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Quantitative microbiological risk assessment

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Abstract

The production of safe food is being increasingly based on the use of risk analysis, and this process is now in use to establish national and international food safety objectives. It is also being used more frequently to guarantee that safety objectives are met and that such guarantees are achieved in a cost-effective manner.

One part of the overall risk analysis procedure—risk assessment—is the scientific process in which the hazards and risk factors are identified, and the risk estimate or risk profile is determined. Risk assessment is an especially important tool for governments when food safety objectives have to be developed in the case of 'new' contaminants in known products or known contaminants causing trouble in 'new' products. Risk assessment is also an important approach for food companies (i) during product development, (ii) during (hygienic) process optimalization, and (iii) as an extension (validation) of the more qualitative HACCP-plan.

This paper discusses these two different types of risk assessment, and uses probability distribution functions to assess the risks posed by *Escherichia coli* O157:H7 in each case. Such approaches are essential elements of risk management, as they draw on all available information to derive accurate and realistic estimations of the risk posed. The paper also discusses the potential of scenario-analysis in simulating the impact of different or modified risk factors during the consideration of new or improved control measures.

Keywords: Quantitative risk assessment; *Escherichia coli* O157:H7; Raw fermented sausages

1. Introduction

Risk analysis, which aims to protect the consumer by setting appropriate food safety objectives (Fig. 1a), is recommended by the World Trade Organisation (WTO) as the most appropriate means of ensuring the production of acceptable safe food. However, examination of a number of published formal risk assessments (Notermans et al., 1997; Whiting and Buchanan, 1997; Cassin et al., 1998; Marks et al., 1998) indicate that, to be successful, this approach requires considerable effort.

A method for risk assessment has been described by the Codex Alimentarius Commission (CAC), and is comprised of three major elements, some of which contain a number of components.

(a) Risk assessment. Risk assessment contains four components:

- hazard identification, in which contaminants are identified as specifically as necessary;
- hazard characterization, in which the health effect of each contaminant is determined, frequently by assessing the dose–response relation;

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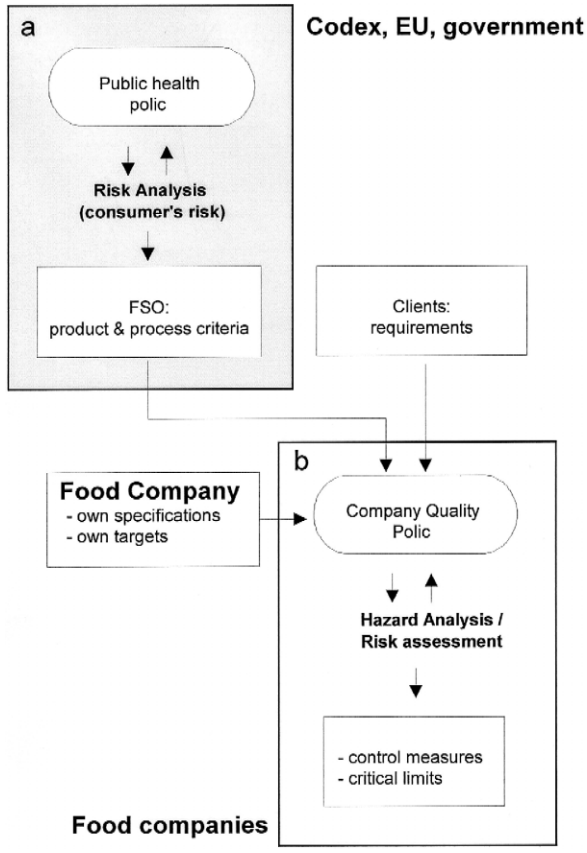

Fig. 1. Risk analysis on two levels: consumer's risk (a) and company's risk (b).

- exposure assessment, in which the probability of intake by the consumer is estimated;
- risk characterization, in which the risk is calculated as the product of exposure (intake) and dose–response estimate (effect).

(b) Risk management. In this element, the risk is evaluated and a decision can be made about the accepted risk within the wider framework of public health objectives (food safety objectives). Options for improvement are considered and new or modified criteria are eventually laid down in guidelines, regulations or legislation.

(c) Risk communication. This involves transparent communication between risk assessors and risk managers, which is important, because they have different interests. Finally, the results of risk assessment and risk management are communicated more widely with the relevant links in the food chain, up to, and including, consumers, by such means as specifications and labelling.

Completion of such a formal risk analysis may take years. To avoid the difficulties and delays caused by requiring all food producers will carry out individual risk analysis for all their products, the WTO also accepts the use of internationally established criteria. Such criteria should be based on previous risk analysis processes, in which international organisations such as the CAC and national governments act as risk managers.

Once food safety objectives have been defined, food companies have to translate these objectives into criteria, etc. that apply to their processes and/or products. To achieve compliance, food companies can take a simple approach, by estimating the probability of occurrence of contaminants in end products,

which gives an assessment of the food companies risks, e.g. of exceeding the criteria set by governments, clients or the company itself. This approach, focussing on assessing the occurrence of contaminants in end products, does not involve hazard characterization (dose–response), or consider the amount of product that might be consumed. These two factors are very complicated, and cannot be effectively assessed by food companies. On the other hand, they are very important for governments in relation to setting food safety objectives. When a food company carries out a risk assessment, the factors contributing to a certain risk will be prioritised and critical limits will be set in order to meet the criteria and specifications (Fig. 1b). Cost-benefit analysis can also be carried out, to assess the economical impact of possible improvements. This approach is in some ways similar to the hazard analysis element of the Hazard Analysis Critical Control Point (HACCP)-plan. However, the systematic method of risk assessment is much more extensive and quantitative in nature. While the process focuses on food safety, i.e. the prevention or control of pathogens, food spoilage also poses important challenges to food companies, e.g. leading to claims, recalls, etc. A systematic risk assessment can include non-pathogenic spoilage microorganisms, underpinning the prediction and extension of product shelf life, within an overall process to food safety and food spoilage problems.

2. Identification and quantification of risk factors

The identification of risk factors is an important and early step in risk assessment procedures. Risk factors contribute to the risk of occurrence of a foodborne hazard. It may contribute to the introduction, increase or decrease of the hazardous agent. Risk factors are influenced by the quality of raw materials, steps within the process environment, as well as the composition, packaging and storage conditions of the final product. When such a method of collecting and analysing information on the characteristics of contaminants, and conditions leading to the food safety risks had been applied, control measures necessary to reduce a risk to acceptable levels can be determined. The impact or the effect of a risk factor can be quantitatively determined using worst-case or statistical approaches.

The worst-case approach considers a succession of extreme situations in the process, under which an improbable series of unfavourable events could occur simultaneously, leading to loss of adequate levels of product safety. If the results from such worst-case analysis still show the product quality to be within the specifications, the product can be considered as safe. In other cases, the results should be subjected to further analysis as the worst-case analysis is by definition always an overestimate of the likely risk.

The statistical approach incorporates results, expert knowledge, literature data and well-reasoned assumptions about the various risk factors into probability distribution functions. This means that calculations are not based on one value, e.g. the average or the extreme (worst-case) value, but on several different values drawn from the probability distribution functions, by Monte Carlo sampling. There are a number of statistical software programmes (e.g. @RISK by Palisade) which can be used to link the probability distribution function for the different process steps.

By definition, a worst-case approach always overestimates the likely situations because the probability of simultaneous occurrence of unfavourable circumstances in relation to every risk factor is very low. Therefore, a statistical approach provides a more likely analysis and clearer insight into the need for process improvements (and the effects of such measure), than a worst-case approach does.

There are different aspects of improvement in product safety, reflecting the fact that risk is made up of uncertainty and variability. *Uncertainty*, arising from lack of sufficient or reliable data, can be reduced by collecting more reliable data, for example, by means of product storage and challenge-testing. Such trials and test procedures yield important data on the extent to which levels of contaminants increase or decrease during the manufacturing process and subsequent storage. *Variability*, on the other hand, can occur when there is sufficient data, but there is variation among these data. Variability can be reduced by improved process control and intervention. Moreover, such interventions may bring about a new situation, for example, in the introduction of alternative ingredients or inclusion of a modified or additional process step. When risks have been calculated based on probability, better choices can be

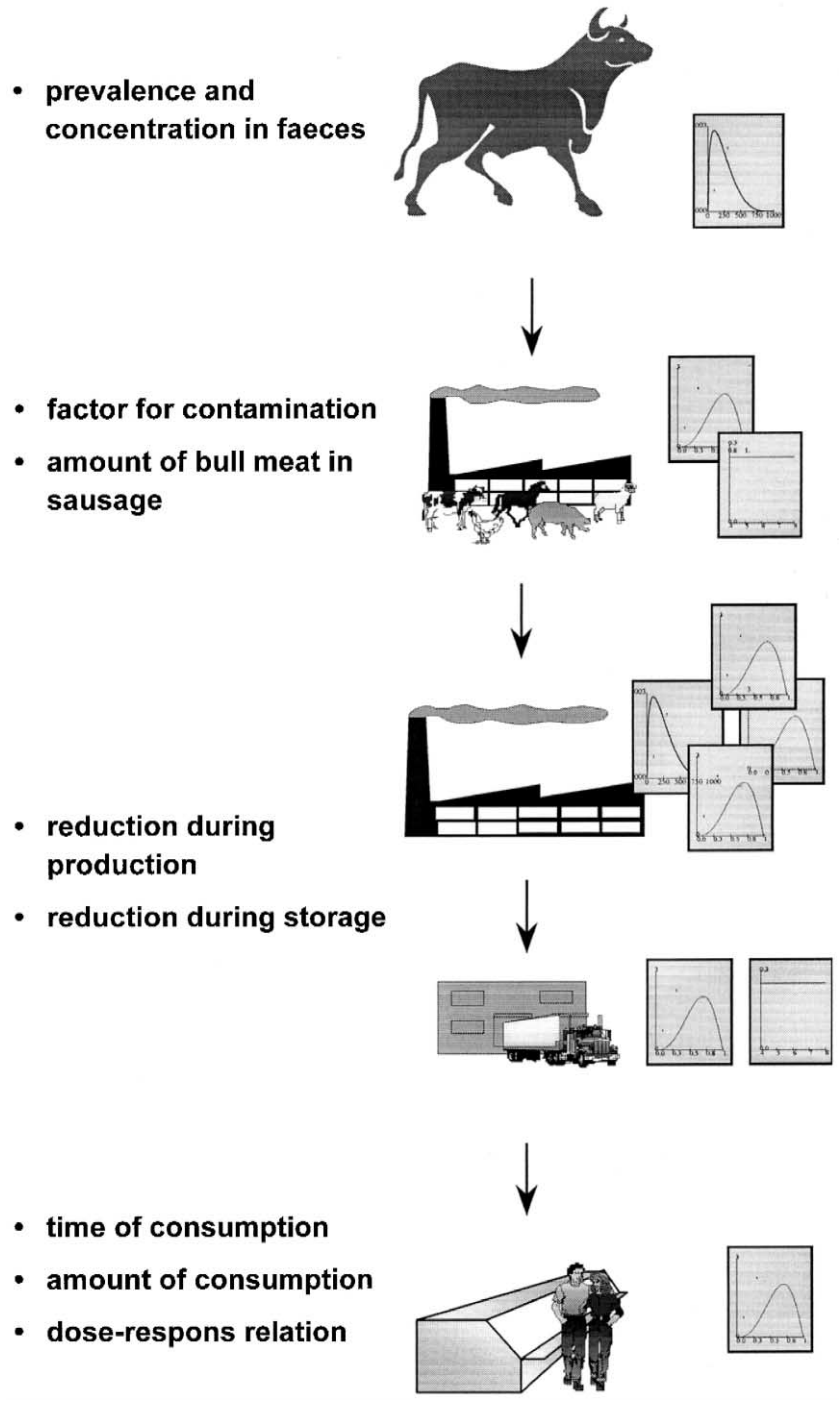

Fig. 2. Risk factors in the production chain for raw fermented sausages.

made among a number of possible means of achieving improvements, taking into account the costs involved.

3. Quantitative risk assessment of *Escherichia coli* O157:H7 used to set criteria

3.1. Results of a practical example

The objective of this risk assessment was to see if a criterion should be set for the reduction of *E. coli* O157:H7 during the production of raw fermented sausages. Quantitative risk assessment integrating data from the literature, challenge tests, other microbiological information and assumptions, combined with the use of applied statistics, was used to complete a risk assessment for two different types of sausages.

The process began with identification of the risk-contributing factors in the food chain (Fig. 2), followed by quantification of the impact of each risk factor using probability distribution functions. Statistical calculations were made including the distribution of the pathogen on the beef meat, the amount of beef meat in the raw material mix, and the distribution of the pathogen during portioning into sausages. Challenge-tested sausages were fermented and stored, during which time a reduction was observed (Fig. 3). The reduction is around 2–3 D. The numbers at day 35 were between 0 and 2 log (negative with counting method, positive after enrichment of 1 g). The numbers at day 78 were between −1.4 and 0 log (negative after enrichment of 1 g, positive after enrichment of 25 g).

Finally, the probability density of *E. coli* O157:H7 in the product at the time of consumption was calculated (Fig. 4). The numbers presented in Fig. 4a–c represent meat from bulls positive for *E. coli* O157:H7 at different stages of the process. It can be observed that not only the numbers of *E. coli* O157:H7 are reduced (*X*-axis) but also the probability that the numbers occur is reduced (*Y*-axis). In Fig. 4d, which relates to meat from both positive and negative bulls, the expected numbers of *E. coli* O157:H7 in all sausages are presented. This probability of occurrence was multiplied by the amount of consumption to assess the likely exposure of the final consumer.

3.2. Some remarks

The probabilistic calculation presented allows a risk manager to decide whether or not control measures, or additional control measures, need to be developed. USDA regulations require that the overall process must be capable of achieving a 5 $\log_{10}$ units reduction in *E. coli* O157:H7 (a performance factor

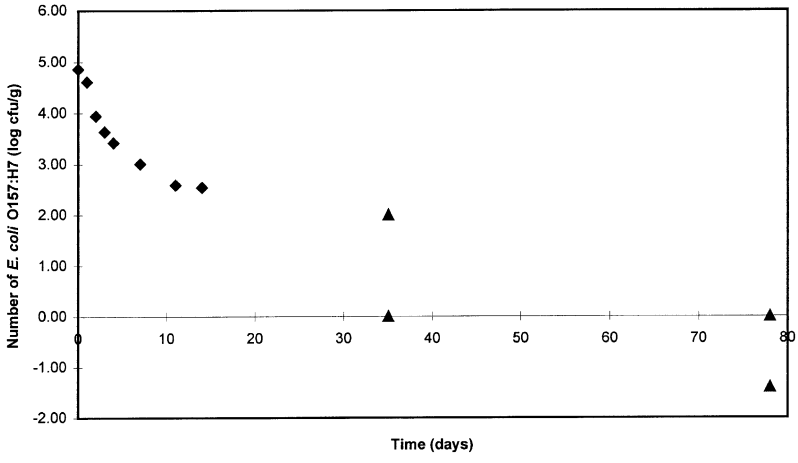

Fig. 3. Reduction of *E. coli* O157:H7 numbers inoculated into raw fermented sausage to an initial concentration of 5 log cfu/g.

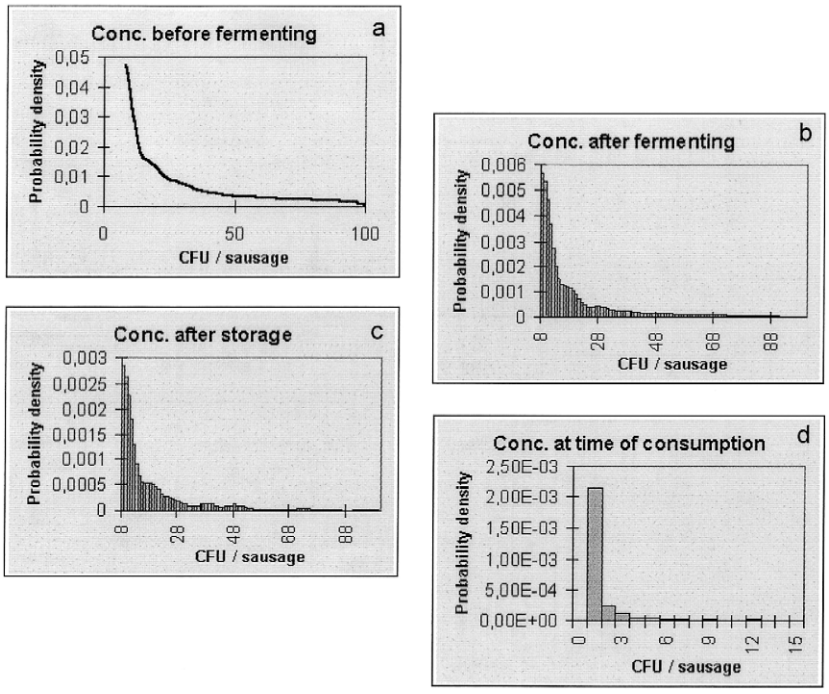

Fig. 4. The probability density of *E. coli* O157:H7 in the product at different stages of the production process.

of 5D) in raw fermented sausages. The requirement for a 5D reduction is a based on the highest number of *E. coli* O157:H7 ever determined on a beef carcass, which is 3 $\log_{10}$ units (Marks et al., 1998) and the requirement for the final sausage to contain less than one *E. coli* O157:H7/100 g. As demonstrated by the results of the challenge test, only a 2–3 $\log_{10}$ reduction was obtained. As a consequence, a 5D process performance was not achieved, and it may be necessary to introduce a heating step within the heating process, to attain a 5D performance overall. Based on identical studies, Riordan et al. (2000) have also proposed a heat treatment for an identical type of sausage. However, the approach of the USDA is a typical example of a worst-case risk assessment. Surveys carried out in Netherlands indicate that around 1% of all beef is contaminated with *E. coli* O157:H7, although there is wide variation among carcasses and meat samples (Heuvelink et al., 1999). Because of this, the distribution of the pathogen within batches of sausages will be very heterogeneous, with considerable variations in the rates of prevalence and in the concentrations of the pathogen. If this more realistic distribution is factored into risk assessment calculations, rather than the above worst-case value of 3 $\log_{10}$ *E. coli* O157:H7 per gram, a different picture will result. In the event that the process achieving a much smaller reduction in pathogen numbers, (2–3 D) during fermentation and storage of the sausages, 0.3% of the sausages could be expected to be positive for *E. coli* O157:H7 and only 0.002% of the sausages could contain more than 10 *E. coli* O157:H7. This more realistic type of risk assessment gives more information and provides a better basis for the setting of food safety criteria. The dose–response relationship, bearing in mind its variations, can be taken into account, to estimate the number of people likely to become ill after consuming the above raw fermented sausage products. The risk assessment presented is based on literature data of Cassin et al. (1998), a few challenge tests and a number of assumptions in relation to the underpinning microbiology. Thus, there is a need to gather more information about the probability of pathogen occurrence in, and pathogen reduction within, different types of raw fermented

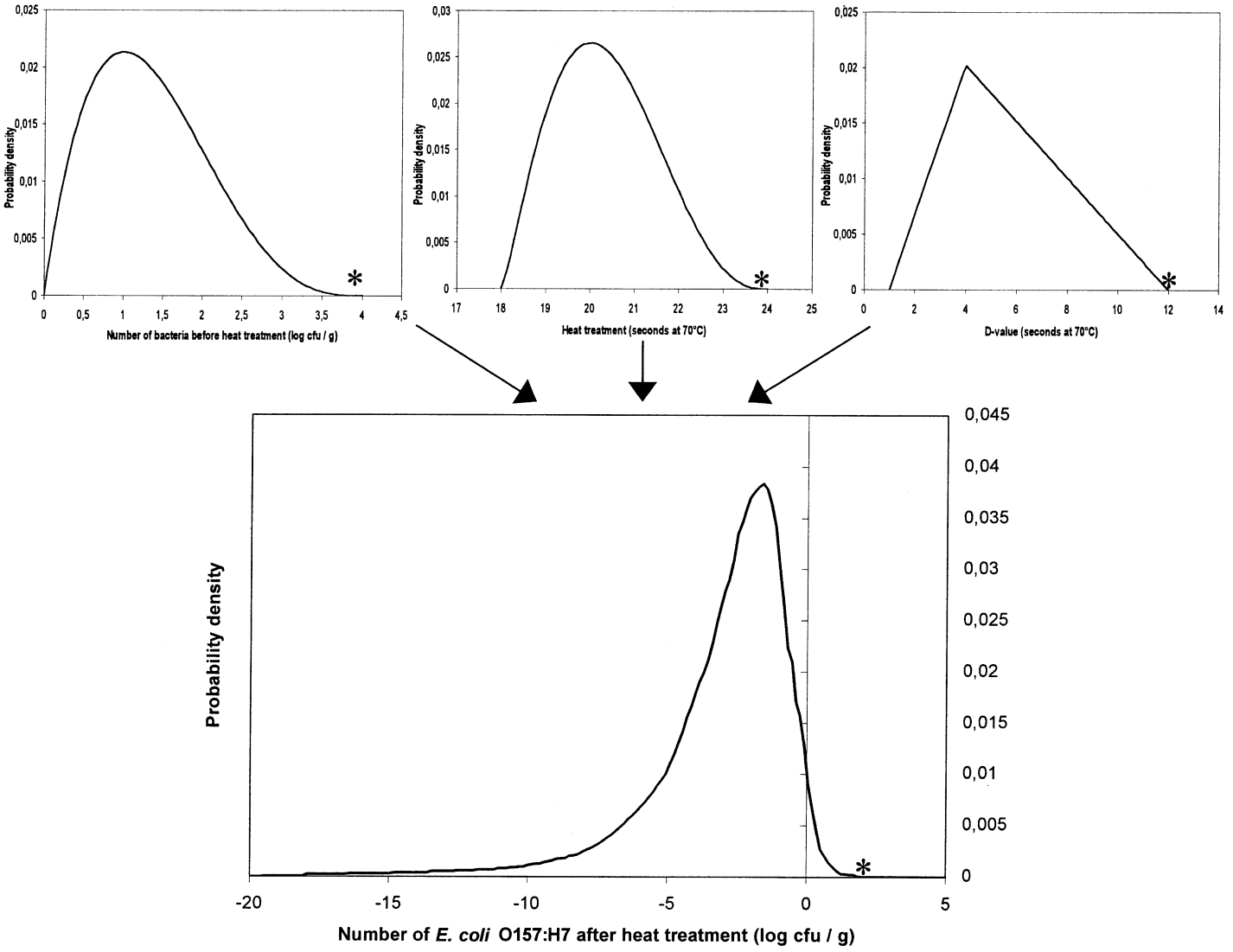

Fig. 5. Monte Carlo sampling: integrating three risk factors (initial contamination, heat treatment (*P*-value) and *D*-value) to estimate the number of *E. coli* O157:H7 present after heat treatment of 2 s at 70°C.

sausages, before a risk manager could decide what additional or alternative control measures need to be taken. It may not be necessary for the fermentation process (with or without a heating step) to achieve a 5D reduction if such a reduction was already being achieved within the existing food chain.

4. Quantitative risk assessment of *E. coli* O157:H7 used to meet criteria set

The objective of this risk assessment was to determine how a food company which produces a pasteurised meat product could achieve a process criterion of a 5D reduction of *E coli* O157:H7. The food company also wanted to know the probability that a 25-g sample of the product would be positive for *E. coli* O157:H7. As previously noted, the occurrence of this pathogen on the raw materials for sausage manufacture is very variable.

Within the fraction of the raw materials that does contain the pathogen (assumed to be 1%), any heat treatment (pasteurization) will entail some reduction in *E. coli* O157:H7 numbers. The required pasteurization time (to achieve a 5D reduction) can be simply derived using the *D*-value at 70°C with the highest probability, i.e. 4 s (Doyle and Schoeni, 1984; Juneja et al., 1997; Food MicroModel V3.02, 1999). This indicates that a treatment time of 20 s at 70°C is required. Next, the variations in initial numbers, *D*-values and pasteurization time (*P*-value) are taken into account by the introduction of probability density functions for each parameter. To assess the expected number of surviving bacteria, values were drawn from the probability density functions leading to a mean process performance, a mean number of surviving *E. coli* O157:H7 bacteria and the calculation of the probabilities that *E. coli* O157:H7 numbers in the product will exceed specified limits after heat treatment (Fig. 5).

Table 1
Calculated numbers of *E. coli* O157:H7 after heat treatment all for the positive fractions (1%)

	Minimum	Mode	Maximum
Numbers before heating ($\log_{10}$ cfu/g)	0	1	4
$D_{70°C}$ (s)	1	4	12
Pasteurization time ($P_{70°C}$) (s)	18	20	24
	Best-case[a]	Mode[b]	Worst-case[c]
Numbers after heating ($\log_{10}$ cfu/g)	−24	−4	2.5

[a] Favourable values occurring together.
[b] Most likely values.
[c] Unfavourable values occurring together.

Table 2
Effects of alternative treatments on the probability that more than one *E. coli* O157:H7 would occur in a 1 g and 25-g sample of product

	70°C; 20 ± 4 s	70°C; 24 ± 1 s	72°C; 14 ± 1 s
Decimal reduction (D)	5	6	7
prob. numbers > 0 $\log_{10}$ cfu/g	3.1%	1.1%	0.4%
prob. numbers > −1.4 $\log_{10}$ cfu/g	0.15	0.09	0.04

The difference between the use of probability functions compared to the best-case and worst-case scenario for the inactivation of *E. coli* O157:H7 during a heat treatment is demonstrated in Table 1. The worst-case values are used to calculate a concentration of 2.5 $\log_{10}$ *E. coli* O157:H7 in 1 g of end product. However, the probability that all these unfavourable events will occur simultaneously is very small. This result is totally different from the mode of the probabilistic approach of −4 $\log_{10}$ *E. coli* O157:H7. The probability that the number of *E. coli* O157:H7 is exceeding −1.4 $\log_{10}$ (positive in 25 g) is assessed at 15% for positive fraction, which makes 0.15% for the total batch (Table 2). Although the objective is a 5D reduction, there is still a chance that some products may be positive because of variance and uncertainty in raw materials, *D*-values and pasteurization time.

A number of measures to improve the situation have been proposed. These include two proposed by the food company: (1) a longer heat treatment with a less temperature variation (70°C, mode 24 ± 1 s), and (2) a higher temperature, shorter time treatment (72°C, mode 14 ± 1 s). Recalculation of the second option to 70°C with the *z*-value conservatively assumed to be 6.5°C gives an equivalent treatment

profile of 28 ± 2 s at 70°C. The two measures proposed above result respectively in a 6D and 7D process performance. The results are presented in Table 2.

5. Discussion

The use of probability distribution functions allows the assessment of the probability that *E. coli* O157:H7 concentrations will exceed certain specified values. The quantification of risk factors identifies cases where there is a need for greater control in relation to raw material quality and processing. Scenario-analysis can be used to show the impact of such interventions, and can also indicate areas where better control is needed. The application of the methods discussed in this paper can provide very useful information, demonstrating that in the examined case, the probability of detecting *E. coli* O157:H7 in a 25-g sample of sausages processed under the original procedures was 0.15% (Table 2). The probability of such detection in sausages subjected to the longer and more controlled heating process was 0.09% and the probability of such detection in sausages subjected to the higher temperature treatment was 0.04%. Provision of this type of information allows the risk manager to decide whether to change the process, depending on the levels of safety required. Such information also allows the accurate prediction of the likely impact of other measures that could be taken to improve levels of safety, such as selection of better quality raw materials or additional decontamination process and new preservation techniques. The ability of the risk assessment approach to provide such information clearly indicates that it goes beyond the more qualitative HACCP-approach, and is an essential tool in the management of food safety problems and in cost effective and efficient product and process development.

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