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Effectiveness of an increasingly stringent microbiological process hygiene criterion to control *Campylobacter* in broiler carcasses

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ABSTRACT

Increasing the stringency of microbiological criteria is a risk management measure that can improve food safety and hygiene, but its adoption by governments around the globe is limited. In 2018, a hygiene criterion for *Campylobacter* in broilers was originally set by the European Commission, which intended to progressively increase its stringency in 2020 and in 2025. In this study, the effects of this regulation on the level of (non-) compliance were estimated based on baseline data on *Campylobacter* for the different European Union (EU) Member States and associated countries in 2008. Qualitative and quantitative baseline data were used to estimate concentration distributions, from which the levels of compliance with the legal limits were determined, making use of the ICMSF sampling plan spreadsheet and considering different batch properties (i.e., standard distributions of microorganism) and different non-compliance detection probability values. Based on the 2008 baseline data, the performance of the criterion is estimated to target the reduction of the mean log contamination level to about 0.25 and 0.5 log (factor 2 and 3 in arithmetic concentration) and to about 0.5 and 1.1 log (factor 4 and 12 in arithmetic concentration) in case the *c*-value decreases from 20 to 15 and from 20 to 10, respectively. Assuming a compliance level in practice of at least 98%, more and more food business operators in EU Member States would fail to meet this level as a consequence of the increasingly stringent criterion. This analysis clearly shows that a timewise push to further improve hygiene standards will be needed in various countries for their food businesses to be able to achieve a high level of compliance with the progressively stringent EU *Campylobacter* process hygiene criterion.

1. Introduction

Food safety needs to be managed by the private sector using a system-based approach with appropriate prerequisite programs such as Good Hygienic Practices (GHP) and, where necessary, a targeted Hazard Analysis and Critical Control Point (HACCP) programme that includes validated critical control points (CCPs) and monitoring of critical limits for CCPs. If this food safety management system is working as intended, then food safety should be under control. Nevertheless, periodic verification is needed, for example, by microbiological testing. Competent authorities may issue quantitative limits for microorganisms as targets for the private sector that reflect the level of food safety expected in market or in international trade. The Codex Alimentarius advocates the use of microbiological criteria to help verify that the limits for the presence of particular pathogens or indicators of process control are met along food supply chains (CAC, 2013). Microbiological criteria can be developed for end products, ingredients, and processes, as well as for the production environment. In European Union (EU) legislation, two types of microbiological criteria are defined: food safety criteria and process hygiene criteria (EC, 2005). A food safety criterion defines the acceptability of a product or a batch of foodstuff and is applicable to products intended to be placed on the market or already on the market. Where a food safety criterion is not complied with, the product should not be placed on the market or should be withdrawn from the market. A process hygiene criterion is a criterion indicating the acceptable functioning of the production process. Such a criterion is not applicable to products

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already placed on the market, but rather to a food processing operation. It sets an indicative contamination value above which corrective actions concerning the processing operation are required in order to maintain the hygiene of the process in compliance with the food law. These corrective actions do not prevent release or impose a recall of released product, but call for improvements in slaughter hygiene, review of process controls, and a review of the origin of animals and the biosecurity measures in the farms of origin (EC, 2005). The utility of microbiological criteria as part of useful testing in food safety management and as risk-based metrics for food safety control has been described by the International Commission on Microbiological Specifications for Foods (ICMSF, 2011, 2018).

It should be realised that microbiological criteria are statistical verification tests, typically set to ensure sufficient consumer safety protection when compliance rates to such legal limits are high. They are not targets, nor acceptable or appropriate levels of organisms in their own right, but rather levels against which verification testing can be done by government and industry of the food safety management system deployed in the production of the food at hand. In practice, in order to keep the producer risk low, industry needs to ensure that it sets its own targets for compliance at a much lower level than those that are needed to just meet the legal criteria. If legal criteria are not complied with, the product can be considered unsafe (food safety criteria) or the process can be considered to be not under control (process hygiene criteria). That a product or process complies with the criteria in itself does not guarantee product safety or that the process is consistently under control. The confidence in product safety and process control should come from data on the combination of key aspects that provide confidence in the food safety management system, such as its design, validation of control measures, day-to-day execution, and microbiological verification and trend analysis.

Various types of microbiological criteria established by the Codex Alimentarius or governments are presented in Table 1 with illustrative examples. Food safety criteria frequently apply to pathogens in the food product, while process hygiene criteria may also apply to utility or indicator organisms (ICMSF, 2018). However, there are exceptions such as generic *Escherichia coli*, although not a pathogen per se, in live molluscs (EC, 2005), which is applied as a food safety criterion. In addition, there are several process hygiene criteria in which a pathogen is tested for to assess control over the food processing, like the EU criterion for *Salmonella* in broiler carcasses in which $c \neq 0$ (with c being the number of positives allowed), and thus a few *Salmonella* positive samples may be acceptable. When this limit for broilers was initially introduced in the EU in 2006, the c-value was 7 (taking a total number of samples $n = 50$, over 10 weeks with 5 samples per week), but this was reduced to $c = 5$ in 2012. Important for Food Business Operators (FBOs) is that the sampling frequency is allowed to be reduced (fortnightly) when satisfactory results have been obtained over a longer period (30 weeks) or if an appropriate control program is in place. In this manner, the stringency of the criterion can be increased over time, but the testing frequency and hence the burden on the FBO can be reduced if additional information is available to underscore good performance.

High standards of food safety and communication with stakeholders about the stringency of control required to comply with food safety expectations of governments is a general public health protection goal. For instance, the United States Department of Agriculture's Food Safety and Inspection Service (USDA-FSIS) launched a regulation in 1996 involving a testing program for *Salmonella* and generic *E. coli* and introducing a more stringent policy of pathogen reduction and the implementation of HACCP. Over the years, the testing program has undergone refinements and updates to reflect advances in scientific knowledge, technological capabilities, and changing industry practices. Over time, the stringency was further increased by successive regulations and evidence of successful reduction of *Salmonella* prevalence in poultry and meat products, as was documented by Ebel and Williams (2020) and Williams et al. (2022), respectively. Rarely have governments actually implemented a planned stringency increase in a single regulation.

In 2017, the European Commission (EC) issued regulation in which a new *Campylobacter* process hygiene criterion for broiler carcasses was introduced that has similar characteristics to the *Salmonella* criterion, being a process hygiene criterion with $n = 50$ and $c \neq 0$, starting initially with $c = 20$ in 2018, but decreasing in time to $c = 15$ in 2020 and to $c = 10$ in 2025, as indicated in Table 2 (EC, 2017). The regulation allows for the sampling frequency to be reduced where satisfactory results have been obtained over a longer period, or if an appropriate control program is in place. Unlike the *Salmonella* criterion (not detected in 25 g, i.e., qualitative), this plan has a quantitative microbiological limit (m = 1000 cfu/g). In case the process hygiene criterion is not met, corrective measures need to be taken to improve slaughter hygiene and/or farm biosecurity.

According to the European Food Safety Authority (EFSA) and the European Centre for Disease Prevention and Control (ECDC), campylobacteriosis is the most commonly reported zoonosis in 36 European countries (28 Member States and eight non-Member States, i.e., associated countries), based on monitoring activities carried out in 2019 (EFSA & ECDC, 2021). With 220,682 reported cases and representing 50% of all reported zoonosis cases in 2019 (for the 13 zoonoses presented in the report), campylobacteriosis has been holding this lead position in Europe since 2005. Notably, in 2011, EFSA issued advice on reducing *Campylobacter* in chicken meat, which included recommendations for pre-slaughter interventions that could reduce the risk to public health by 50%, meat production measures that could reduce public health risk by 90% or more, and an evaluation of the effectiveness of achieving particular prevalence reduction targets (EFSA BIOHAZ Panel, 2011). This opinion was recently updated (EFSA BIOHAZ Panel et al., 2020). These opinions focus a lot on the effectiveness of on-farm and in-chain intervention measures when implemented by FBOs. A

Table 1
Examples of various types of sampling plans for foodborne pathogen or hygiene control.[a]

Class	Type[b]	$c = 0$ or $c \neq 0$	Example[c]	n	c	m	M	Reference
2	Qualitative	0	• ***Salmonella* in PIF**	60	0	n.d./25g		CAC (2008)
			• ***Listeria monocytogenes* in RTE foods supporting its growth**	5	0	n.d./25g		CAC (2007)
2	Qualitative	$\neq 0$	• Enterobacteriaceae in PIF	10	2	n.d./10g		CAC (2008)
			• *Salmonella* in broiler carcasses	50	5	n.d./25g		EC (2011)
2	Quantitative	0	• ***Listeria monocytogenes* in RTE foods not supporting its growth**	5	0	100/g		CAC (2007)
2	Quantitative	$\neq 0$	• *Campylobacter* in broiler carcasses	50	20,15,10	1000/g		EC (2017)
3	Quantitative	$\neq 0$	• Mesophiles in PIF	5	2	500/g	5000/g	CAC (2008)
			• ***Escherichia coli* in molluscs**	5	1	230/100g	700/100g	EC (2015)

[a] n = number of sample units, c = acceptable number of sample units giving values between m and M (3-class plan) or above m (2-class plan), m = lower microbiological limit, M = upper microbiological limit.

[b] Qualitative is a detected or not detected (n.d.) criterion in a given sample volume, for example m = n.d./25 g or n.d./10g. Quantitative is a quantitative test where there is a quantitative limit, for example m = 100 cfu/g.

[c] Bold: food safety criterion; not bold: process hygiene criterion. PIF is powdered infant formula, RTE is ready-to-eat.

Table 2
Process Hygiene Criterion for *Campylobacter* in broiler carcasses.[a]

Food category	Micro-organisms/their toxins, metabolites	Sampling plan		Limits		Analytical reference method	Stage where the criterion applies	Action in case of unsatisfactory results
		n[b]	c[c]	m	M			
2.1.9 Carcasses of broilers	*Campylobacter* spp.	50	20	1000 cfu/g		EN/ISO 10272-2	Carcass after chilling	Improvements in slaughter hygiene, review of process controls, of animals' origin and of the biosecurity measures in the farms of origin

[a] As stipulated in EC Regulation 2073/2005 (EC, 2005) and updated by EC Regulation 2017/1495 (EC, 2017), coming into force 1.1.2018.
[b] The 50 samples shall be derived from 10 consecutive sampling sessions in accordance with the sampling rules and frequencies laid down in EC Regulation 2017/1495 (EC, 2017).
[c] From 1.1.2020 onward $c = 15$; from 1.1.2025 onward $c = 10$.

possible strategy for governments to achieve a reduction of *Campylobacter* contamination levels is to establish microbiological criteria at one or more specific points in the farm-to-fork poultry products supply chain as benchmarks for operational level interventions. Quantitative Microbiological Risk Assessment has been used as a tool to derive risk-based microbiological criteria (Nauta et al., 2012; Nauta & Havelaar, 2008) and the impact of such criteria on *Campylobacter* contamination in broilers has been published (Lee et al., 2015; Reich et al., 2018).

In this paper, the performance of the process hygiene criterion for *Campylobacter* in broiler carcasses (EC, 2005, 2017) is quantified, taking into account the progressively lowered c-values that increase the stringency of control expected from FBOs. Furthermore, how this plan works to decrease pathogen levels is illustrated by using the 2008 baseline data for the EU and two associated countries on the prevalence and concentration of *Campylobacter* in poultry (EFSA, 2010).

2. Methods

2.1. Statistics of the performance of a sampling plan

To evaluate the performance of sampling plans, one needs to investigate three relevant statistical phenomena (Jongenburger et al., 2015). The first one is the actual spatial and statistical distribution of microorganisms in the food product. Microorganisms can be fully mixed within a product, but generally there will still be a level of inhomogeneity or clustering. In many cases, it is assumed that microorganisms are log-normally distributed in the food product, i.e., the log concentration is normally distributed. Since microbial growth and inactivation can occur in an exponential manner and since actual levels can differ by orders of magnitude, this distribution is often suitable and works well in practice. The second aspect is the statistical process of taking a sample unit and the sample being either defective or not. For a qualitative test, this is the probability that at least one organism is present and detected in a sample unit, and for a quantitative test it is the probability of the concentration being above the m-value in the sample unit. The third aspect is that the sampling plan consists of several sample units (n) and a certain number of these samples (c) are allowed to test positive or are allowed to be higher than the microbiological limit (m). The latter process may follow a binomial distribution, since it can be assumed that all samples are independent and have an equal probability to result in a positive outcome. These three aspects together can be used to describe the performance of a sampling plan. In the case of the *Campylobacter* criterion, we could assume for the first aspect that the concentration C of *Campylobacter* is log-normally distributed, with a certain mean, $\mu_{\log C}$ (i.e., log geometric mean) and a particular standard deviation, $\sigma_{\log C}$. The second aspect, the probability that one sample is defective, can be determined as follows (Jongenburger et al., 2015):

$$P_{\text{defective}} = \text{normal}(\log C > m, \mu_{\log C}, \sigma_{\log C}) = 1\text{- normal}(\log C \leq m, \mu_{\log C}, \sigma_{\log C}) \quad (1)$$

where $P_{\text{defective}}$ is the probability that a sample is defective (assuming that $P_{\text{defective}}$ equals the proportion of defective products in the batch), $\log C$ is the log microbiological concentration (log cfu/g), m is the microbiological limit (log cfu/g), $\mu_{\log C}$ is the mean of the normal distribution of $\log C$, $\sigma_{\log C}$ is the standard deviation of the normal distribution of $\log C$.

The third aspect is the process of taking n samples and the probability that the criterion is complied with, i.e., the probability that no more than c samples are allowed to be positive. This can be determined as:

$$P_{\text{compliant}}(c, n, P_{\text{defective}}) = \text{binomial}(k \leq c, n = n, P = P_{\text{defective}}) \quad (2)$$

where $P_{\text{compliant}}$ is the probability that the criterion is complied with, c is the number of samples allowed to be positive (i.e. allowed to be higher than m), n is the number of samples and k is the numerical outcome. It should be noted that *defective* ($P_{\text{defective}}$) relates to one sample unit and that *compliance* ($P_{\text{compliant}}$) relates to the full sampling plan, in which all n samples are considered.

2.2. Performance of the EU criterion for Campylobacter in poultry with increasing stringency

The performance of a sampling plan can be defined as the mean log concentration, $\mu_{\log C}$, that is detected with a certain probability, for example 95% (Zwietering et al., 2014). In that case, the probability to comply with the criterion ($P_{\text{compliant}}$) is 5%. This performance stringency depends on the number of samples (n), the number of samples allowed to be positive (c), the standard deviation of the log concentration ($\sigma_{\log C}$), and the microbiological limit (m). The $\sigma_{\log C}$ can, for example, be assumed to be 0.4 for well mixed products, 0.8 as a generally applicable value, and 1.2 for products characterised by a more heterogenous distribution of microorganisms. Here, we develop scenarios, based on $\sigma_{\log C} = 1$ and $\sigma_{\log C} = 2$ because chicken products are likely characterised by highly heterogenous distributions of microorganisms. These values are in line with reported standard deviations for *Campylobacter* contamination levels on chicken carcasses (neck and leg skin samples) in France in different seasons of the year (Duqué et al., 2018). The performance of a specific sampling plan can be determined making use of the ICMSF sampling plan tool (ICMSF, 2020), but can also be determined by the following equations:

$$P_{\text{compliant}}(c, n, P_{\text{defective}}) = \text{binomial}(k \leq c, n = n, 1\text{- normal}(\log C \leq m, \mu_{\log C}, \sigma_{\log C})), \quad (3)$$

$$P_{\text{non-compliant}}(c, n, P_{\text{defective}}) = 1\text{ - binomial}(k \leq c, n = n, 1\text{- normal}(\log C \leq m, \mu_{\log C}, \sigma_{\log C})), \quad (4)$$

and for this specific *Campylobacter* criterion when $c = 20$, $n = 50$ and $m = 3$ log cfu/g

$$P_{\text{compliant}}(20, 50, P_{\text{defective}}) = \text{binomial}(20, 50, 1\text{- normal}(\log C \leq 3, \mu_{\log C}, 1.0)) = 0.05, \quad (5)$$

in which a batch with a specific mean log concentration $\mu_{\log C}$, and a $\sigma_{\log C}$ of 1.0 log has a 5% probability of being compliant when 50 samples are taken and with only 20 samples being accepted to be higher than m (i.e., 1000 cfu/g, which equals 3 log cfu/g). This batch has thus a 95% (100%

minus 5%) probability of being non-compliant. Should more than 20 samples be above *m* and thus a batch be non-compliant, then the FBO should take actions to make further batches of product compliant.

2.3. Arithmetic mean

The log arithmetic mean ($log_{10}(\overline{C})$) of a log-normal distribution can be determined from the log geometric mean ($\overline{log_{10}\, C}$) and the standard deviation ($\sigma_{log_{10}\, C}$) as (Jongenburger et al., 2015):

$$log_{10}(\overline{C}) = \overline{log_{10}\, C} + 0.5 \bullet \ln(10) \bullet \sigma^2_{log_{10}\, C} \quad (6)$$

3. Results

3.1. Performance of the Campylobacter sampling plan in theory

The theoretical performances of the *Campylobacter* sampling plan with progressively higher stringency are reflected in the pre-defined values of *c* at 20, 15 and 10 that are mandated from 2018, 2020 and 2025, respectively, are presented in Table 3. For illustrative purposes, two standard deviation values were selected (i.e., $\sigma_{logC} = 1$ and $\sigma_{logC} = 2$) next to two non-compliance probability values (i.e., 95 and 99%).

From Tables 3 and it can be seen that both the choice of the standard deviation (σ) and the selected non-compliance probability ($P_{non\text{-}compliant}$) have an effect on the performance stringency. However, generally, one can conclude that the mean log concentrations (μ_{logC}) are not that dissimilar for different values of σ and $P_{non\text{-}compliant}$, and that, depending on the standard deviation, the higher stringency will have μ_{logC} values 0.26 to 0.52 log cfu/g lower [factor 2 (i.e., $10^{0.26}$) to 3 (i.e., $10^{0.52}$) in arithmetic concentration] when going from $c = 20$ to $c = 15$, while a change from $c = 20$ to $c = 10$ would reduce the μ_{logC} with 0.55–1.1 log cfu/g (factor 4 to 12 in arithmetic concentration).

Let us assume that all slaughterhouses in all Member States and associated countries work at an operational non-compliance level of no more than 1% with $c = 20$ and $\sigma_{logC} = 2$. When achieved, this would mean that only in 1% of the sampling rounds the sampling outcome is not according to the criterion. To remain at the level of no more than 1% non-compliant when the criterion becomes progressively more stringent (i.e., going to $c = 15$ and to $c = 10$), the slaughterhouses would need to introduce appropriate interventions. Appropriate interventions would be those that help reduce the *Campylobacter* concentration on chicken by a factor of 3–12 (see Table 3). Assuming these interventions would translate linearly to public health impact, the health burden associated with *Campylobacter* on poultry would be reduced by a factor of 3–12 too. In practice, there will be slaughterhouses within and across Member States and associated countries that already comply at a high level with each progressively stringent criterion and others that do not comply. As a consequence, the exact effect of current and future interventions on *Campylobacter* numbers and prevalence in the poultry supply chain may differ significantly for individual countries. Hence, the effect of these interventions on public health in countries and overall in the EU is difficult to predict. What can be done is to assess the impact of the criterion on non-compliance levels of FBOs in countries on the basis of the 2008 baseline data assuming no interventions would be introduced.

Table 3
Performance of *Campylobacter* sampling plans, defined as μ_{logC} detected with a 95% or 99% probability ($P_{non\text{-}compliant}$) when 50 samples are taken and the microbiological limit *m* is 3 log cfu/g, for different standard deviations (σ) and *c*-values (*c*).

c	σ_{logC}	$P_{non\text{-}compliant}$	μ_{logC} (log cfu/g)	difference μ_{logC} compared with $c = 20$[a]
20	1.0	0.95	3.1	
15	1.0	0.95	2.8	0.26 (factor 2 on arithmetic scale)
10	1.0	0.95	2.5	0.55 (factor 4)
20	2.0	0.95	3.1	
15	2.0	0.95	2.6	0.52 (factor 3)
10	2.0	0.95	2.0	1.09 (factor 12)
20	1.0	0.99	3.2	
15	1.0	0.99	2.9	0.26 (factor 2)
10	1.0	0.99	2.6	0.54 (factor 3)
20	2.0	0.99	3.4	
15	2.0	0.99	2.9	0.51 (factor 3)
10	2.0	0.99	2.3	1.07 (factor 12)

[a] Difference mean concentration is on logarithmic scale, μ_{logC}, but can also be expressed as factor on non-logarithmic or arithmetic scale as $10^{difference\ \mu logC}$.

3.2. Performance of the Campylobacter sampling plan in actual situations

In 2010, the EFSA published its 2008 baseline data on *Campylobacter* in various EU Member States and associated countries (EFSA, 2010). A total of 10,132 broiler batches were sampled from 561 slaughterhouses in the 25 contributing EU Member States and two countries not belonging to the EU. Caecal and carcass samples were used to assess the presence and concentration of *Campylobacter*. With these data, the performance of the *Campylobacter* in broilers hygiene criterion can be determined in terms of non-compliance (i.e., the $P_{non\text{-}compliant}$ value). To determine the $P_{non\text{-}compliant}$, one can follow two approaches. First, one can simply calculate the proportion of samples higher than the *m*-value from the original sampling data results. With this proportion (i.e., $P_{defective}$ based on original sampling data) and using equation (2), the compliance with the criterion can be determined. The second approach is to fit a probability distribution to the occurrence of different possible concentrations, and to then estimate the parameters of that distribution. For such specific distributions of organisms (e.g. a normal distribution of the log concentrations with a given mean and standard deviation) the non-compliance probability can be determined either with the ICMSF spreadsheet (ICMSF, 2020) or with equation (4). Both procedures are used below and explained in Tables 4 and 5 for the first four countries listed by EFSA (2010) but performed for all countries in this study (Table 6). For every country, the categorised *Campylobacter* concentrations numbers were copied from the baseline study (Table 4). Then, the frequency of the reported concentrations (Table 5) and the cumulative frequencies and overall prevalence levels were calculated (Table 6) based on the combined results of detection and enumeration method in the 2008 baseline study.

The normal distribution can be fitted to the cumulative frequency data of the log concentrations to determine the mean and the standard deviation of the log concentrations in the various countries. Example distribution fits for six countries and the total EU data are presented in Fig. 1, and fitting outcomes (fitted mean and standard deviation of the log concentrations) for the various countries are presented in Table 7.

With these parameters of the distribution for FBOs in a specific country, the probability that one sample is above the *m*-value ($P_{defective}$) can be calculated using equation (1), which can be compared to the probability of a sample being above the *m*-value based on the original sampling results using the data from Table 6. It can be seen that these two probabilities are rather comparable for FBOs in all countries (Table 7). The probability determined with the fitted distribution has the advantage that it has smoothed the data and is not dependent on the often rather small frequencies in the two last bins with the higher concentrations (Table 6), but on the other hand relies heavily on the assumption that the normal distribution is the appropriate model to describe the log concentrations.

Using both approaches, the non-compliance probability of the three sampling plans with progressively higher stringency (i.e., *c*-value of 20, 15 and 10) has been determined using equation (4) based on the

Table 4
Categorised *Campylobacter* concentrations (cfu/g) present in broiler carcasses, in the EU, 2008 (EFSA, 2010) for four countries.

cfu/g	<10	10–39	40–99	100–999	1000–10000	>10000	Total
Austria	146	37	45	86	63	31	408
Belgium	188	20	19	74	66	13	380
Bulgaria	163	1	15	52	28	21	280
Cyprus	352	0	1	2	2	0	357

Table 5
Frequency of *Campylobacter* concentrations (cfu/g) present in broiler carcasses for four countries.

cfu/g	<10	10–39	40–99	100–999	1000–10000	>10000
Austria	0.358	0.091	0.110	0.211	0.154	0.076
Belgium	0.495	0.053	0.050	0.195	0.174	0.034
Bulgaria	0.582	0.004	0.054	0.186	0.100	0.075
Cyprus	0.986	0.000	0.003	0.006	0.006	0.000

contamination distribution for FBOs in individual countries determined from the 2008 baseline data (Table 8).

It can be seen that, qualitatively, the results of both approaches are similar and general conclusions will be the same. It can be concluded from Table 8 (fitted distribution), and assuming the hypothetical 2% operational target for maximum non-compliance, that based on the 2008 data, six countries would have non-compliant probabilities above 2%, even with a *c*-value of 20. However, before the implementation of this legislation in 2018, there was time to improve performance, which the calculations here do not consider as these data are not available. Due to the change in the *c*-value in 2020 from 20 to 15, the poultry industry in seven additional countries (and the EU as a whole) are also above a 2% non-compliance target, if the situation would have been unchanged from 2008. In 2025, with the *c*-value lowered to 10, a further five countries would end up above the 2% non-compliance rate, when no interventions had been implemented since 2008. Notably, the poultry industry in nine countries would remain below the 2% non-compliance rate assuming their *Campylobacter* concentration data of 2008 stayed at the same reference level.

It should be noted that these calculations are based on data from 2008, and the data have been collected over the whole year and averaged over various slaughterhouses within a country. Importantly, the EU criteria apply to each single FBO over time. The specific standard deviations (and mean) of the log concentrations will be very different for specific FBOs as their individual baseline data would depend on specific factory/supply chain variability as well as flock-to-flock variability. Notably, the variation in one single plant will vary considerably since even on a single day, the poultry being processed likely comes from multiple farms. When 50 samples are taken over 10 weeks, then the variability will be even larger. Evidently, the variability in the data of the baseline study used as input into the current calculations, which are at the country level, will yet again be larger. Thus, it could well be that, for example, a specific slaughterhouse in Ireland would comply already in 2020 with the 2025 criterion and that a specific slaughterhouse in the Netherlands would fail the 2020 criterion. Moreover, the sample period, the sampled carcass area and the size of the sample all can have a significant effect on *Campylobacter* contamination levels, as demonstrated by Duqué et al. (2018) for French slaughterhouses, and this would also affect compliance levels.

The analysis in this paper gives an estimate of expected average

Table 6
Cumulative frequency of *Campylobacter* concentrations present on broiler carcasses for 25 EU Member States, the EU total, and Norway and Switzerland, and the overall prevalence of *Campylobacter*-contaminated broiler carcasses based on the combined results of detection and enumeration method.[a]

cfu/g log(cfu/g)	<10 <1.0	<40 <1.6	<100 <2.0	<1000 <3.0	≤10000 ≤4.0	$prevalence_{overall}$
Austria	0.358	0.449	0.559	0.770	0.924	0.806
Belgium	0.495	0.547	0.597	0.792	0.966	0.527
Bulgaria	0.582	0.586	0.639	0.825	0.925	0.452
Cyprus	0.986	0.986	0.989	0.994	1.000	0.141
Czech Republic	0.486	0.495	0.514	0.732	0.917	0.686
Denmark	0.763	0.788	0.816	0.912	0.985	0.314
Estonia	0.980	0.980	0.990	0.990	1.000	0.049
Finland	0.978	0.989	0.995	0.997	1.000	0.055
France	0.242	0.370	0.481	0.846	0.974	0.887
Germany	0.569	0.632	0.676	0.845	0.961	0.608
Hungary	0.502	0.617	0.673	0.875	0.953	0.553
Ireland	0.038	0.190	0.259	0.581	0.911	0.983
Italy	0.626	0.684	0.718	0.875	0.962	0.496
Latvia	0.664	0.779	0.820	0.959	1.000	0.336
Lithuania	0.540	0.738	0.786	0.947	0.995	0.458
Malta	0.054	0.057	0.071	0.204	0.681	0.943
Netherlands	0.676	0.725	0.748	0.895	0.977	0.376
Poland	0.234	0.270	0.308	0.630	0.921	0.804
Portugal	0.390	0.466	0.511	0.758	0.957	0.702
Romania	0.370	0.381	0.403	0.524	0.857	0.642
Slovakia	0.313	0.360	0.438	0.694	0.948	0.791
Slovenia	0.194	0.584	0.707	0.942	0.998	0.778
Spain	0.075	0.183	0.224	0.558	0.841	0.926
Sweden	0.910	0.932	0.954	0.990	1.000	0.146
United Kingdom	0.329	0.367	0.416	0.728	0.953	0.863
EU total	0.470	0.544	0.591	0.784	0.942	0.758
Norway	0.987	0.992	0.995	1.000	1.000	0.051
Switzerland	0.480	0.532	0.578	0.797	0.968	0.717

[a] Exceptionally, *Campylobacter* enumeration was not performed for broiler carcass samples in Luxembourg and Greece did not participate in the baseline survey; United Kingdom was an EU Member State at the time of the 2008 baseline study.

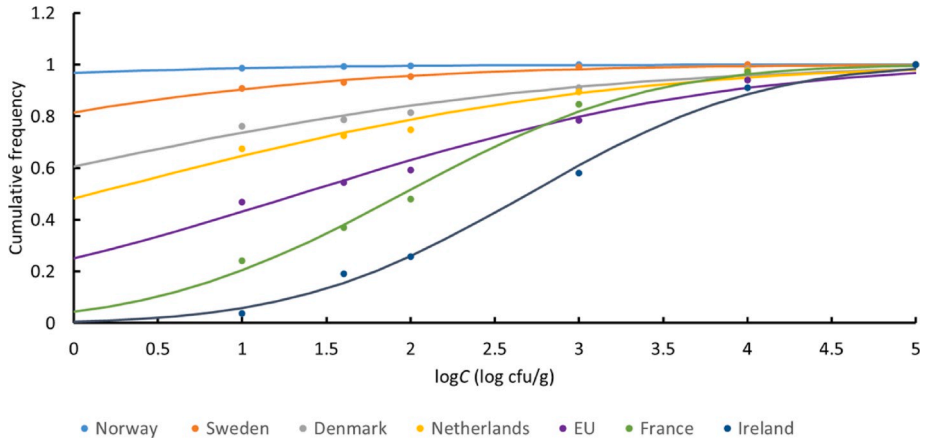

Fig. 1. Fitted cumulative distribution of *Campylobacter* concentrations (log cfu/g) in six countries and the EU total data.

Table 7
Estimated parameters, mean log concentration (μ_{logC}) and standard deviation (σ_{logC}) of the normal distribution describing the cumulative frequency distribution of the log concentrations, and the probability of a sample having a concentration larger than 3 log cfu/g ($P_{defective}$) based on this distribution and based on the original sampling data.

	μ_{logC}	σ_{logC}	$P_{defective}$(log$C$>3) Fitted normal distribution[a]	$P_{defective}$ (log$C$>3) Original sampling data
Austria	1.73	1.71	0.229	0.230
Belgium	1.26	2.04	0.197	0.208
Bulgaria	0.80	2.52	0.192	0.175
Cyprus	−9.74	5.01	0.006	0.006
Czech Republic	1.49	2.33	0.259	0.268
Denmark	−0.83	2.84	0.089	0.088
Estonia	−8.33	4.60	0.007	0.010
Finland	−3.27	2.11	0.001	0.003
France	1.95	1.15	0.181	0.154
Germany	0.78	2.21	0.157	0.155
Hungary	1.05	1.82	0.142	0.125
Ireland	2.70	1.09	0.392	0.419
Italy	0.40	2.35	0.134	0.125
Latvia	0.32	1.68	0.056	0.041
Lithuania	0.84	1.35	0.054	0.053
Malta	3.63	0.86	0.766	0.796
Netherlands	0.062	2.44	0.114	0.105
Poland	2.46	1.41	0.351	0.370
Portugal	1.72	1.76	0.233	0.242
Romania	2.30	2.40	0.385	0.476
Slovakia	2.08	1.60	0.283	0.306
Slovenia	1.55	0.73	0.023	0.058
Spain	2.82	1.22	0.442	0.442
Sweden	−2.21	2.46	0.017	0.010
United Kingdom	2.05	1.56	0.272	0.272
EU total	1.33	2.06	0.208	0.216
Norway	−4.81	2.61	0.001	0.000
Switzerland	1.35	1.96	0.199	0.203

[a] $P_{defective}$ is calculated as 1 – cumulative frequency log cfu/g < 3.0 (see Table 6).

Table 8
Non-compliance probabilities[a] calculated based on 2008 baseline data for various EU and associated countries considering different c-values and using either the fitted normal distribution or the original sampling data.

	$P_{non\text{-}compliant}$ Based on fitted distribution			$P_{non\text{-}compliant}$ Based on original sampling data		
	$c = 20$	$c = 15$	$c = 10$	$c = 20$	$c = 15$	$c = 10$
Austria	0.002	**0.089**	**0.612**	0.002	**0.094**	**0.623**
Belgium	0.000	**0.027**	**0.396**	0.001	**0.043**	**0.472**
Bulgaria	0.000	**0.022**	**0.361**	0.000	0.009	**0.250**
Cyprus	0.000	0.000	0.000	0.000	0.000	0.000
Czech Republic	0.010	**0.203**	**0.783**	0.014	**0.246**	**0.821**
Denmark	0.000	0.000	0.004	0.000	0.000	0.004
Estonia	0.000	0.000	0.000	0.000	0.000	0.000
Finland	0.000	0.000	0.000	0.000	0.000	0.000
France	0.000	0.013	**0.287**	0.000	0.003	**0.137**
Germany	0.000	0.003	**0.153**	0.000	0.003	**0.142**
Hungary	0.000	0.001	**0.089**	0.000	0.000	**0.041**
Ireland	**0.392**	**0.883**	**0.997**	**0.547**	**0.942**	**0.999**
Italy	0.000	0.001	**0.065**	0.000	0.000	**0.041**
Latvia	0.000	0.000	0.000	0.000	0.000	0.000
Lithuania	0.000	0.000	0.000	0.000	0.000	0.000
Malta	**1.000**	**1.000**	**1.000**	**1.000**	**1.000**	**1.000**
Netherlands	0.000	0.000	**0.024**	0.000	0.000	0.013
Poland	**0.190**	**0.725**	**0.985**	**0.276**	**0.809**	**0.992**
Portugal	0.003	**0.103**	**0.642**	0.004	**0.133**	**0.695**
Romania	**0.354**	**0.862**	**0.996**	**0.826**	**0.991**	**1.000**
Slovakia	**0.026**	**0.329**	**0.876**	**0.057**	**0.466**	**0.933**
Slovenia	0.000	0.000	0.000	0.000	0.000	0.000
Spain	**0.672**	**0.971**	**1.000**	**0.675**	**0.972**	**1.000**
Sweden	0.000	0.000	0.000	0.000	0.000	0.000
United Kingdom	0.017	**0.269**	**0.838**	0.017	**0.267**	**0.837**
EU total	0.001	**0.044**	**0.476**	0.001	**0.058**	**0.528**
Norway	0.000	0.000	0.000	0.000	0.000	0.000
Switzerland	0.000	**0.030**	**0.412**	0.000	**0.036**	**0.440**

[a] Using maximum of 2% non-compliance as the operational target, probabilities above 0.02 are indicated in bold.

effects for poultry slaughterhouses in various EU countries as such estimates, despite all notable variability, give insights to countries and FBOs into the magnitude of the effort needed to improve their stringency of control of *Campylobacter* to remain compliant over time with the prevailing EU regulation.

4. Discussion

Campylobacter is a leading zoonotic illness in many parts of the world and many governments and industries are striving to reduce the burden of illness caused by this pathogen through food and water. Different approaches for achieving this have been reported. In New Zealand, for instance, the focus has been on achieving a reduction in *Campylobacter* levels on carcasses at the end of primary processing by improving the food safety control systems for slaughter and dressing of broiler chickens (Lee et al., 2015). As a means to verify that the control systems are effective, the New Zealand government established a microbiological criterion referred to as a regulatory *Campylobacter* Performance Target (CPT), which follows a "moving window approach". In the CPT, a "moving window limit" failure occurs when the log count values for seven or more samples out of 45 samples in the moving window are greater than the established maximum level of 3.78 log cfu/carcass. If processors fail the "target", they are expected to take corrective actions

to restore control. In the United States of America, a regulatory *Campylobacter* performance standard has been introduced (USDA-FSIS, 2015), which included a microbiological criterion stipulating $n = 51$ samples and $c = 8$, so allowing 15.7% positives. This criterion applies to the overall process control of an FBOs establishment, not to individual products, with an aim towards pathogen reduction. Products are not tested to determine their disposition, but rather to measure the effectiveness of the slaughter and grinding process in limiting contamination of the pathogen. Establishments do not have to hold products or recall products based on results of the *Campylobacter* sampling and testing.

In the above examples of control over foodborne campylobacteriosis, authorities issue specific microbiological criteria in regulations that allow FBOs flexibility in how compliance is achieved and that allow positive samples to be found within certain boundaries of concentration or prevalence. Over time, new regulations with stricter criteria could be issued to achieve further illness reduction. The *Campylobacter* process hygiene criterion stipulated by the EC is rather unique in using an approach that drives for continued improvement over time in controlling foodborne illnesses from the moment that the criterion is put into a single regulation. This gives FBOs and Member State governments an early warning and time to prepare for their poultry industries to achieve compliance. The rationale of the EC for selecting this approach in the case of campylobacteriosis (EC, 2017) can be summarized as.

- human campylobacteriosis is the most reported human foodborne illness in the European Union
- in the 2008 baseline study, broiler carcases were found to be contaminated at an average of 75.8% with significant variations between Member States and also between slaughterhouses
- the handling, preparation and consumption of broiler meat is likely to account for 20%–30% of campylobacteriosis, while 50%–80% can be attributed to the chicken reservoir as a whole
- a public health risk reduction from the consumption of broiler meat of greater than 50% could be achieved if chicken carcases complied with a *Campylobacter* limit of 1000 cfu/g
- such a limit could be introduced as a process hygiene criterion for *Campylobacter* on broiler carcases (using EN ISO 10272–2 as the method for the enumeration of *Campylobacter*), with the aim to keep contamination of carcases during the slaughtering process under control
- a cost-benefit analysis showed that setting a process hygiene criterion for *Campylobacter* in broiler carcases would provide one of the best balances between reducing human campylobacteriosis attributed to the consumption of poultry meat and economic consequences from the application of the criterion
- based on the challenge that remains in controlling *Campylobacter* in the supply chain, a step-by-step approach should be considered, making the process hygiene criterion gradually stricter over time

So, very intentionally, the *Campylobacter* process hygiene criterion developed by the EC has several interesting characteristics that competent authorities and FBOs may want to be aware of. The first is that it tests for a rather serious pathogen, although it is considered a process hygiene criterion, and a few positive samples are allowed to be found since it is an intermediate product not considered to be eaten raw. Although risk management actions have to be taken to improve hygiene at slaughter, the product does not have to be withdrawn or recalled when the testing results are showing that a batch is not compliant. Therefore, the negative effects of intensive sampling are limited. Secondly, since a certain number of samples are allowed to be positive, trend analysis applied for verification testing can provide useful management information before batches actually are out of compliance with the criterion and definite action has to be taken to improve process hygiene. Trend analysis can be performed within the sampling window of 10 weeks, to already take action within this time frame, before surpassing the criterion. In addition, trend analysis over various subsequent 10 week periods can also be informative to visualize and to take action on slower developing trends. Thirdly, the criterion is made more stringent over time at specified dates, so FBOs and country governments have clear timelines to work on improvements. Lastly, the criterion also rewards good performance of FBOs, as such that the sampling frequency can be reduced, saving financial and human resources, in case satisfactory results have been obtained over a longer period or if an appropriate control program is in place.

As illustrated in this study, the probability of non-compliance to an expected compliance level can be determined either directly from available concentration data in poultry production in a country or it can be based on fitted distributions. Using the same approaches, the rate of non-compliance could be derived from data of a specific slaughterhouse. Where possible, it is best to apply both approaches and compare the outcomes of the analyses in terms of either a compliance or non-compliance rate. Generally, the two approaches may give comparable results and, when this indeed is the case, it provides confidence in the projected (non-)compliance rates.

As shown in Table 3, the performance of the increasingly stringent criterion reduces the mean log contamination level theoretically by about 0.25–0.5 log when the *c*-value decreases from 20 to 15 (factor 2–3 in arithmetic concentration); the *c*-value changing from 20 to 10 would give about a 0.5–1.1 log reduction (factor 4–12 in arithmetic concentration). In reality, there is no single level of performance for all slaughterhouses in a country, so there also will not be single levels of improvement in performance. As shown in Table 7, there rather are individual country differences in poultry processing that probably relate to differences at slaughterhouse level. Conceivably, a number of slaughterhouses are already below certain target levels while others need to improve their operational performance in order to be compliant at the next change in *c*-value stringency.

Overall, from our calculations and assuming no further improvement to the baseline data since 2008, the *Campylobacter* criterion that was introduced in 2018 in the EU is not expected to result in large operational non-compliance rates as the poultry industry in 17 out of 27 EU and associated countries would keep non-compliance <0.1%, while in six countries it would not meet a 2% operational target of non-compliance that was hypothetically used in this study to compare possible non-compliance levels based on fitted *Campylobacter* 2008 baseline data. However, in 2020, the poultry industry in 13 countries would be over 2% non-compliance if no improvements of hygiene conditions were made, and in 2025, 18 countries (about 60%) would be experiencing serious non-compliance. Evidently, these possible non-compliance level calculations are for illustrative purposes only as *Campylobacter* data for the EU and associated countries after 2008 have not been updated systematically and made publicly available. Nevertheless, our findings stress the fact that already in 2018 the EU criterion is a challenge for the poultry industry in several countries and thus action had to be taken by these between 2008 and 2018 to comply; other countries may have been pro-active and implemented improvements in anticipation of the planned stringency increases. A case in point to illustrate that the 2008 baseline *Campylobacter* data may not accurately reflect levels in countries in 2018, when the criterion came into force, is a recent Irish study that demonstrated that the overall prevalence of *Campylobacter* on broiler carcasses reduced from 98.3% in 2008 (see also Table 6) to 53% in 2017–2018, although prevalence levels cannot be exactly compared because the sampling schemes were different in the two surveys (Lynch et al., 2022). In this Irish study, neck skin samples were collected from the three largest broiler processing plants and it was found that 11%, 10% and 21% of the samples had *Campylobacter* counts equal or higher than 3.0 log cfu/g, in plant A, B, and C, respectively, and with an overall average of 12% (see Supplementary Table S1). This would indicate that the non-compliance rate in all three plants was lower than 2% at the moment the criterion with $c = 20$ came in force in 2018. Assuming that no further *Campylobacter* reduction could be achieved after the survey investigation in 2017–2018, then only plant C has

a non-compliance rate higher than 2% in 2020 ($c = 15$) and 2025 ($c = 10$). This demonstrates that processing plant/slaughterhouse specific survey data are useful to reflect on the necessity for continuous improvement and in combination with the approach presented here, can help stakeholders to assess compliance levels valid for their circumstances. From a government perspective, such calculations can help to estimate to what extent different jurisdictions or industry sectors may achieve the gradual improvement in the hygiene level aimed for when establishing the criterion. An update of the 2008 data would be helpful as a reference to the more current state of compliance in the EU region in relation to the EU criterion.

5. Conclusion

The EC has established a rather unique microbiological criterion to improve food safety by regulation, pre-defining a time horizon of increased stringency requirements, but also focusing on the increase of stringent process hygiene measures. While it may be a criterion that is somewhat complicated in description ($m \neq 0$, $c \neq 0$, changing value for c over time, allowing reduced frequency of sampling), the performance of such a criterion in practice has the advantage that a continuous improvement goal of government can be achieved over time, whilst giving the private sector time to prepare for expected performance increases. In addition, management information becomes available to operators before the criterion is surpassed and thus the costs of compliance can be kept low for well-performing operators. By phasing in increased stringency, every actor or stakeholder knows beforehand what future challenges they need to tackle and by when. The poultry industry in countries or specific FBOs with the highest levels of *Campylobacter* obviously have the greatest challenges to overcome.

CRediT authorship contribution statement

Marcel H. Zwietering: Conceptualization, Methodology, Formal analysis, Writing – original draft, Writing – review & editing. **Wayne Anderson:** Conceptualization, Methodology, Writing – review & editing. **Jeffrey M. Farber:** Conceptualization, Methodology, Writing – review & editing. **Leon G.M. Gorris:** Conceptualization, Methodology, Writing – review & editing. **Heidy M.W. den Besten:** Conceptualization, Methodology, Writing – review & editing.

Declaration of competing interest

The authors of the paper declare that there is no financial/personal interest or belief that could affect their objectivity.

Data availability

Data will be made available on request.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.foodcont.2023.109962.

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