperatures <7 °C resulted in a significant reduction of over 80 % in predicted illness cases. Among all illness cases, the elderly represented nearly 90 %. When assessing the impact of the disease in terms of Years of Life Lost (YLL), the contribution of the elderly was 59 %. Targeted communication, particularly directed towards the elderly, on the importance of storing RTE cooked meat products at the recommended temperature on the bottom or middle shelf as well as consuming within two to three days after opening, holds the potential to significantly reduce the number of cases.

1. Introduction

Insufficient food temperature control is one of the most common causes of foodborne illness according to the official standards of the Codex Alimentarius Commission (2003). The World Health Organization (WHO) also states, in its five keys to safer food, that all cooked and perishable food should be quickly refrigerated below 5 °C (WHO, 2006). Recommended refrigerator temperatures vary throughout the world, but the maximum recommended temperatures are mostly below 7 °C (FDA, 2022; FSA, 2022; Terpstra et al., 2005), with many countries recommending below 5 °C. In the Netherlands, it is recommended that the temperature of the domestic refrigerator should be 4 °C (Van der Vossen and Van Dooren, 2012).

The safety of many foods is dependent on ensuring the cold chain

until the time of consumption. One of the weakest links is the consumer part of the chain as the temperatures of domestic refrigerators may be above the recommended temperature. A review of 23 available survey studies in mostly European countries from 1991 to 2016 showed mean, minimum and maximum temperatures ranging from 5 to 8.1 °C, -7.9 to 3.8 °C and 11.4 to 20.7 °C, respectively (EFSA BIOHAZ Panel, 2018). The temperature in domestic refrigerators varies among consumers and over time, but can vary also within the refrigerators. Studies reporting simultaneous recordings of temperature at different positions in the refrigerators using data loggers (temperature measurement every 1 to 10 min) showed lower mean temperature values on the bottom shelves than top shelves (Jofré et al., 2019; Koutsoumanis et al., 2010). Available data indicate that the domestic refrigerators mean temperature, as well as the fluctuation within the refrigerator, is affected by their type

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and age (Dumitrascu et al., 2020; EFSA BIOHAZ Panel, 2018; George

Several surveys have shown that few householders know the actual temperature of their refrigerators. A fridge thermometer is not often used by consumers to check the temperature of the refrigerator (James et al., 2017; Janjić et al., 2016; George et al., 2010). More often, personal judgement and tactile senses are used when it comes to evaluation of the temperature of the refrigerator (George et al., 2010). However, the subjective experience of tactile coldness can easily be misleading. Borda et al. (2020) showed that the probability to give the exact temperature value after touching a surface or food kept at 8 °C is very low. Consumers demonstrated limited ability to assess the food and surface temperature by tactile senses when the skin touches refrigerated foods. The estimated temperature is highly related to the thermal properties of the packaging materials, for example an aluminium can is estimated colder than a PET bottle when kept at refrigeration temperature. Food handling behaviour may also differ among age groups. EFSA BIOHAZ Panel (2018) noted that unsafe practices, including prolonged storage time and too high temperatures, are not uncommon among elderly. However, there is a wide variation in behaviour among elderly, which makes it challenging to generalise about knowledge and food handling behaviours (Dumitrascu et al., 2020).

Temperature abuse during storage can support the growth of Listeria monocytogenes to levels that largely increase the health risk (Mataragas et al., 2006). L. monocytogenes remains a significant public health concern, despite not being among the most commonly reported causes of food-borne illnesses (Buchanan et al., 2017). The capacity of L. monocytogenes to grow under extreme conditions such as low temperature, low pH and high salt concentrations, is among the most important factors affecting the risk of human listeriosis associated with the consumption of ready-to-eat (RTE) foods. These foods are of particular concern, because of the lack of a cooking step to inactivate L. monocytogenes prior to consumption (Georgalis et al., 2020). Cooked meat and heat-treated sausages with extended shelf lives were the RTE food subcategories with the most consumed servings per person and per year in the European Union (EU) (EFSA BIOHAZ Panel, 2018). Cooked meat products were associated with the largest number of listeriosis cases per year (Sampedro et al., 2022; Hadjicharalambous et al., 2019). Risk assessment for L. monocytogenes in deli meats predicted that 63 %-84 % of human listeriosis cases and deaths attributable to deli meats are due to retail-sliced products (FSIS, 2010; Gombas et al., 2003; Pradhan et al., 2011). Contaminated RTE meat products constitute a high risk for the susceptible populations, such as pregnant women, the elderly and immune-compromised persons (FAO and WHO, 2022; Mataragas et al., 2008; Sampedro et al., 2022). Since the start of EU-wide surveillance, most listeriosis cases have been reported in the elderly, in particular those over 64 years of age. At the EU level, the proportion of listeriosis cases in this age group steadily increased from 56.1 % in 2008 to 72.5 % in 2020; in the age group over 84, there was an increase from 7.3 % to 17.1 % in the same time period (EFSA and ECDC, 2021).

Quantitative microbiological risk assessment (QMRA) is a modelling approach used to estimate the risk of illness when a population is exposed to a pathogen. The process of QMRA involves four steps: i) hazard identification, ii) exposure assessment, iii) hazard characterization and iv) risk characterization (EC, 1997). In the exposure assessment step of the QMRA, the microbial kinetics of the identified hazards are quantitatively described for all subsequent steps of the product's value chain, using mathematical models. However, this system is inherently stochastic. For that reason, it is essential to include uncertainty and variability in the analysis (EFSA, 2018). In this context, variability refers to sources of variation that are inherent to the system (e.g., biological differences between microbial cells or human beings), whereas uncertainty is related to lack or imprecise knowledge. This can be accomplished by a stochastic modelling approach where the relevant kinetic parameters are described using probability distributions (Garre et al., 2020; Nauta, 2000).

OMRA's that have incorporated the consumer phase indicate that storage time and storage temperature in the consumer phase are important parameters to control (Hadjicharalambous et al., 2019; Mataragas et al., 2010). These consumer practices have a crucial impact on the final dose of exposure. Neves et al. (2018) concluded that, for adequate risk assessments, it may be necessary to include the variation in consumer practices (e.g., variation in storage time and temperature), because this variation is expected to be large. However, lack of information about these consumer practices makes it difficult to draw quantitative conclusions on the variation in food handling practices and how this may contribute to the number of human listeriosis cases (FAO and WHO, 2022). However, EFSA estimated, based on predictions of the QMRA model, that the expected number of invasive listeriosis cases per year is reduced by 37 % in the absence of growth after retail (i.e., at the consumer phase) (EFSA BIOHAZ Panel, 2018). Another risk assessment that focussed on L. monocytogenes in deli meats estimated an up to a million-fold increase in risk due to consumer mishandling, and storage practices appeared to be more important in terms of risk than crosscontamination (Yang et al., 2006).

To date, still little is known about the consumer practices regarding refrigeration of RTE food and its impact on the number of listeriosis cases. Therefore, in this study we conducted a survey to obtain quantitative insight into the knowledge and behaviour of Dutch consumers regarding refrigeration, as well as quantitative information about the temperature profiles in domestic refrigerators. The outcomes of this survey were used to quantitatively estimate the number of listeriosis cases per year due to the consumption of RTE cooked meat products among different risk groups of the Dutch population. The most risky food-handling practices were identified. These results help in designing risk communication messages that target most risky food-handling behaviour.

2. Materials and methods

2.1. Consumer knowledge and temperature data collection of domestic refrigerators

A questionnaire was developed to evaluate consumers' knowledge and behaviour regarding the temperature of their refrigerator (Table 1). The included questions covered four topics: (i) socio-demographic information regarding the respondents, (ii) general information about their refrigerator, such as type, age, and digital temperature indication (iii) user's habits regarding checking and changing the temperature, and (iv) consumer's knowledge regarding the temperature setting.

List of questions as mentioned in consumer survey about refrigerators' temperature.

Questions of consumer survey

Table 1

- 1. Does your refrigerator have a (digital) temperature display?
- 2. What temperature is indicated on the temperature display?
- 3. Do you know, without measuring the temperature of your refrigerator, what the temperature of your refrigerator is?
- What do you think the temperature of your refrigerator is? (in case of no temperature display)?
- 5. Do you, or someone else, ever change the temperature of your refrigerator?
- 6. Do you ever check the temperature of your refrigerator?
- 7. How do you check the temperature of your refrigerator?
- 8. If your refrigerator is not at the right temperature, what do you usually do?
- 9. What type of refrigerator do you have?
- 10. How do you change the temperature of your refrigerator?
- 11. If you want to make the refrigerator colder, what do you usually do?
- 12. How old is your refrigerator?
- 13. What is in your opinion the ideal temperature of a refrigerator?
- 14. What is the temperature you just measured in your refrigerator?

 $^{^{\}mathrm{a}}$ Question 14 was asked to 536 participants measuring the temperature of their refrigerator.

Data collection was performed by the panel agency Flycatcher. Selected consumers were asked by e-mail to participate in the survey. The goal was to sample 1000 consumers to obtain a representative result. Considering an expected response rate of 65 % (based on the experience of the panel agency), an initial sample of 1630 consumers was randomly selected from the participant database of the panel agency based on socio-demographic representativeness, i.e., age, gender, educational level, and geographics. In case of a positive response a questionnaire was taken by a website survey. This message included detailed information about the purpose of the study (e.g., aim, estimated time required for completion, information about anonymity). Inclusion criteria were being aged 18 years or older and currently living in the Netherlands. Consumers who participated (n = 1020), completed the first part of the questionnaire (questions 1-13) and were asked to participate in the follow-up study to measure the temperature of their refrigerator. These participants (n = 536) received a fridge thermometer (Hendi 271117, temperature range - 40 °C to 40 °C, accuracy 1 °C) by post mail. The participants were instructed to place the thermometer in a glass (150 mL) of cold water from the tap at the bottom shelf of the refrigerator. The thermometer was read after being kept in a closed refrigerator for at least 2 h (preferably read in the morning after an overnight). Two hours proved to be sufficient to reduce the temperature of the cold tap water to the stable temperature of the refrigerator. The results were recorded in question number 14 of the questionnaire. Mean scores, standard deviations, and 75th and 95th percentiles were calculated for the measured temperature of the household refrigerators. A Shapiro-Wilk test was performed to check whether the data was normally distributed. The correlation between the ideal mentioned temperature (question 13) and the measured temperature (question 14) was calculated by the Spearman's correlation coefficient (r_s), and chi-square statistic was used to calculate the *p*-value with a level of significance α is 0.05. To understand if consumer characteristics (age, gender, education level) were associated with different temperatures, a multiple linear regression was conducted with age, gender, and education level (low, middle, high) as independent variables and measured temperature as the predicted dependent variable.

2.2. 24-hour temperature measurement

Following the collection of information on the temperature status of 536 domestic refrigerators at the bottom shelf, a more detailed followup study was performed to investigate the temperature distribution inside a set of domestic refrigerators. The temperature was assessed at three different locations in the refrigerator (upper, middle and bottom shelf) in 50 households (17 student homes, 16 family homes, 15 elderly homes, 2 single persons). The temperature was monitored in a container $(2.5 \cdot 2.5 \cdot 5 \text{ cm})$ filled with water (60 mL) for a minimum period of 24 h with a temperature datalogger (DS1921G-F5# Thermochron iButton, accuracy 0.5 °C and programmed to record the temperature at 10 minute intervals). Recorded values were downloaded from the dataloggers using 1-WireViewer and analysed with Microsoft Excel 365 (Microsoft, Redmond, USA). A Mann-Whitney test (level of significance α is 0.05) was used to check differences between the temperature distribution of the different locations in the refrigerator (upper, middle or bottom shelf), and differences between the temperature distribution of households. Relevant information was collected from the participants in an interview, such as the method and place of storage of RTE cooked meat products. To check whether water reacts the same to temperature changes in the refrigerator as RTE cooked meat products, data loggers were placed inside pâté and in a container with water (n = 50 measurements). Both products were stored side-by-side in the same place in the refrigerator (upper, middle or bottom shelf) and the temperature was monitored for 24 h. A Mann-Whitney test (level of significance α is 0.05) was used to check differences between the temperature distribution of pâté and water.

2.3. Quantitative microbiological risk assessment

A QMRA was performed with emphasis on consumer handling and storage of RTE cooked meat products before consumption. Stages of manufacturing and distribution of the product before the retail phase were not included. A stochastic model was developed that describes the prevalence and growth of *L. monocytogenes* on RTE cooked meat products, considering the effects of temperature and time, during transport from supermarket to home and storage at home. The QMRA modules were based on information derived from the consumer survey, refrigerator survey and previous risk assessments.

2.3.1. Prevalence

Information from the EU-wide baseline survey 2010–2011 was used to estimate the frequency of contamination (i.e., prevalence) (EFSA BIOHAZ Panel, 2018). The EU prevalence of L. monocytogenes contaminated RTE heat-treated meat products was 2.07 % (CI: 1.63% - 2.64%) (72 positive samples out of 3470, at the end of shelf-life). A food sample was considered contaminated with L. monocytogenes if either the detection test result was positive (detected in 25 g of sample) and/or the enumeration test result was positive, i.e., having at least one colony on a plate of the first decimal dilution (resulting in a count of at least 10 CFU/g in the original sample). The prevalence was described by a PertAlt distribution, assuming a most likely value of 2.07%, $P^{2.5}$ of 1.63%, $P^{97.5}$ of 2.64%, and always between 0% and 100% (EFSA BIOHAZ Panel, 2018).

2.3.2. Initial concentration

The initial concentration (i.e., at point of sale, in case of being contaminated) was estimated using information from the EU-wide baseline survey 2010–2011. A beta-general distribution was chosen to describe the initial concentration, assuming α equal to 0.502 and β equal to 2.908 and a minimum of -1.69 log CFU/g and a maximum of 6 log CFU/g (EFSA BIOHAZ Panel, 2018).

2.3.3. Consumer handling

Accurate data about temperature profiles during transport from retail to home were not available. Possas et al. (2019) assumed that the transport from retail to home can last a minimum of 15 min and a maximum of 2 h. A uniform distribution was used with a minimum of 0.25 h and a maximum of 2 h (Possas et al., 2019). The temperature during transport is largely unknown, but in general, foods are not refrigerated during transport by the consumer and at this phase there is an increase in temperature which depends on many uncertain and variable factors. A beta-pert distribution was used to describe the temperatures during transport from retail to home with minimum 4 °C, most likely 10 °C and maximum 25 °C, as proposed by expert opinion published by Nauta et al. (2003). Although it is unlikely that the temperature will rise from refrigeration temperature to a maximum of 25 °C within 2 h, on a hot day, it could potentially happen due to the thinness of a package of ham and the potential for a rapid temperature increase.

Household storage times were determined from a consumer survey, executed in 2021 (Voedingscentrum, 2021). In this survey 1690 participants were asked to answer the question 'within how many days do you eat opened cooked deli-meat'. Possible answers were: 1. Within one day (0.5); 2. within two days (1); 3. within three to four days (3.5); 4. within five to six days (5.5); 5. for a week or longer (9); 6. until durability date (an equal allocation of days between 4 and 14); 7. I use my senses (an equal allocation of days between 4 and 14). The number of days used to determine the distribution is shown in brackets. A pert distribution was used to describe storage time with a most likely of 1.52 days, a minimum of 0 days and a maximum of 18 days, which was the maximum storage time according to the survey. Refrigerated storage temperatures at the household were used from the survey among the 534 households. A normal distribution was fitted to describe the temperature data with a mean (μ) of 5.68 °C and a standard deviation (σ) of

2.23 °C. A minimum of 0 °C (below zero products will freeze) and a maximum of 18 °C was chosen (the maximum temperature of the refrigerators in the survey).

2.3.4. Consumption data

Data about individual consumption patterns (typical serving size) of cooked deli-meat of the Dutch population were retrieved from the Dutch National Food Consumption Survey 2012–2016 (Van Rossum et al., 2020). The food group 'Cold processed meat' was chosen, and deli-meats which were cooked or boiled (e.g., pâté or ham), pre-packaged and pre-sliced were chosen as representative for RTE cooked meat (Appendix A). A pert distribution was chosen with a mean serving size of 30 g per consumption day, a minimum of 8.1 g, a most likely of 20 g and a maximum of 100 g. On average people consumed 48 % of the days 'cold processed cooked meat' (=175 days (servings) per year). A normal distribution was chosen with μ is 175 servings and σ is 50 servings.

2.3.5. Growth model for Listeria monocytogenes

Maximum growth rates μ_{max} of RTE pâté and cooked ham were retrieved from Combase at www.combase.cc (USDA, 2023). For a reliable estimation of growth parameters, experimental growth curves were only used for estimation of the maximum growth rate if they met the following criteria: (i) if there was exponential growth, there should be at least four data points in the exponential phase, (ii) parameters of the product like aw, pH, NaCl and sodium nitrite concentration should be reported, (iii) no specific package conditions like vacuum or modified atmosphere. For pâté the selection criteria 'organism: 'Listeria monocytogenes/innocua'; food category: 'other or unknown type of meat'; food name: 'pâté' were used. In total, maximum growth rates (log/h) from 143 pâtés were collected in a temperature range of 0 °C and 15 °C, with a_{w} 0.989-0.991, pH 6.2-6.3, NaCl 1.6 % - 2.0 %, and sodium nitrite 81.3-102 ppm. For ham the selection criteria 'organism: 'Listeria monocytogenes/innocua'; food category: 'pork'; food name: 'ham' were used. In total, maximum growth rates (log/h) from 192 hams were collected in a temperature range of 0 $^{\circ}$ C and 15 $^{\circ}$ C, with a_{w} 0.988–0.995, pH 6.1–6.4, NaCl 1.0 %-2.2 %, and sodium nitrite 84-110 ppm.

The square root model described by Ratkowsky et al. (1982) was used to describe μ_{max} as function of temperature (T).

$$\sqrt{\mu_{\max}} = b \cdot (T - T_{\min}),\tag{1}$$

where μ_{max} is the maximum growth rate (log/h); b is the regression parameter determined during the modelling process ($\sqrt{(\log/h/^\circ C)}$); T is the storage temperature (°C); and T_{min} is the theoretical lower temperature at which the estimated maximum growth rate of L. monocytogenes first becomes 0. A linear regression line was plotted for pâté and cooked ham, and the mean, standard error, 95 % confidence and 95 % prediction interval and the correlation coefficient (R^2) of the model were calculated (Appendix B). To express the variability of μ_{max} , the deviation of the $\sqrt{\mu_{max}}$ (delta $\sqrt{\mu_{max}}$) was represented by a normal distribution with mean of 0 and standard deviation represented by the standard error of the model fit, which was found independent of temperature on this square root scale. This $delta\sqrt{\mu_{max}}$ was added to the square root model, i. e.,

$$\sqrt{\mu_{max}} = b \cdot (T - T_{min}) + delta\sqrt{\mu_{max}}.$$
 (2)

This approach differed from the method described in the FDA & FSIS risk assessment, which utilizes a baseline exponential growth rate at 5 °C with its deviation, and from that the growth rate is converted to the actual temperature with $\mu_{max} = (\mu_{max,5} + delta\mu_{max}) \cdot [(T - T_{min}) / (5 - T_{min})]^2$ (FDA and FSIS, 2003). The approach described in the FDA & FSIS risk assessment was not used because simulations showed that the variability of $\sqrt{\mu_{max}}$ increased with increasing temperature above 5 °C. However, our study and other studies showed that the variability of $\sqrt{\mu_{max}}$ remains constant and is independent of temperature (Abe et al., 2023; Aryani et al., 2016; Le Marc et al., 2021). Note that the use of

 $delta\sqrt{\mu_{max}}$ may yield a negative value for $\sqrt{\mu_{max}}$. Squaring this negative value would still produce a positive μ_{max} , which is incorrect. To address this issue, first $\sqrt{\mu_{max}}$ was calculated. If the result was negative, the μ_{max} was set at zero, and when it was positive, the $\sqrt{\mu_{max}}$ was squared and used in the simulations. A more detailed description of the followed approach can be found in Appendix C.

To verify the results, the outcome of the square root model was compared with maximum growth rates as described in the literature, but not present in Combase. Maximum population densities of *L. monocytogenes* were derived from Combase, involving 179 hams, with a recorded increase of >0.5 log CFU/g. A pert distribution was selected with a minimum of 3.6 log CFU/g, a most likely of 8.2 log CFU/g, and a maximum of 8.5 log CFU/g (Szczawiński et al., 2017; Zhang et al., 2012).

2.3.6. Hazard characterization

An exponential dose-response model was used to estimate the probability of illness given a dose:

$$P_{iii}(D;r) = 1 - e^{(-r \cdot D)}$$
 (3)

where D represents the ingested dose and r is the dose-response parameter, representing the probability of illness per CFU. Two different r values were considered: r is 2.37×10^{-14} for the low-risk population and r is 1.06×10^{-12} for the susceptible population as suggested by the World Health Organization/Food and Agricultural Organization of the United Nations (WHO and FAO, 2004).

The QMRA was based on the Dutch population. According to Statistics Netherlands (CBS) the total number of inhabitants was 17,475,415 dated January 1st, 2021 (Statistics Netherlands CBS, 2021). The fraction of the total population being at risk, i.e., 'susceptible or high-risk' is considered 20 % of the total population as described by Mataragas et al. (2010). Included groups were:

- Aged population (65 years or older). It is estimated that 95 % of the high-risk group consists of elderly people (Statistics Netherlands CBS, 2021).
- Pregnant women. It is estimated that 5 % of the high-risk group consists of pregnant women (Friesema et al., 2022).

Immunocompromised people and new-born infants who are susceptible for *L. monocytogenes* were excluded, because they constitute a relatively small group and new-born infants do not consume cooked deli meats.

Cross-contamination during transport from supermarket to home and during household storage was not included in the QMRA, because storage practices appeared to be much more important in terms of risk for deli-meats than cross-contamination (Yang et al., 2006).

2.3.7. Risk characterization

Monte Carlo simulations were performed to determine the distribution of P_{ill} using @Risk simulation software (@Risk 8.2 for Excel, Palisade, Ithaca, USA), as an add-in to Microsoft Excel, with 1,000,000 iterations. For each iteration, the risk per serving for the high-risk population $(R_{serv,hr})$ and the low-risk population $(R_{serv,hr})$, respectively was calculated by multiplying P_{ill} with the prevalence. The number of cases per year was calculated by multiplying the average arithmetic risk per serving $(R_{serv,\ hr}$ and $R_{serv,\ lr})$ with the average number of servings per year per person (Nrserv) and with the number of persons in the high-risk (Pophr) and low-risk population (Pophr), respectively. The number of cases for the total population ($Nr_{cases\ avg,\ tot}$) was calculated by summating the number of cases of the high-risk population ($Nr_{cases\ avg,\ hr}$) and the low-risk population ($Nr_{cases\ avg,\ lr}$). The number of cases was also calculated performing a Bernoulli trial with 1,000,000 iterations using the P_{ill} distribution. A value of 1 indicated an illness case, whereas a value of 0 indicated no illness case. The total number of cases was then

 Table 2

 Overview of input variables for the QMRA model for 'consumption, dose-response and risk of infection module'.

ID	Input variable	Distribution or formula	Unit	Data source/reference
	eria monocytogenes			
$/\mu_{ m max}$	Growth rate (square root) of <i>Listeria</i> monocytogenes in cooked ham	$b\cdot (T-T_{min})$	$\sqrt{\log/h}$	Fitted with data from Combase (b: 0.015; T_{min} : -0.7 °C)
lelta $\sqrt{\mu_{max}}$	Deviation of the growth rate (square root) due to variability	Normal (0; 0.0286)	$\sqrt{log/h}$	Calculated from linear regression (square root mode
emax	Maximum concentration of <i>Listeria</i> monocytogenes in ham	Pert (3.6; 8.2; 8.5)	log CFU/g	Fitted with C_{max} data from Combase
nitial concenti	ration and prevalence			
70 20	Initial concentration	BetaGeneral (0.502; 2.908; -1.69; 6)	log CFU/g	EFSA (2018)
Prev	Prevalence at end of shelf life	PertAlt (2.07 %; 1.63 %; 2.64 %)	%	EFSA (2018)
ransport to h	ome			
ime _{tr}	Transport time from retail to home	Uniform (0.25, 2)	h	Possas et al. (2019)
tr	Transport temperature from retail to home	BetaPert (4; 10; 25)	°C	Nauta et al. (2003)
max,tr	Growth rate during transport to home	If $(b \cdot (T_{tr} - T_{min}) + delta\sqrt{\mu_{max}}) < 0$ then 0, else:	log/h	Calculated
S _{tr}	Concentration at end of transport	$(b \cdot (T_{tr} - T_{\min}) + delta\sqrt{\mu_{max}})^2$ Min $(C_0 + \mu_{max,tr} \cdot time_{tr}, C_{max})$	log CFU/g	Calculated
·u	towards home	(50 + pmax,ir the as omax)	100 01 0/8	
Consumer store	age			
ime_h	Household storage time	Pert (0; 1.52; 18.3)	days	Own survey with max 21 days
h	Household temperature refrigerator	Normal (5.68; 2.23)	°C	Own survey with min: 0 $^{\circ}\text{C}$ and max: 18 $^{\circ}\text{C}$
max,h	Growth rate during home storage	If $(b \cdot (T_h - T_{min}) + delta\sqrt{\mu_{max}}) < 0$ then 0, else: $(b \cdot (T_h - T_{min}) + delta\sqrt{\mu_{max}})^2$	log/h	Calculated
cons	Concentration at consumption	$\min (C_{tr} + \mu_{max,h} \cdot time_h \cdot 24, C_{max})$	log CFU/g	Calculated
Concumption d	lata			
Consumption of Size	Serving size (mass of cold processed	Pert (8.1; 20.0; 100.0)	g	Van Rossum et al. (2020)
Vr _{serv}	meat per serving) Number of servings (consumption days) per year per person	Normal (175, 50)	servings	Van Rossum et al. (2020) with max: 350 servings
Pose-response		$10^{C_{cons}} \cdot S_{size}$	CELL	Colordotod
)	Ingested dose (number of cells consumed per single serving)		CFU	Calculated
Pill, hr	Probability of illness per contaminated serving for high-risk population	1- e ^(-r_{hr} · D)		WHO and FAO (2004) with r_{hr} : 1.06×10^{-12}
R _{serv, hr}	Risk per serving for high-risk population	P _{ill, hr} · Prev		Calculated
ill, Ir	Probability of illness per contaminated serving for low-risk population	$1 - e^{(-r_l \cdot D)}$		WHO and FAO (2004) with r_{lr} : 2.37 \times 10 ⁻¹⁴
R _{serv, lr}	Risk per serving for low-risk population	$P_{ill, lr} \cdot Prev$		Calculated
Population				
Pop_{tot}	Total population	17,475,415	persons	Statistics Netherlands CBS (January 1st, 2021)
Pop_{hr}	High-risk population	$0.2 \cdot Pop_{tot}$	persons	Calculated (Mataragas et al., 2010)
Pop _{lr}	Low-risk population	$0.8 \cdot Pop_{tot}$	persons	Calculated (Mataragas et al., 2010)
llness occurre				
Nr _{cases avg, hr}	Nr of illness cases per year (average) for the high-risk population	$\overline{R_{serv,hr}} \cdot \overline{Nr_{serv}} \cdot Pop_{hr}$	cases per year	Calculated with $\overline{x} = \frac{1}{n} \sum_{i=1}^{n} x_i$ and $n = 1,000,000$ iterations
Nr _{cases avg, lr}	Nr of illness cases per year (average) for	$\overline{R_{serv,lr}} \cdot \overline{Nr_{serv}} \cdot Pop_{lr}$	cases per	Calculated with $\overline{x} = \frac{1}{n} \sum_{i=1}^{n} x_i$ and $n = 1,000,000$
	the low-risk population	V V	year	iterations
Ir _{cases avg,}	Nr of illness cases per year (average) for the total population	$Nr_{cases\ avg,\ hr} + Nr_{cases\ avg,\ lr}$	cases per year	Calculated
tot N r cases bern, hr	Nr of illness cases per year (Bernoulli) for the high-risk population	Bernoulli ($< P_{ill, hr}$), if illness = 1, if not = 0	cases per year	Calculated by summation with $n = 1,000,000$ iterations, and multiplied by Nr_{serv} and corrected by
				Pop_{hr} .
	Nr of illness cases per year (Bernoulli)	Bernoulli ($\langle P_{ill, lr} \rangle$), if illness = 1, if not = 0	cases per	Calculated by summation with $n = 1,000,000$
$Nr_{cases\ bern,\ lr}$	for the low-risk population		year	
Nr _{cases bern, lr}		$Nr_{cases\ bern,\ hr} + Nr_{cases\ bern,\ lr}$	year cases per	iterations, and multiplied by <i>Nr_{serv}</i> and corrected by <i>Pop_{lr}</i> . Calculated

calculated by summing up all cases of the 1,000,000 iterations and multiplying this by the average prevalence, the average number of servings and the high-risk population and low-risk population per 1,000,000, respectively. This was repeated 100 times to calculate the estimated mean and the standard deviation of the number of cases. To validate the developed QMRA model, the outputs were compared with the epidemiological data for listeriosis in the Netherlands. An overview of the QMRA model and the parameters' values and/or distributions is shown in Table 2.

2.3.8. Sensitivity analysis

Three types of sensitivity analyses were conducted on the stochastic model (Table 2) to identify the most crucial factors influencing the probability of illness (P_{ill}) and the number of cases. A tornado chart was created to illustrate the impact of different sources of variability on the final output (Pill), using Spearman's rank correlation coefficient. Additionally five simulations (each with 1,000,000 iterations) were performed, and iterations that resulted in illness cases (denoted by a value of 1 from the Bernoulli trial) were analysed to evaluate the underlying input values. These input values were illustrated in the distribution figures as a bar code plotting. In addition, a deterministic approach was used to determine the effect of worst-case scenarios of the input variables on the final output. For this an input variable was set at a high value (e.g., P⁹⁵ for a normal distribution) while keeping the rest of the input variables at the mean value, and then comparing the resulting risk per serving to the risk per serving in the default scenario (i.e., all input variables set to the mean value).

2.3.9. Intervention scenarios

To evaluate the impact of input variables on the number of listeriosis cases, 'what if' scenario analyses were conducted. The baseline model (Table 2) was modified by truncating the following input variables at various maximum limits: (i) initial concentration (maximum: 5, 4, 3 and 2 log CFU/g), (ii) storage time (maximum: 14, 10, 7, and 4 days), (iii) refrigerator temperature (maximum: 14, 10, 7, and 4 °C), (iv) a combination of storage time and refrigerator temperature (maximum: 12 days and 12 °C, 10 days and 10 °C, 7 days and 7 °C, and 4 days and 4 °C). The model was re-run for each scenario with 1,000,000 iterations.

Furthermore, it was evaluated how variations in storage practices between high-risk populations and low-risk populations affected the number of listeriosis cases and the Years of Life Lost (YLL). According to the World Health Organization, 98 % of the Disability-Adjusted Life Years (DALY) estimates for listeriosis are due to YLL, which highlight the high case-fatality rate of listeriosis (FAO and WHO, 2022). The YLL were estimated based on the following assumptions: i) 95 % of the high-risk population cases consists of elderly of 65 years and older (Statistics Netherlands CBS, 2021), and 5 % of the high-risk population cases were pregnant women that gave birth to an infected baby (Friesema et al., 2022); ii) a case-fatality ratio of 0.15 was applied for perinatal listeriosis (pregnant women) and 0.25 for acquired listeriosis (in elderly and lowrisk population) (De Noordhout et al., 2014; WHO and FAO, 2004); and iii) the YLL per case was estimated to be 85 years for perinatal listeriosis (life expectancy in the Netherlands (Statistics Netherlands CBS, 2021)), 10 years for acquired listeriosis in elderly above 65 years old (the median age of reported listeriosis cases in the Netherlands in 2020 was 75 years (Friesema et al., 2022)), and 43 years for acquired listeriosis for the low-risk population (the average age of people in the Netherlands is 42 years (Statistics Netherlands CBS, 2021)). The YLL of the total population was calculated using the following formula:

$$YLL_{total} = \sum_{i=1}^{i} number of cases_{i} \cdot case-fatality ratio_{i} \cdot YLL_{i}$$
 (4)

where i represents the persons of 65 years and older, the pregnant women (neonates), and the low-risk population. The following variations in household storage temperatures were included: a) default with 5.7 °C for both high-risk and low-risk population (baseline as shown in Table 2); b) difference between storage temperature of older individuals of 6.1 °C (persons 65 years and older) and younger individuals of 5.5 °C (low-risk population and pregnant women), based on the results of the consumer survey; c) storage temperature of the upper shelf of 7.7 °C for both high-risk and low-risk population, as determined in the 24-hour temperature measurements; and d) extreme storage temperature of the upper shelf of 13.4 °C for both high-risk and low-risk population, as determined in the 24-hour temperature measurements. To estimate the number of listeriosis cases and the YLL, the model was re-run with 1,000,000 iterations for each variation. The ratio for each variation was determined by dividing the listeriosis cases of the specific scenario by the listeriosis cases of the default scenario.

3. Results

In total 1020 consumers participated and completed the first part of the questionnaire (questions 1–13) and 536 consumers also measured the temperature of their refrigerator and completed the second part of the questionnaire (questions 14). Two results of the measured temperatures were invalid (temperature refrigerator of -9 and $-13\,^{\circ}\text{C}$), leaving 534 participants with valid temperature results.

3.1. Consumer knowledge and domestic refrigerators temperature analysis

The analysis of the measured temperatures of 534 domestic refrigerators (bottom shelf) showed a mean temperature of 5.7 °C (SD 2.2 °C). The measured temperatures varied from -1 °C to 17 °C, with 66.8 % of the refrigerators with a temperature of 6 °C or lower. Following a normal distribution, the 75th percentile was determined at 7.2 °C and the 95th percentile at 9.4 °C. An overview of the distribution and the cumulative frequency is given in Fig. 1.

The survey showed that around 28 % of the consumers indicated 4 °C as the temperature a refrigerator should have, which is the recommended temperature in the Netherlands. Around 8 % of the respondents indicated a temperature below 4 °C, 22 % at 5 °C, 16 % at 6 °C, 18 % at 7 °C, and 8 % of the respondents at 8 °C. Based on Spearman's correlation analyses, and confirmed by the chi-square statistic, a positive and significant correlation was seen between the mentioned temperature the refrigerator should have and the measured temperature of the refrigerator ($r_s = 0.667$, p < 0.01). This means that consumers who indicated a lower recommended temperature, also measured a lower temperature of their refrigerator.

Most of the respondents never checked the temperature of the refrigerator (61 %), 37 % of the participants occasionally, and 2 % regularly. Of those who indicated that they checked the temperature, approximately half of the respondents checked it with a thermometer (45 % measuring the air temperature and 6 % the temperature in a glass of water), making it in total 20 % of all respondents checking the refrigerator with a thermometer.

Multiple linear regression analyses with age, gender, and education level as independent variables and the measured or indicated recommended temperature as the predicted dependent variable showed no significant impact of gender (men, women) and education (low, middle, high). Age had a significant impact on both the measured temperature and on the indicated recommended temperature. The measured temperature of the refrigerators of older consumers (65 years and older) were on average 0.6 °C higher than that of younger consumers below 35 years of age, i.e., 6.1 °C and 5.5 °C respectively. Older consumers

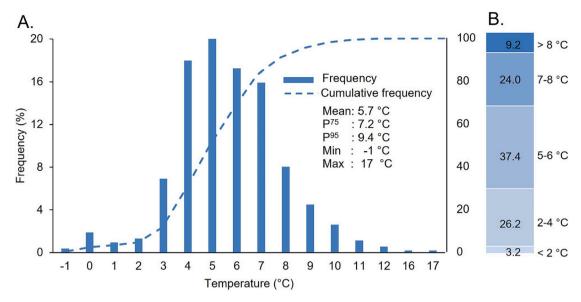


Fig. 1. Frequency (%) and cumulative frequency (%) of the 534 domestic refrigerators with mean temperature, P^{75} , P^{95} , minimum and maximum (A). Percentage of refrigerators with temperature below 2 °C, from 2 to 4 °C, from 5 to 6 °C, from 7 to 8 °C and above 8 °C (B).

significantly indicated a higher recommended temperature than younger consumers (5.9 $^{\circ}$ C versus 5.0 $^{\circ}$ C). Younger consumers were better aware of the recommended temperature of 4 $^{\circ}$ C. Notably, the refrigerators with the highest measured temperature (16 and 17 $^{\circ}$ C) were both owned by old participants (79 and 86 years old). Statistical analysis showed that refrigerator's type and refrigerator's age did not significantly affect the measured temperature.

3.2. 24-hour temperature profiles

The analysis of the 24-hour temperature profiles of 50 surveyed refrigerators showed differences in temperature inside the household refrigerators (Fig. 2). The highest temperature was measured on the upper shelf (mean 7.7 °C, SD 2.7 °C) and the lowest temperature was measured on the bottom shelf (mean 5.7 °C, SD 2.1 °C). This result is almost perfectly equal (5.7 °C, SD 2.2 °C) to the results from the 534 measurements of the consumer survey. The difference was significant between the upper shelf and the middle and bottom shelf (p < 0.05). No significant difference was found between the middle and the bottom

shelf. In general, a cyclical fluctuation of the temperature was noticed, and depending on the brand and the on-off cycles of the compressor, less or more fluctuation was noticeable per refrigerator. Seven out of fifty refrigerators showed the lowest temperatures on the upper shelf, which was the opposite from the other refrigerators. These seven refrigerators were a specific type of refrigerator with a small freezer unit at the top of the refrigerator. This freezer unit lowered the temperature of the upper shelf. As also found in the consumer survey, refrigerator's brand and refrigerator's age did not significantly affect the measured temperature.

The refrigerators at the homes of the elderly were significantly warmer than the refrigerators at the other homes (student, family, single person) (p < 0.05). Especially the top shelf of the elderly was warmer than the top shelf of the other homes (0.9 °C warmer with an average of 8.3 °C). Information retrieved from the interviews showed that 34 % of the participants stored meat products on the bottom shelf, 30 % on the middle shelf, and 36 % on the top shelf or on every shelf where space is available.

No significant difference was found between 24-hour temperature measurement of pâté and water (p = 0.734). Water turned out to be a

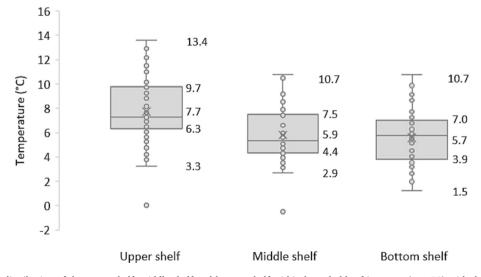


Fig. 2. The temperature distribution of the upper shelf, middle shelf and bottom shelf within household refrigerators (n = 50) with the mean (X), median temperature, and the 5th, 25th, 75th and 95th percentile.

good predictor of the temperature course of pâté meat products in the refrigerator. It should be noticed that in pâté it takes a longer time to go from room temperature to refrigerator temperature than in water. This can be explained by the fact that in more solid material no circular flow develops during cooling and the internal heat transfer is lower (Kandhai et al., 2009). A reliable temperature measurement of cooked ham and water could not be conducted due to the challenge of consistently finding a suitable measuring point within the thin slices of the cooked ham using the data logger.

3.3. Maximum growth rates of Listeria monocytogenes in pâté and ham

In Fig. 3 the maximum growth rates in 143 different pâté meat products and 192 different cooked ham meat products are shown that were extracted from Combase. A square root model was fitted to the data to determine the maximum growth rate (μ_{max}) of *L. monocytogenes* and its variation in pâté and cooked ham meat products as function of temperature.

The maximum growth rates found in the literature, and which were not included in Combase (Beumer et al., 1996; Farber et al., 1995; Glass and Doyle, 1989; Hayrapetyan et al., 2012; Hudson and Mott, 1993; Hunt et al., 2018), fitted within the prediction intervals of the respective product types. In addition, more variability was visible in the cooked ham meat products than in pâté. This may be due to more variability in the components of ham (e.g., more variation in salt content) and therefore the ability of L. monocytogenes to grow also varies. In the QMRA model, the growth rate of cooked ham meat product was chosen to represent the RTE cooked meat products. This decision was based on the fact that in the Netherlands, the consumption of cooked ham meat products is more than seven times higher than that of pâté (Van Rossum et al., 2020). Furthermore, the composition of cooked ham is more similar to the other RTE cooked meat products mentioned in Appendix A than pâté. A lag phase was not included in the primary growth model. According to the data reported in Combase, with a maximum culturing and pre-culturing temperature of 15 °C, in 132 out of the 192 hams, the lag time was not significantly different from zero. For those studies reporting a lag time, it ranged from 2 h to 522 h, with an average lag time of 95 h. Therefore, since in most cases (approximately 68 %) the lag phase was zero, and in cases where a lag phase did exist, the assumption was made that the lag time would have passed by the time the consumer purchases the product. This approach opts for the safe side.

3.4. Quantitative microbiological risk assessment (QMRA)

The QMRA model predicted an average number of 191 illness cases per year (SD 54) for the total population. As expected, the high-risk populations displayed a significant higher risk of listeriosis compared to the low-risk population (175 cases versus 16 cases). With 1,000,000 iterations the Bernouilli trial gave on average the same results as the calculation with the arithmetic mean.

3.4.1. Sensitivity analysis

To evaluate the influence of input variability on the outcome variability, a sensitivity analysis was conducted, and the results are visualized in a tornado plot (Fig. 4). Variability in the initial concentration of L. monocytogenes has the largest impact on the variability in the probability of illness (P_{ill}) associated with the consumption of RTE cooked meat products. Household storage time and household storage temperature were also found to be significant contributors, followed by the variability on the growth rate of L. monocytogenes and the serving size. Using a deterministic approach with fixed input variables at their respective minimum and maximum, also showed that the initial concentration, household storage time, household storage temperature and variability in the growth rate had the largest impact on the final output (Appendix D). This confirmed the output of the sensitivity analysis as shown in Fig. 4.

To further explore the factors that contributed to illness cases, the input values of the most important variables that resulted in an illness case were visualized with a barcode of (orange) lines plotted in the input distributions in Fig. 5, following the approach proposed by Abe et al. (2023). Based on model simulations, the average storage time for illness cases was 9.7 days (SD 2.6 days), and the average refrigerator's temperature for these cases was 9.1 °C (SD 1.9 °C). Most of the cases were attributable to storage for >7 days or at refrigerator's temperature above 7 °C. Conversely, the distribution of illness cases for initial concentration was more evenly spread (mean 1.3 log CFU/g; SD 2.1). For growth rate variability, the cases of illness were associated with a positive deviation of the mean (mean 0.0356 $\sqrt{\log/h}$; SD 0.0263). Fig. 5E and F show that cases of illness were linked to a relatively very high risk per serving, where the risk per serving was higher for the high-risk population (mean -4.2 log; SD 0.6) than for the low-risk population (mean -5.9 log; SD 0.6), and considerably higher than the mean risk per serving for all 1,000,000 iterations (-13.1 log (SD 1.8) and - 14.7 log (SD 1.8) for high- and low-risk populations, respectively).

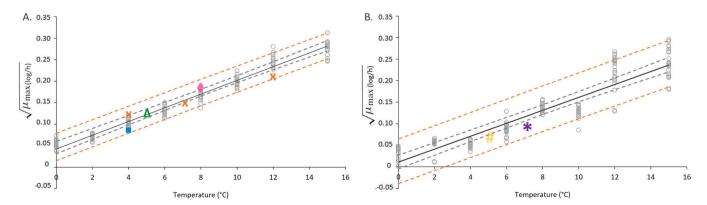


Fig. 3. Square root plot of maximum growth rate ($\sqrt{\mu_{\text{max}}}$) of *L. monocytogenes* in pâté meat products (A) and cooked ham meat products (B) as function of temperature. Open dots represent the maximum growth rates extracted from Combase, solid line indicates the square root function (A: $\sqrt{\mu_{\text{max}}} = 0.0159 \cdot (T + 2.67)$; B: $\sqrt{\mu_{\text{max}}} = 0.0150 \cdot (T + 0.75)$), grey dashed lines indicate the upper and lower 95 % confidence intervals of the model and orange dashed lines indicate the upper and lower 95 % prediction interval. Calculated with a standard error of 0.0144 for pâté and 0.0286 for cooked ham. The coloured symbols represent growth rates from literature \mathbf{X} (Hayrapetyan et al., 2012), \mathbf{I} (Hudson and Mott, 1993), $\mathbf{\Delta}$ (Farber et al., 1995), \mathbf{V} (Hunt et al., 2018), \mathbf{W} (Beumer et al., 1996), \mathbf{V} (Glass and Doyle, 1989).

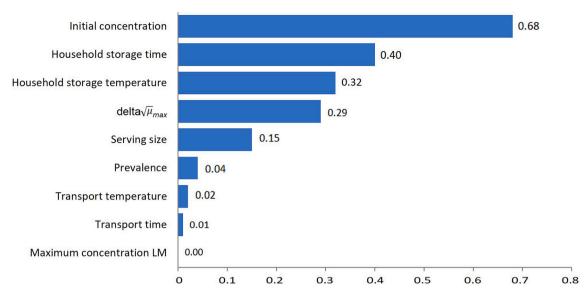


Fig. 4. Sensitivity analysis showing the correlation coefficients (Spearman's rank) of the variability of the input factors on the variability of the probability of illness (P_{ill}).

3.5. Scenarios

A scenario analysis was conducted to determine the impact of various input variables on the estimation of the risk of listeriosis from RTE cooked meat products. Four input variables were selected for assessing potential interventions to reduce the number of listeriosis cases: (i) initial concentration, (ii) household storage time, (iii) household storage temperature, and (iv) combination of household storage time and temperature (Fig. 6). The grey boxplot refers to the default scenario (Table 2), while the red, green, blue, and yellow boxplots represent different scenarios with truncated input values. The analysis showed that a maximum temperature of 4 °C or a maximum storage time of 4 days reduced the estimated number of cases of listeriosis to almost zero. A maximum storage time of 7 days or a maximum storage temperature of 7 °C also reduced the number of cases significantly (83 % and 80 % respectively). A reduction was also visible by reducing the maximum initial concentration, but to a smaller extent than reducing the household storage time and temperature. Improving both household storage time and temperature resulted in even more significant decrease of annual cases of listeriosis. These results demonstrate that consumer behaviour, such as reducing storage time and controlling refrigerator temperature at home, can significantly reduce the risk of listeriosis.

Additional scenario analyses were performed to account for differences in household storage practices among high-risk and low-risk consumer groups (Appendix E). Comparing the default scenario, where the mean temperature of the refrigerator was 5.7 °C for all consumer groups, with the scenario where the mean temperature of the refrigerator was 6.1 °C for the elderly and 5.5 °C for the younger generation, resulted in an overall 1.3-fold increase of illness cases. This increase was primarily observed in the elderly, with a significant higher number of cases, while the younger generation showed a slightly lower number of cases. Furthermore, comparing the default scenario with the scenario where the mean temperature of the refrigerator was set at 7.7 °C (average temperature of the upper shelf) for all consumer groups, resulted in a 3.3-fold increase in number of illness cases. Moreover, comparing the default scenario with the scenario where the temperature of the refrigerator was set at 13.4 °C (extreme temperature measured at the 24-hour temperature measurements) for all consumer groups, resulted in a 30-fold increase in the number of illness cases. It is important to note that it is unrealistic to assume that all individuals store their RTE cooked meat products at such high temperatures. However, it is important to consider this factor of increase for individuals who do store their RTE cooked meat products at elevated temperatures.

4. Discussion

Consumers play an important role in risk reduction associated with L. monocytogenes, as individuals make food handling decisions in purchasing, storage and cooking practices in their homes. One of the most commonly reported consumer food safety practices that increases the concentration of L. monocytogenes are not using proper refrigeration storage (FAO and WHO, 2022). The current study analysed the knowledge and practice of household refrigeration of Dutch consumers. A mean temperature of 5.7 °C was found, which is within the range of temperatures appropriate for the preservation of perishable RTE products (<6 °C) as recommended by the Codex Alimentarius (2007). However, 33.7 % of the refrigerators had a temperature above the recommended value of 6 °C (Fig. 1), the 75th percentile being at 7.2 °C, and the 95th percentile being at 9.4 °C. These results are in agreement with the outputs of a review by James et al. (2017) showing that the temperature performance of household refrigerators has not remarkably changed over the last decades, with mean air temperatures ranging from 3.5 to 9.3 °C in different European countries. Despite the temperature recommendations included in numerous food safety campaigns among consumers, the storage temperature of chilled foods is frequently above 6 $^{\circ}\text{C}$. The reasons for the difference between recommendations and actual practices are unclear. A lack of knowledge of the recommended temperature may be a contributing factor. The current study demonstrated that a majority (72 %) of the Dutch consumer is not familiar with the recommended temperature of 4 °C. Consumers who were familiar with the recommended temperature, have a significantly lower temperature of household refrigerator and this temperature came more in line with the recommendation. This could indicate that knowledge does have an effect on the behaviour regarding refrigeration. Especially younger consumers were more aware of the recommended temperature. Another contributing factor could be a lack of perceived importance or risk control benefit. Optimistic bias, a cognitive bias that causes people to believe that they are less likely to experience a negative event than others, may encourage people to risky food behaviours (Clayton and Griffith, 2004; Da Cunha et al., 2015; Evans et al., 2020; Rodrigues et al., 2020). In this study most respondents never checked the temperature of the refrigerator (61 %), indicating a lack of perceived importance or risk control.

The measured temperature of the refrigerator of older consumers

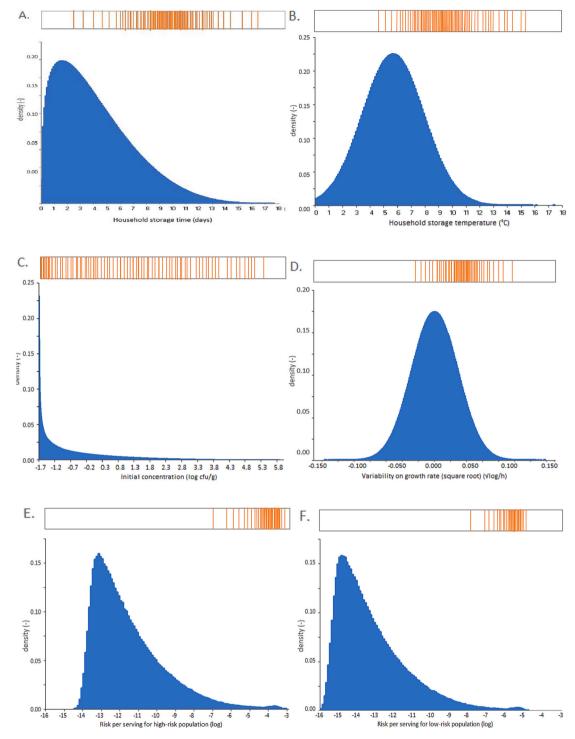


Fig. 5. The distribution of the household storage time (A), household storage temperature (B), initial concentration (C), variability on growth rate (square root) (D), risk per serving for the high-risk population (E), and risk per serving for the low-risk population (F) of 1,000,000 iterations (blue distribution chart) and the corresponding input values from five simulations that resulted in an illness case (orange barcode chart). The orange bars represented are all the cases from both the high-risk and the low-risk group in all the panels.

(above 65 years old) was higher than that of younger consumers. Also the outliers of measured temperatures of $16\,^{\circ}\text{C}$ and $17\,^{\circ}\text{C}$ were measured at households of older consumers (above 80 years old). Looking at the risk of serious health impact due to listeriosis, this is also the group of people who have an increased risk. For example, in the cantaloupe outbreak in the United States of America, the median age of patients who became ill was 78 years; the median age of persons who died was 81 years (McLauchlin et al., 2004). The surveillance data in the

Netherlands showed that the highest mean annual number of listeriosis cases was recorded for the age group above 65 years old. The median age of the reported cases in 2020 was 75 years old, with 62 % being male (Friesema et al., 2022). Since the beginning of EU-level surveillance, most listeriosis cases have been reported in elderly people, in particular those over 64 years of age (EFSA BIOHAZ Panel, 2018). The population of older susceptible consumers is increasing and may represent up to 30 % of the general population in the future (Farber et al., 2021). The

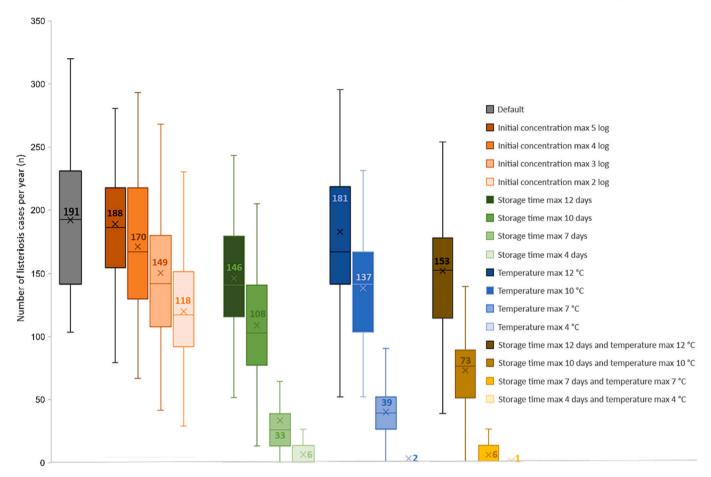


Fig. 6. Number of listeriosis cases per year with four different scenarios: initial concentrations (maximum of 5, 4, 3 and 2 log CFU/g – red boxplots), household storage times (maximum of 12, 10, 7 and 4 days - green boxplots), household storage temperatures (maximum of 12, 10, 7 and 4 °C - blue boxplots), and combinations of maximum household storage time and temperature (maximum of 12, 10, 7 and 4 days and °C – yellow boxplots), compared to the default (grey boxplot). The mean (x), median (line), P²⁵ (bottom boxplot), P⁷⁵ (upper boxplot), minimum and maximum are given. All other variables are varied as in the default scenario.

importance of good temperature and time control is an important control measure to reduce the risk of getting listeriosis, especially for the older generation.

The temperature measured inside the different compartments of the refrigerators varied. The 24-hour temperature profile of the surveyed refrigerators in this study showed that the temperature measured on the upper shelf was significantly higher (7.7 °C) than the temperature measured on the bottom shelf (5.7 °C). Both the 24-hour temperature measurement with 50 refrigerators and the temperature measurement of the consumer survey with 534 refrigerators had, on average, the same temperature on the bottom shelf (5.7 °C (SD 2.1 °C) and 5.7 °C (SD 2.2 °C), respectively). The large difference of temperature between the upper and lower shelf indicates that for the best temperature measurement in the refrigerator, two or three temperature points (top, middle, and bottom) may be needed, as also suggested in other studies (George et al., 2010; Laguerre et al., 2002). But the question is whether consumers are prepared to use multiple thermometers to check the temperature of their refrigerator. The survey showed that only a minority of the consumers check the temperature of the refrigerator (39 %). And if they did, it was only with one thermometer at one point in the refrigerator. This suggests that a more intelligent system for monitoring refrigerator temperatures is needed; one that provides more interactions with, and feedback to, the householder. To meet the recommendation of the Codex Alimentarius (2007) that perishable RTE products should be stored below 6 °C, consumers should be informed to store these products on the bottom or middle shelf. The survey showed that two third of the participants stored meat products indeed on the bottom or middle shelf (32 out of 50 participants), but more research is needed to evaluate how

this varies among different countries. Jofré et al. (2019) noticed that most of the interviewed consumers in Spain claimed to store cooked meat products in the refrigerator core (94 %), especially on the upper shelves. Although the general advice towards consumers should be to store RTE cooked meat products on the bottom or middle shelf, there can be some exceptions. The survey showed that refrigerators with a freezer unit on top of the fridge showed the lowest temperatures on the upper shelf, which was the opposite from the other refrigerators.

RTE cooked meat products are associated with the highest number of listeriosis cases (FDA and FSIS, 2003; Sampedro et al., 2022). Deli meats were ranked as very high risk by the U.S. Food and Drug Administration, both per serving and per annum. This high risk ranking is due to their relatively high contamination levels, their ability to support growth of L. monocytogenes under refrigerated storage, their prolonged storage, and their high consumption frequency (FDA and FSIS, 2003). To assess the ability of L. monocytogenes to grow under refrigerated conditions in cooked meat products, growth rates are required. The growth of L. monocytogenes is described by the relationship between growth rate and temperature represented by a square root linear regression model (Ratkowsky et al., 1982). Many growth rates were collected from Combase, and it was possible to generate a linear regression model for pâte ($R^2 = 0.97$) and cooked ham ($R^2 = 0.86$). The constituents (e.g., the use of growth inhibitors or salt) and the type of packaging or package atmosphere are important parameters for the growth of L. monocytogenes. Several studies showed that modified atmosphere packaging slows down the growth of L. monocytogenes compared to air packaging or vacuum packaging (Devlieghere et al., 2001; EFSA BIO-HAZ Panel, 2018). However, since the packaging is typically opened at

home, this QMRA analysis only considered results without specific packaging conditions. This approach is considered a worst-case scenario because even during the transportation from the supermarket to home and the first few days at home, the package may remain sealed under conditions that partly inhibit the growth of *L. monocytogenes*, such as modified atmosphere packaging. Additionally, RTE cooked ham was selected as a representative for RTE cooked meat products in the QMRA model. It is important to note that in reality, there is a lot of variation in RTE cooked meat products due to differences in composition, processing and packaging.

A quantitative microbiological risk assessment (QMRA) was performed to estimate the risk per serving and predict the annual number of listeriosis cases. The model predicted an average number of 191 cases of illness per year for the total population through the consumption of RTE cooked meat products, which translates to 1.09 cases per 100,000 individuals. In comparison, the National Institute for Public Health and Environment reported an average of 92 cases of listeriosis per year in total (0.53 cases per 100,000 individuals) through the mandatory notification system in the Netherlands from 2011 to 2020 (Friesema et al., 2022). The OMRA model thus predicted a higher incidence of listeriosis cases than what was observed through epidemiological data. This may be due to the fact that risk assessment models tend to rely on worst-case scenarios, assumptions, and estimates. For example, the model does not take into account the variability of the virulence of L. monocytogenes strains, variability of susceptibility within the population, or changes in susceptibility to L. monocytogenes infection over time. Additionally, products that are stored for longer periods at higher temperatures (which are associated with higher predicted risk) are more likely to be spoiled and not consumed. Also, the presence of background flora (e.g., lactic acid bacteria) is considered an additional factor that affects the growth of L. monocytogenes, since lactic acid bacteria act as competitors and may suppress the pathogen when they reach a critical density population (Lardeux et al., 2015; Mejlholm and Dalgaard, 2015; Tsaloumi et al., 2021). Furthermore, not all packages will be opened immediately upon arrival at home, but some may remain closed in the refrigerator for a few days. On the other hand, discrepancies between the model predictions and reported incidence rates may also be attributed to underdiagnosis within microbiological surveillance systems, as has been observed for listeriosis. Scallan et al. (2011) estimated that there was a 2.1-fold factor of underdiagnosis for listeriosis. Taken all these factors into account, caution is required when interpreting the absolute results of QMRA models, and the limitations of the model inputs and assumptions should be taken into consideration. However, the outcomes derived from the QMRA model, and especially the relative effects, can be useful in evaluating key input variables and anticipating potential consequences of various scenarios related to the risk of listeriosis.

The sensitivity analysis showed that initial concentration, storage time and temperature at home were important factors that contributed to the risk of listeriosis. Most of the predicted illness cases were attributed to the storage of RTE cooked meat products for >7 days and/or at a temperature of 7 °C or above. Besides home storage, also initial concentration was an important factor that contributed to the risk of listeriosis. This was to be expected since a high initial concentration is also more likely to end up in a final concentration that can make people sick. A scenario where the RTE cooked meat products that are contaminated with *L. monocytogenes* do not exceed the 100 CFU/g at the point of sale, predicted a reduction of the illness cases by 50 %. This suggests that approximately half of the current occurrences of listeriosis cases are likely caused by foods that have a level of *L. monocytogenes* exceeding the legal limit, even already at the point of sale.

The QMRA model showed that the high-risk population accounted for >90 % of predicted illness cases (175 cases of the 191 cases in total). When assessing the impact of disease in terms of Years of Life Lost (YLL), the contribution of the elderly was 60 %. Both outcomes indicate a significant contribution of the elderly group. Targeted risk

communication about the risk of listeriosis is available in many countries, as well as in the Netherlands, for immunocompromised persons and pregnant women. Specific advice is given about (avoiding) risky foods and control measures that can reduce risks in the home kitchen (Health Council of the Netherlands, 2021). For the elderly, targeted communication to reduce the risk of listeriosis is much less available and less common. Yet, the QMRA model showed that the vast majority of illness cases and YLL are in this group and it is expected that this will even further increase (EFSA BIOHAZ Panel, 2018). The rapid aging population in the Netherlands and Europe, and the increase in chronic age-related diseases contributes to this high predicted number of illnesses (Statistics Netherlands CBS, 2021). In addition, specific foodhandling behaviours which are more common among the older generation than the younger generation (e.g., a lack of adherence to use-by dates and ineffective refrigerated storage of RTE foods) increase this risk (FAO and WHO, 2022). Our study showed that especially the more extreme high storage conditions were found in refrigerators of elderly and this indicates that there is a need for more targeted communication about good storage practices that can be taken in the home kitchen. Especially prevention of outliers can have a serious effect. Specific advice can be given to the elderly, such as setting a maximum temperature for the home refrigerator, storing RTE cooked meat products on the bottom or middle shelf of the refrigerator, and establishing a maximum storage time for opened RTE cooked meat product. From the distribution data, as shown in Fig. 5A, it can be seen that the first cases of illness occur after a storage time of 2 to 3 days. This aligns with the Dutch Health Council's recommendation for pregnant women, suggesting consumption of products capable of supporting L. monocytogenes growth within the stated shelf life date and, once opened, within 2 to 3 days (Health Council of the Netherlands, 2021). However, the exact duration for storing opened RTE cooked meat products can vary due to factors like storage temperature, the presence of preservatives or inhibitors, and the timing of product opening (Devlieghere et al., 2001; Couvert et al., 2017). Manufacturers should be encouraged to conduct appropriate challenge tests to more accurately determine the postopening storage duration based on specific product characteristics, as also stipulated in Regulation (EU) No 1169/2011 (2011). Also awareness of risky products need to be improved as mentioned in a meeting report about L. monocytogenes in RTE foods (FAO and WHO, 2022). Further research is necessary to understand how targeted communication can be effectively organized. Reducing the risk of listeriosis is a shared responsibility. Consumers must be involved, yet the manufacturer remains also an important stakeholder in reducing the risk of listeriosis for the simple reason that if *L. monocytogenes* is not present in the product, it cannot lead to any disease.

In conclusion, while the mean temperature of household refrigeration of Dutch consumers is appropriate for preserving perishable ready-to-eat products, a significant proportion of refrigerators had temperatures above the recommended value. A lack of knowledge, optimistic bias, and a lack of perceived importance of risk control may contribute to this difference. To reduce the risk of listeriosis, the importance of storing RTE foods at the recommended temperature on the bottom or middle shelf as well as consuming them within a few days after opening, should be highlighted. Raising awareness among stakeholders, especially towards older consumers who are at higher risk, is essential in decreasing the disease burden.

CRediT authorship contribution statement

Wieke P. van der Vossen-Wijmenga: Conceptualization, Formal analysis, Visualization, Writing – original draft, Methodology. Heidy M. W. den Besten: Conceptualization, Methodology, Supervision, Writing – review & editing. Marcel H. Zwietering: Methodology, Supervision, Writing – review & editing, Conceptualization.

Declaration of competing interest

The authors have no competing interests to declare.

Data availability

The used QMRA model, along with the outcomes of one simulation with 10,000 iterations and another simulation with 1,000,000 iterations, is available at https://doi.org/10.4121/98b4d5dd-7b07-48c0-bbc7-62a2f10944b1.

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Appendices. Supplementary data

Supplementary data to this article can be found online at $\frac{\text{https:}}{\text{doi.}}$ org/10.1016/j.ijfoodmicro.2023.110516.

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