

## Using consumer acceptance to guide microbial thresholds for the shelf life of fresh chicken breasts

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### ABSTRACT

A declared shelf life aligned with consumer acceptance is essential to avoid unnecessary food waste. The relationship between microbial development and consumers' odour-based decisions to accept or discard industrially packed chicken breasts from three European countries (Hungary, Norway and Portugal) was investigated. Products were stored at 4 °C up to 27 days, with or without a shift to 8 °C after 3–7 days to mimic temperature abuse at retail/consumer stages. Bacterial levels were quantified by culture-based enumeration (total viable counts, TVC), and the microbiota assessed by 16S rRNA gene amplicon sequencing. In each country  $\geq 120$  consumers assessed acceptance based on odour. Acceptance was modelled through survival analysis using time (days) and TVC as predictors.

Initial TVC differed among products (2.5 - 4.3 log cfu/g). Dominant taxa were *Photobacterium* (Hungary), *Lactobacillales* (Norway), and *Brochothrix* together with *Yersiniaceae* (Portugal) at the end of the declared shelf life. Temperature abuse shifted the microbiota towards higher proportions of *Enterobacteriales* (Norway and Portugal). Sensory shelf life varied markedly, with 25% rejection rates reached after 9 days (Hungary), 25 days (Norway), and 12 days (Portugal) during storage at 4 °C, corresponding to TVCs of 6.4, 7.4 and 7.3 log cfu/g.

We propose a consumer-anchored methodological approach for shelf life assessment that (i) predefines an acceptable rejection level (ii) models acceptance as a function of TVC under foreseeable abuse and (iii) complements culture-dependent analyses with culture-independent profiling. This approach links microbial criteria to consumer perception and behaviour and providing a basis for more accurate, consumer-anchored shelf-life determination.

### 1. Introduction

Poultry meat is a valuable food product, with a high protein content and a relatively low carbon footprint, land use and production costs compared with other meats (Poore and Nemecek, 2018; Statista, 2025). The production of poultry meat globally is about 127 million tons (Food and Agriculture Organisation of the United States, 2025) and is growing faster than for beef and pork (U.S. Department of agriculture, 2025). While being a valuable food source, chicken meat is also a vehicle for foodborne pathogens, such as *Salmonella* spp. and *Campylobacter* spp.

(Khalid et al., 2020). *Campylobacter* spp. may cause illness even at low doses, and foodborne illness is primarily caused by consumption of undercooked meat or cross-contamination from raw meat, resulting in ingestion of pathogens (Colin et al., 2025; Khalid et al., 2020; Sharma et al., 2025). Since *Salmonella* spp. and *Campylobacter* spp. do not grow under refrigerated storage, the risk of foodborne illness is in most cases constant or declining during the storage time and illness is therefore not linked to consumption of products after passing the expiry date (Bhaduri and Cottrell, 2004; Silva et al., 2022). In contrast, sensory quality deterioration proceeds as psychrotrophic spoilage bacteria grow,

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producing off-odours, off-flavours, discolouration and slime (Rouger et al., 2017). Shelf life for chilled fresh poultry is therefore primarily restricted by spoilage rather than by pathogen growth.

Several factors affect the consumers' decision to discard food, and the relative importance of microbial issues or concerns is not known (Quintieri et al., 2023). However, a significant contributor to food waste is food that has been stored for so long at the consumer stage that it is either spoiled or has passed the date label (Llagas et al., 2025). Declaring shelf life that is too short may increase premature discarding, whereas declaring it too long may result in unacceptable quality at end of life, where both scenarios can elevate food waste. In a food waste context, a consumer-anchored shelf life is therefore essential, though whether more accurate shelf-life determination translates directly into reduced food waste at the consumer stage remains an open question.

The declared shelf life is the responsibility of each food business operator. For products restricted by spoilage, guidance documents recommend durability studies with microbial and sensory assessments, incorporating reasonably foreseeable storage conditions including temperature abuse at retail and consumer stages (Koutsoumanis et al., 2020; Everis, 2019; Food Standards Scotland, 2025; MPI, 2016).

The guidance documents do not define microbial thresholds for spoilage organisms, but a critical limit of 10<sup>7</sup> cfu/g bacteria is widely applied in practice as a default criterion for unacceptable spoilage (ASTM International, 2020; Koutsoumanis et al., 2020). While this threshold provides a practical default in the absence of product-specific data, the literature shows considerable variation in rejection levels spanning from 6.5 to 9.5 log cfu/g across different studies (Alakomi et al., 2017; Augustynska-Prejsnar et al., 2024; Balamatsia et al., 2006; Chouliara et al., 2008; Dogu-Baykut and Gunes, 2014; Dold et al., 2022; Economou et al., 2009; Franke et al., 2017; Herbert et al., 2015; Holck et al., 2014; Ntzimani et al., 2023; Rossaint et al., 2015; Rotabakk et al., 2006; Smolander et al., 2004; Tsaloumi et al., 2023; Wang et al., 2020). One likely reason for the variations between studies is differences in the spoilage potential of the microbiota, which depends both on the species or strain, but also on storage conditions affecting their metabolism (Hansen et al., 2023).

Many previous studies used selective media to identify the spoilage microbiota, resulting in an underestimation of the dominating microbiota (see e.g. (Balamatsia et al., 2006; Herbert et al., 2015; Rossaint et al., 2015; Rotabakk et al., 2006)). To avoid such biased results, it has been proposed to use culture-independent techniques (e.g., 16S amplicon sequencing) to characterize the microbial development in durability studies (Everis, 2019).

No specific recommendations on how to set sensory threshold values are defined in the guidance documents. Terms like “retain specific qualities” and “fit for consumption” are applied, indicating respectively sensory analysis and consumer evaluation. The sensory shelf life of foods is typically set as the timepoint where 25-50% of consumers reject the sample. This threshold can be estimated from sensory data from consumer panels evaluating the product combined with a statistical modelling approach referred to as “survival analysis” (Hough, 2010). Survival analysis focuses on the probability of consumer rejection after a certain storage time rather than measuring changes in product quality (Hough and Garitta, 2012).

To link the microbial quality to consumer acceptance, an approach parallel to quantitative microbial risk assessment (QMRA) has been proposed for spoilage (Koutsoumanis et al., 2021). Whereas a food safety QMRA models the dose-response for illness, a food spoilage QMRA (Quantitative microbiological spoilage risk assessment; QMSRA) models dose-response for consumer rejection as a function of specific spoilage organisms (Tsaloumi et al., 2023). Consistently, the EFSA Biohazard panel recommends defining the “maximum acceptable levels” of spoilage organisms that corresponds to the end of the sensory shelf life (Koutsoumanis et al., 2020). However, such maximum values are largely missing in the literature. Most shelf life studies use too small panels of assessors (10 or less, in-house) to evaluate product acceptance. To get

reliable results, recruiting at least 120 consumers belonging to the target group is recommended (Hough and Garitta, 2012; Hough et al., 2007). The largest consumer study on shelf life of chicken meat identified in the literature included 45 panellists recruited from a university population. (Soederqvist et al., 2024). However, no sensory changes were observed within the timeframe of the study, and it was not possible to draw conclusions about the relationship between consumer acceptance and microbial development.

The aim of this study was to quantify the link between microbial development and consumer acceptance (decision to consume or discard) for industrially packed chicken breasts. To cover different microbiota in chicken meat, products from large producers in three countries (Hungary (vacuum packaged), Portugal (modified atmosphere with O<sub>2</sub> and CO<sub>2</sub>), Norway (modified atmosphere with CO<sub>2</sub>) were stored at cold and abuse temperatures. During storage, samples were taken at different time points varying from 1 to 27 days. The microbial load and the composition of the microbiota was determined, and the likelihood of rejection was measured with consumer panels in each country. Survival analysis with either storage time or TVC as predictor variables was used to explore the association between the microbial development to consumer acceptance. Because the design combines several product-specific factors, it was not intended to support causal inference regarding country, packaging method, or consumer group. We further propose a consumer-anchored assessment methodology to translate acceptance targets into microbial decision criteria for product-specific shelf-life assessment.

2. Materials and methods

2.1. Raw materials and storage

Fresh chicken breasts (without skin) in consumer packages were shipped at 4 °C from chicken processing factories located in Norway, Portugal, and Hungary to laboratories in the same country. An overview of products is given in Table 1. The chicken products were received the same day (Portugal) or the day after (Hungary, Norway), slaughter and processing and stored in their original package. Two different scenarios were investigated: i) 4 °C throughout the whole storage period and ii) 4 °C for 30-40% of the declared shelf life and shifted to 8 °C for the remaining storage time, to simulate foreseeable abuse at retail/consumer stages (see Table 1). Samples were taken throughout the storage period for assessment of the gas composition, microbial numbers, microbiota and for the consumer test. At each sampling point, chicken breasts to be used in the consumer test were vacuum packed and frozen until evaluation, while other samples were analysed immediately either before (gas composition) or after opening the packages (microbiology). Frozen storage may affect sensory and quality traits of chicken meat, including flavour, aroma, texture, drip loss, and lipid and protein oxidation, particularly during extended storage at around -20 °C (Augustynska-Prejsnar et al., 2018; Soyer et al., 2010). As samples in the

Table 1  
Overview of chicken products and storage conditions.

| Country  | Packaging gas <sup>a</sup>                      | Size of packages         | Shelf life <sup>b</sup> | Moved to 8 °C | Sampling days        |                |
|----------|-------------------------------------------------|--------------------------|-------------------------|---------------|----------------------|----------------|
|          |                                                 |                          |                         |               | 4 °C                 | 8 °C           |
| Hungary  | Vacuum                                          | 1 kg/4 breasts           | 10 days                 | 4 days        | 1, 4, 8, 11, 15, 18  | 7, 9, 11       |
| Norway   | 45-50% CO <sub>2</sub>                          | 1 kg/3-5 breasts         | 20 days                 | 7 days        | 1, 7, 18, 20, 25, 27 | 15, 18, 20, 23 |
| Portugal | 55-70% O <sub>2</sub><br>20-30% CO <sub>2</sub> | 0.5-1 kg/<br>3-4 breasts | 7 days                  | 3 days        | 0, 5, 7, 11, 14      | 4, 7, 11       |

<sup>a</sup> Remaining gas N<sub>2</sub>.

<sup>b</sup> Shelf life according to the expiry date set by the food producer.

present study were stored frozen for up to 8 months, an effect on the absolute intensity of sensory attributes cannot be excluded. However, storage temperatures were partly lower than conventional frozen storage, with Norwegian samples stored at  $-40^{\circ}\text{C}$ , which may improve preservation of meat quality by reducing structural damage, drip loss, and oxidation (Chang et al., 2022; Dang et al., 2021; Sujiwo et al., 2025). Since all samples were handled consistently, freezing was considered less likely to systematically affect the relative differences between samples.

## 2.2. Gas composition

The headspace composition ( $\text{O}_2$  and  $\text{CO}_2$ ) of the unopened packages was measured using Check Mate instruments (PBI Dansensor, Denmark).

## 2.3. Microbial numbers

Three packages of chicken breasts were analysed for each sampling point. From each package, two breast fillets were placed in a stomacher bag. The fillets were added 150 mL of diluent (0.1% peptone, 0.85% NaCl) and hand-massaged for 1 min. Serial dilutions of homogenates were made in peptone water and aliquots plated on Plate count agar (PCA). For samples from Hungary, samples were also spread on Long & Hammer (LH) Agar to allow growth of *Photobacterium* spp. LH agar was used because a pre-test showed dominance of *Photobacterium* spp. that did not grow on PCA in the products from Hungary. The plates were incubated aerobically at  $15^{\circ}\text{C}$  for 5–7 days before counting of colonies. The total viable counts (TVC) were calculated from the weight of the fillets and the number of colony forming units for each sample. Estimated counts of dominant spoilage taxa were obtained by multiplying TVC by their relative abundances from microbiota profiling. All microbial numbers were log transformed before calculating mean values.

## 2.4. Identification of colonies

Bacterial isolates were identified by MALDI-TOF MS following a common analytical framework applied by both the Norwegian and Portuguese teams, with minor differences related to colony handling procedures and analytical platform (instrumentation, software versions, and reference libraries).

Briefly, approximately ten colonies were randomly selected from a randomly chosen sector of each agar plate; when fewer than ten colonies were present, all colonies were analysed. Colony material was transferred to a MALDI target plate either by smearing a small amount of biomass as a thin film using a sterile wooden application stick or by transferring approximately 0.1 mg of cell material directly onto the target plate. Subsequently, 1  $\mu\text{L}$  of HCCA matrix solution (10 mg/mL  $\alpha$ -cyano-4-hydroxycinnamic acid in 50% acetonitrile and 2.5% trifluoroacetic acid) was added to each spot, using a new pipette tip for each position to prevent cross contamination.

In Portugal, spectra were acquired using a MALDI-TOF Biotyper Sirius mass spectrometer (Bruker Daltonics, Germany) and analysed using MBT Compass HT software (version 5.1.450.98) with the MBT Compass HT reference library (version 2023) and FlexControl 3.4 software (Bruker Daltonics). In Norway, spectra were acquired using a Biotyper MALDI-TOF mass spectrometer (Bruker Daltonics, Germany) and processed using MBT Compass 4.1, FlexControl 3.4, and BioTyper 3.1 software, in combination with the MBT 9607 MSP Library (released November 2020) and an in-house reference library. In both cases, spectra were acquired according to the manufacturer's standard measurement procedures.

Instrument calibration was carried out using the Bacterial Test Standard (Bruker Daltonics, Germany). Identification scores  $\geq 2.0$  were considered reliable for species level identification, while scores between 1.7 and 2.0 were accepted for genus level identification. Scores below 1.7, in the absence of Sanger sequencing confirmation, were considered

unreliable and recorded as "NoID".

## 2.5. Microbiota analysis

Homogenates (2–10 mL ( $>6 \log \text{cfu/mL}$ )) were transferred to sterile tubes and pelleted by centrifugation ( $13\,000 \times g$  for 5 min). For the Portuguese samples, 10 mL homogenate was used for all samples. The pellets were washed once in 0.5 mL Phosphate-Buffered Saline and frozen at  $-20^{\circ}\text{C}$  until DNA extraction and analysis. DNA extraction was done using DNeasy PowerLyser PowerSoil kit (Qiagen) as described by the manufacturer except that cells were lysed with mechanical lysis (bead beating) using Precellys Evolution,  $3 \times 7400 \text{ rpm}$  for 40 s (the plates were put on ice before and after bead beating). Extracted DNA was stored at  $-20^{\circ}\text{C}$  until sequencing. 16S rRNA gene PCR (V4 region) and paired-end sequencing (2x150 bp) using MiSeq reagent v3 kits on a MiSeq instrument (Illumina) were performed following the protocol by (Caporaso et al., 2012, 2023) with modifications as previously described (Moretto et al., 2021). The sequences were processed in QIIME2 (qiime2-2023.5) (Bolyen et al., 2019) as previously described (Moen et al., 2026a). The sequencing data have been deposited in the National Center for Biotechnology Information (NCBI) archives under BioProject no. PRJNA1310835 and PRJNA1315198.

Three MiSeq runs were performed, and counts exported from QIIME2 were merged and further processed in Python v3.13.5. In total, 81 samples were analysed and, altogether, 1414 sOTUs were detected from a total of 2 564 560 sequences after removing singletons. The mean number of sequences obtained per sample was 31 661 (min-max: 2061–73 405). The mean number of sequences per sOTU was 1814 (min-max: 2–1 208 517).

## 2.6. Consumer recruitment

Participants in the consumer experiment were required to be between 20 and 79 years old and to be consumers (incl. purchase and preparation) of chicken fillets. In Norway, recruitment was conducted via Nofima's consumer database of leisure-time associations (e.g., sports and music clubs) in the Akershus county, in the vicinity of our lab. These associations received an incentive of  $\approx 40$  euros per member participating in the test. In Hungary, convenience sampling was used by advertising to family, friends, students, and to the public. Each participant received a 5 euros voucher, and an entry to a lottery in which two 130 euros vouchers were drawn. Recruitment in Portugal was handled by Sense Test - Sociedade de Estudos de Análise Sensorial a Produtos Alimentares ([www.sensetest.pt](http://www.sensetest.pt)), an ISO/IEC 17025 accredited laboratory. Participants were volunteers recruited from their consumer database and received a reward for their involvement.

## 2.7. Consumer evaluation

A total of 365 consumers participated in the consumer experiment with valid data for analysis (Hungary: 117, Norway: 120, Portugal: 128). Consumers conducted the test in groups of up to 40 participants over several days, in sessions of maximum 1 h. Upon arrival, they received a short introduction about the aim of the experiment and were shown a package of chicken fillet. The moderator then presented the task:

"Imagine you are having chicken fillets for a meal. You are now about to open the pack of raw chicken fillets which looks like this [shows the pack] and prepare it. This is the context which you are to have in mind when assessing the chicken samples.

*You will have in front of you different samples of raw chicken that may vary in smell. The task is to smell each of them in the order indicated by the software, and to answer the on-screen questions."*

Each consumer received 11 samples of chicken fillet to be evaluated. Chicken breasts were thawed overnight in a refrigerator, divided into 6-

8 samples, and acclimated at room temperature for 3 h prior to the consumer test. The samples were presented in odour-free plastic containers covered with a lid to preserve the meat odour and identified with random three-digit codes. Evaluation was conducted individually in sensory booths. Participants were instructed to open the lid of the container, use a fork to lift the piece of chicken, and evaluate the sample by smelling the underside of the chicken piece. For each sample, the consumer reported if they found it fit for consumption according to the on-screen instructions:

*Find the sample marked with [sample code]. Imagine you are at home, preparing chicken fillet for a meal. Based on the smell, would you prepare and eat this chicken sample, or would you throw it? (I would prepare and eat it / I would throw it)*

The EyeQuestion software (Logic8 B.V., The Netherlands) was used for data collection in Norway and Hungary, and Qualtrics (Qualtrics LLC, USA) was used in Portugal. The questionnaire was developed in English, then translated from English to the three national languages by native speakers in the research team. The experiment was part of a larger study; not all questions are included in the present publication.

## 2.8. Statistical analysis

### 2.8.1. Microbial levels

All microbial counts were log-transformed before statistical analysis. Mean values and the standard error of the mean were calculated using Minitab (Minitab 22.2.2, 2026). Differences between means were tested using a two-sample *t*-test in Minitab. Estimated counts of dominant spoilage taxa were obtained by multiplying TVC by their relative abundances derived from microbiota profiling.

### 2.8.2. Survival analysis

Typically, storage time is used as the predictor in survival analysis models, but we also included TVC as the predictor. This approach incorporates variability in bacterial load arising from differences in storage conditions into the statistical model. Since the time for rejection can only be observed for intervals and not as an exact time point, censoring of consumer rejections was performed as described in (Hough, 2010). Consumers that rejected the fresh samples were not included in the analyses.

The survival function  $S(x)$  is estimated as a function of the predictor variable  $x$  and is the probability of survival after timepoint  $x$ . In classical applications, this is time to death, more generally it is the time to an event. Here we are interested in time (storage days) or TVC (log cfu/gram) until the event rejection by consumers. In survival analysis, a statistical distribution is assumed for the error distribution of the random variable  $\log(x)$ . For the sensory shelf life of foods, the Weibull distribution is usually applied, and the survival function can then be expressed as (Hough, 2010):

$$S(x) = e^{\left(e^{\frac{\log(x) - \mu}{\sigma}}\right)}$$

where  $\mu$  and  $\sigma$  are model parameters.

Models were fitted using either a) storage time (days) or b) the bacterial counts (log cfu/gram) as predictor variables. The survival model was fitted to consumer acceptance data (coded as 0 = reject, 1 = accept) using the survreg R-function (Therneau, 2024; Therneau and Grambsch, 2000). Shelf life was estimated for 10, 25, and 50% rejections.

Alternative error distributions were also evaluated, but differences in model fit were negligible.

## 2.9. Ethics

The processing of personal data was assessed and approved by the

Norwegian Agency for Shared Services in Education and Research (sikt. no, reference number 271025) in accordance with GDPR regulations. An informed consent was obtained from all participants. They were informed about the purpose of the study, which data would be collected and processed, who would have access to data and their right to withdraw from the study at any time.

## 3. Results and discussion

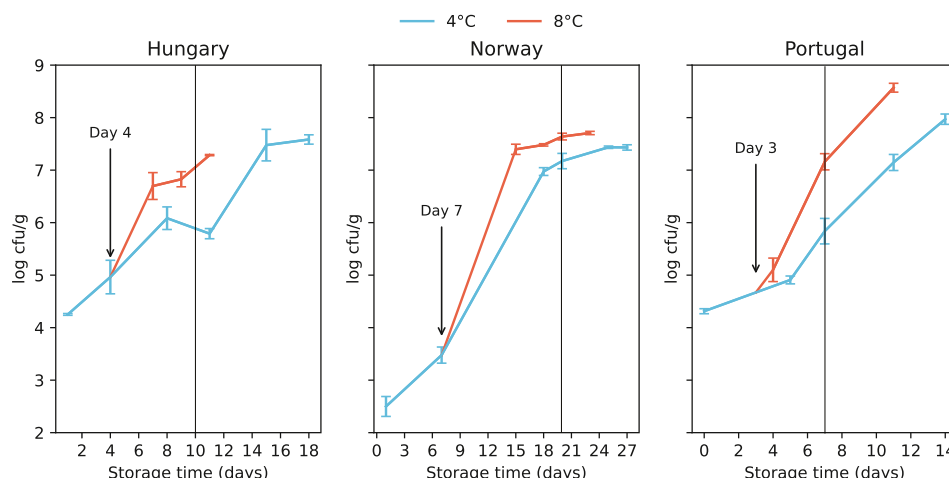
### 3.1. Initial contamination of products

Initial bacterial loads varied considerably among products from the three countries (Fig. 1). Initial contamination was substantially higher on products from the Hungarian producer than from the Norwegian producer (4.3 vs 2.5 log cfu/g;  $p = 0.012$ ). Portuguese products showed 4.3 log cfu/g at production, aligning with the contamination levels observed in chicken breasts from the Hungarian producer. Large variations in initial bacterial levels on chicken meat have also been reported in previous studies, ranging from low levels (<3 log cfu/g) in investigations from Norway (Hansen et al., 2023; Pettersen et al., 2021), Poland (Augustynska-Prejsnar et al., 2024; Chmiel et al., 2020) and Germany (Höll et al., 2016), intermediate levels (3–4.5 log cfu/g) in studies from Italy (Alessandroni et al., 2022), and Germany (Franke et al., 2017) and high levels (5.8 log cfu/g) in products from Slovakia (Necidová et al., 2024).

Culture-independent analysis showed a diverse microbiota in all products at the beginning of the storage period. Across all samples, a total of 280 taxa were detected after collapsing the data to genus level where possible and retaining taxa at the lowest assigned taxonomic rank. Of these, 93 taxa were shared across all three factories. Portugal showed the highest diversity, with 224 taxa detected, followed by Norway (169) and Hungary (148). Only a few taxa exceeded 1% relative abundance in each country, including *Lactobacillus*, *Acinetobacter*, *Pseudomonas*, *Psychrobacter*, *Lachnospiraceae*, *Chryseobacterium* and *Flavobacterium*. In products from Norway, *Lactobacillus* (17%), *Pseudomonas* (14%) and *Acinetobacter* (7%) were found in highest proportions. The microbiota resembled what was found in a study from the US, although the US samples also contained high proportions of *Salmonella* spp. (Rothrock et al., 2019). The microbiota in the products from Portugal was similar, with *Lactobacillus* (15%), *Acinetobacter* (13%), and *Pseudomonas* (5%). However, *Yersiniaceae* (8%) and *Megamonas* (8%) were also present at significant levels. To our knowledge, *Megamonas* spp. have not been reported on chicken meat earlier, although it is a common bacterium in chicken faeces (Rychlik et al., 2023). Among the genera within the family *Yersiniaceae*, earlier studies have reported presence of *Serratia* (Höll et al., 2016; Indriani et al., 2025) in similar products from Germany and Thailand. The initial microbiota on Hungarian products was dominated by *Psychrobacter* (24%) followed by *Acinetobacter* (14%) and *Pseudomonas* (10%). This pattern was similar to that reported in a recent South-Korean study (Jeong et al., 2023).

Differences among producers and across studies may reflect variation in the microbiota of the live birds, hygiene practices, processing factors, and packaging conditions. All dominant bacterial taxa identified in the present study have previously been reported as part of the live chicken microbiome in one or more studies (Jeong et al., 2023; Marmion et al., 2021; Rothrock et al., 2019; Rychlik et al., 2023). The importance of contamination from the live bird seems to vary. For example, while (Jeong et al., 2023) identified *Psychrobacter* both in the faeces and corresponding products (Rothrock et al., 2019), did not find *Psychrobacter* in either. Differences in processing and hygienic level could potentially also impact the microbiota on the chicken breasts. For example, cross-contamination in the air-chiller can increase dominance of *Pseudomonas* spp. (Chen et al., 2020; Moen et al., 2026a). Although other studies have reported an initial microbiota different from the present study, a consistent finding from culture-independent analyses is the high diversity at the start of storage (Dourou et al., 2021; Höll et al.,





**Fig. 1.** Microbial growth in chicken breast products. Total viable counts (log cfu/g) is plotted against the days of storage for one scenario where temperature is kept at 4 °C throughout the storage time (blue line) and another simulating temperature abuse (8 °C) at retail and consumer part of the chain (orange line). The timepoint where products were moved from 4 °C to 8 °C is indicated by an arrow. The vertical line is indicating the shelf life declared by the producer. Mean values and standard error of the mean is shown. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

2016; Jeong et al., 2023; Wang et al., 2020).

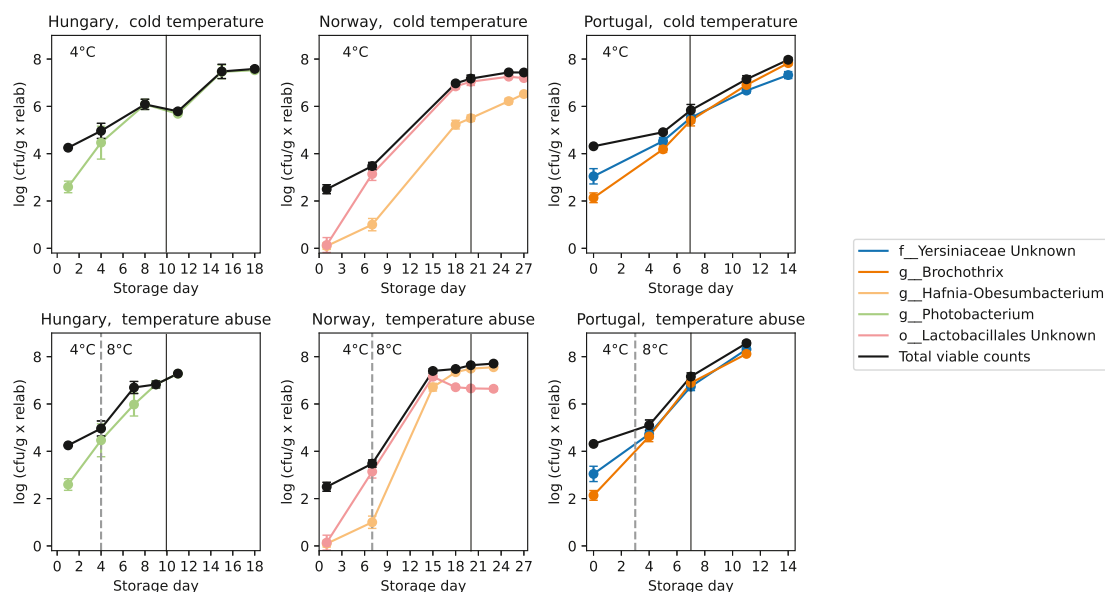
### 3.2. Microbial growth

TVC increased during storage, with similar growth rate in the products from all three factories, and faster growth after products were moved to abuse temperature (Fig. 1). At the end of the declared shelf life, the microbial levels for products from Hungary, Norway, and Portugal were approximately 6 log cfu/g (approximation as sample was not taken at the expiry date), 7.2 log cfu/g, and 5.8 log cfu/g, respectively, for products stored at 4 °C.

At or near the end of the declared shelf life, the microbiota composition differed markedly between products of different origins (Fig. 2, Supplementary 1). While *Photobacterium* spp. dominated in chicken breasts from Hungary (vacuum packed) stored at 4 °C, *Lactobacillales* dominated in products from Norway (CO<sub>2</sub>/N<sub>2</sub>). In contrast, products

from Portugal (CO<sub>2</sub>/O<sub>2</sub>/N<sub>2</sub>), were dominated by *Brochothrix* spp. together with the family *Yersiniaceae*.

Except for one single package with a diverse microbiota after 11 days of storage, *Photobacterium* spp. dominated in the products from Hungary after storage (8 and 11 days, 4 °C). *Photobacterium* spp. in vacuum packed chicken has, to our knowledge, only been reported in one earlier study (Sequino et al., 2025). In other studies, a high microbial diversity has been reported in vacuum packed chicken, with different microbial compositions between studies (Hansen et al., 2023; Indriani et al., 2025; Li et al., 2020). *Photobacterium* spp. are rarely mentioned as a spoilage bacterium in chicken, although it has been reported to dominate in products packed under air, vacuum, CO<sub>2</sub>/O<sub>2</sub> and CO<sub>2</sub>/70% N<sub>2</sub> in products from Germany, Italy and Greece (Dourou et al., 2021; Hilgarth et al., 2018; Holl et al., 2019; Sequino et al., 2025). It is likely that its presence as a spoilage organism in chicken is underreported since the microbial methods used in most earlier studies do not support growth of



**Fig. 2.** Growth of the most dominant spoilage bacteria in chicken breast products purchased from three different food factories. The total viable counts and estimates of viable counts of each dominant spoilage bacterium (log cfu/g) is plotted against the days of storage for one scenario where temperature is kept at 4 °C throughout the storage time (cold temperature) and another simulating temperature abuse (8 °C). The vertical line is indicating the shelf life declared by the producer. Mean values and standard error of the mean is shown.

all strains of *Ph. phosphoreum* and *Ph. carnosum*, which require low temperatures and supplement of salt (Dourou et al., 2021; Hilgarth et al., 2018). The exact spoilage characteristics of *Photobacterium* spp. in poultry have not been fully elucidated; however metatranscriptomics suggests that its spoilage characteristics resembles that of *Brochothrix* spp. and *Carnobacterium* spp. (Holl et al., 2019). This corresponds to a sensory profile characterized by fermented, cloying, pungent, and sweet notes in the spoiled products (Hansen et al., 2023).

In products from Norway, the microbiota identified by culture-independent analysis was dominated by lactic acid bacteria (*Lactobacillales* (74%), *Lactobacillus* (10%), *Leuconostoc* (3%)) after 20 days of storage at 4 °C. MALDI-TOF analysis of colonies from samples stored for 25 days (no colonies selected after 20 days) indicated dominance (9/10 colonies) of *Carnobacterium* spp. (*C. divergens* and *C. maltaromaticum*). *Carnobacterium* spp. and other lactic acid bacteria have been reported to dominate in products packed with CO<sub>2</sub> in several past studies (Alakomi et al., 2017; Bjorkroth et al., 2005; Holck et al., 2014; Pettersen et al., 2021; Tsafrakidou et al., 2021) and it may be accompanied by *Brochothrix* spp. (Chouliara et al., 2008). Several malodours (fermented/sour, cloying, yeast, pungent, sulphur and sweet) are produced by *Carnobacterium* spp. growing on chicken (Hansen et al., 2023). After the end of the declared shelf life (20 days to 27 days) a decline in the proportion of *Lactobacillales* (from 74% to 58%) and increase in *Hafnia* spp. (from 2% to 12%) was found, indicating a slower growth rate of the former after reaching approximately 7 log cfu/g.

After 7 days of storage at 4 °C, microbiota on chicken fillets from Portugal contained respectively 49% and 38% *Yersiniaceae* and *Brochothrix* spp. MALDI-TOF analysis of colonies from samples stored for 7 and 11 days identified *Brochothrix thermosphacta* (3/8) and *Serratia proteamaculans* (5/8) (only two colonies identified at day 7, one of each). *Brochothrix* is a well-known spoilage organism on chicken, especially in MAP with O<sub>2</sub> and CO<sub>2</sub>, and produces butter-like, cloying, yeast, fermented and sulphur odours (Franke et al., 2017; Hansen et al., 2023; Herbert et al., 2015; Pettersen et al., 2021). To our knowledge, the dominance of *Serratia* or other genera within *Yersiniaceae* in products packed with O<sub>2</sub> and CO<sub>2</sub> has not been previously reported and the role of *Yersiniaceae* in spoilage of chicken is not clear. Only moderate sensory changes were found at levels >9 log cfu/g for *Serratia liquefaciens* in a challenge test with chicken meat stored at aerobic conditions (Wang et al., 2017). In another challenge study with a cocktail of *Serratia* spp. (*S. liquefaciens*, *S. quinivorans*, *S. fonticola*) on chicken fillet stored in vacuum, no sensory changes were observed, even at a total number of >8 log cfu/cm<sup>2</sup> (Hansen et al., 2023). After passing the expiry date, a gradual shift towards dominance of *Brochothrix* spp. up to 72% after 14 days was observed, with corresponding reduction of *Yersiniaceae* and *Acinetobacter*.

Temperature abuse led to a shift in the microbiota from dominance of lactic acid bacteria (*Lactobacillales*, *Lactobacillus* and *Leuconostoc*, 87%) to *Hafnia* spp. (73%) on chicken from the Norwegian processor after 20 days of storage. A similar pattern was reported by Höll (Höll et al., 2016), with *Hafnia alvei* dominating on chicken fillets stored at 10 °C in CO<sub>2</sub>-rich atmosphere but only constituting a minor part at 4 °C. At high concentrations, *Hafnia* spp. will produce several off-odours, such as sulphur, cloying and fermented odours (Hansen et al., 2023). For the chicken from the Portuguese producer, a similar microbiota at both temperature regimes was observed when total numbers reached approximately 7 log cfu/g, but after further storage, *Brochothrix* spp. dominated at 4 °C storage temperature, and *Yersiniaceae* at 8 °C. Thus, as for the Norwegian products, bacteria belonging to the *Enterobacterales* tended to dominate more at abuse storage temperatures. For the chicken samples processed in Hungary, *Photobacterium* spp. dominated at both storage temperatures.

### 3.3. Consumer test

Around 120 consumers participated in each country (Table 2). The

**Table 2**  
Consumer characteristics.

|                    | Hungary | Norway | Portugal |
|--------------------|---------|--------|----------|
| <b>Number</b>      | 117     | 120    | 128      |
| <b>Age (years)</b> |         |        |          |
| Mean               | 29.6    | 37.0   | 49.6     |
| StDev              | 12.0    | 15.2   | 11.6     |
| Min                | 20      | 20     | 22       |
| Max                | 74      | 72     | 72       |
| <b>Sex (%)</b>     |         |        |          |
| Women              | 67.6%   | 60.0%  | 68.9%    |
| Men                | 32.3%   | 40.0%  | 31.3%    |
| Other/no response  | 0%      | 0%     | 0%       |

youngest consumers were 20 years, while the oldest 74 years. The average age was lowest in Hungary (30 years) and highest in Portugal (50 years) and in all countries, there was an overrepresentation of female participants (60–69%). Differences in demographic composition between countries can partly be explained by variations in recruitment procedures. While in Portugal and Norway participants were recruited through established consumer panels, participation relied on voluntary recruitment in Hungary. This could have led to differences in sample characteristics due to self-selection effects commonly observed in non-probability sampling (Baker et al., 2013; Bethlehem, 2010).

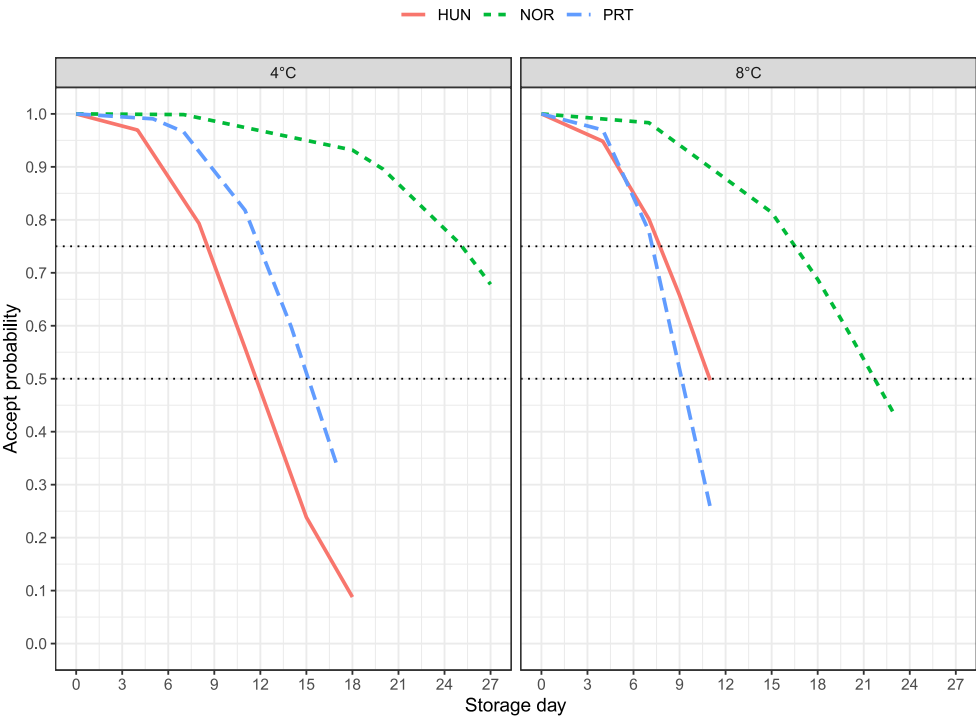
For all three countries, the fraction of consumers that reported that they would discard the chicken based on the odour increased with storage time (Fig. 3). The acceptance ratio decreased continuously with no clear cut off point. Moreover, acceptance varied considerably between individuals, with some still accepting a product stored 10 days longer than the timepoint where others rejected it.

The declared shelf life varied between the products from different countries, and this was also reflected in the number of storage days before consumers rejected the products. For example, 25% of consumers rejected the chicken product after 9 days of storage in Hungary, 25 days in Norway, and 12 days in Portugal (Table 3, Fig. 3). No other studies are directly comparable with the present study, due to much lower numbers of panellists and different breakpoints for defining the threshold for spoilage. However, a large variation of shelf life has been reported earlier, ranking from 10 to 26 days for chicken fillet stored at 4 °C (see e. g. Balamatsia et al., 2006; Chouliara et al., 2008; Holck et al., 2014; Rossaint et al., 2015). The experiment was not designed to identify explanatory factors behind the differences found, which could be caused by different microbial and sensory development in the products, differences in acceptance between the three consumer panels, or a combination of both. Experimental differences cannot be excluded either, even though coordination of the experimenters across countries and detailed protocols were used. Nevertheless, it is plausible that longer sensory shelf life in the products from the Norwegian producer was mainly related to longer microbial shelf life (Figs. 1 and 3).

For products from Norway and Portugal, storing at abuse temperature led to lower acceptance of products at the same number of storage days, corresponding to more rapid growth of spoilage microorganism (Figs. 2 and 3). For the products from Hungary, the decline in acceptance followed a similar curve for storage at cool and abuse temperatures. The reason is most likely that the growth of the most important spoilage bacterium, *Photobacterium* spp., was similar for the two temperature scenarios (Fig. 2).

### 3.4. Acceptance as a function of bacterial levels

The relationship between the bacterial levels and the acceptance was modelled across the two temperature scenarios (Fig. 4). Comparison of the fitted survival curves and their confidence intervals, together with the estimated TVC thresholds at 10–50% rejection, indicated no appreciable difference between the 4 °C and 4–8 °C scenarios across the range most relevant to threshold-setting (Supplementary material,



**Fig. 3.** Fraction of consumers that accept (respond they would prepare the sample of chicken fillet for dinner based on odour) in a) Hungary b) Norway c) Portugal. The fraction of accepting the product is plotted against the time of storage of the sample. 25% and 50% rejection rate shown as horizontal lines.

**Table 3**  
Shelf life (in days) of chicken products with different criteria for setting the expiry date and according to declared shelf life on the label.

|                      | Criteria                           | Hungary | Norway | Portugal |
|----------------------|------------------------------------|---------|--------|----------|
| 4°                   | Declared shelf life                | 10      | 20     | 7        |
|                      | 10% reject <sup>b</sup>            | 6       | 20     | 9        |
|                      | 25% reject <sup>b</sup>            | 9       | 25     | 12       |
|                      | 50% reject <sup>b</sup>            | 12      | 31     | 15       |
|                      | 10 <sup>7</sup> cfu/g <sup>c</sup> | 14      | 18     | 10       |
| 4 °C-8C <sup>a</sup> | 10% reject                         | 5       | 12     | 6        |
|                      | 25% reject                         | 8       | 17     | 7        |
|                      | 50% reject                         | 11      | 22     | 9        |
|                      | 10 <sup>7</sup> cfu/g              | 10      | 14     | 7        |

<sup>a</sup> Simulating 4 °C at transport and 8 °C at retail and consumer.  
<sup>b</sup> 10%/25%/50% of the consumers report they would discard and not prepare the product for dinner based on odour.  
<sup>c</sup> The total viable count of psychrotrophic bacteria has reached log 7 cfu/g.

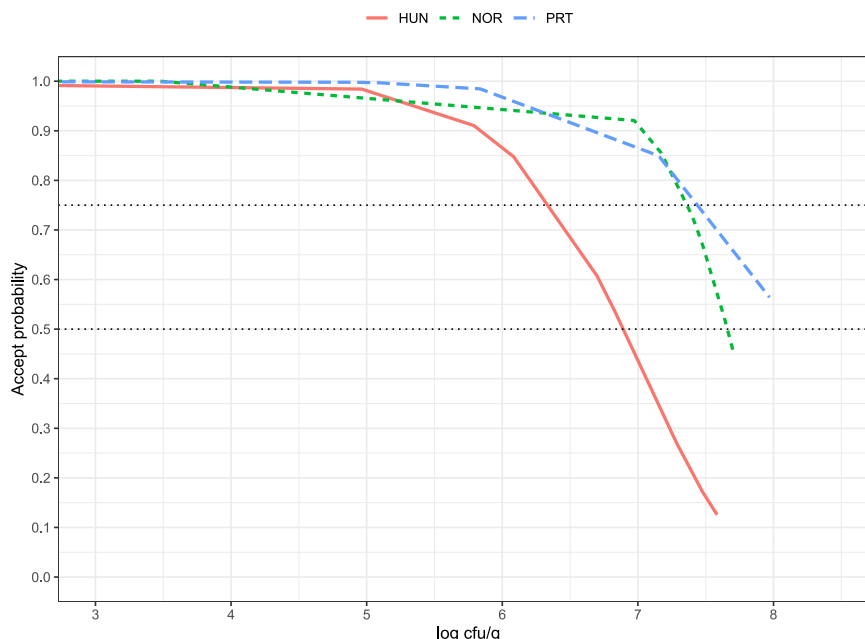
Fig. S2). For the Norwegian samples, there were no observations between approximately log 3 and log 7.5 cfu/g under the 8 °C scenario, so this comparison was limited to the overlapping range. Also, for a food producer, one microbial threshold acquired from a model based on two temperatures is more useful than different thresholds depending on different temperature scenarios. Interestingly, the degree of acceptance at different bacterial levels seemed similar for products from Norway and Portugal, despite different microbiota (Figs. 2 and 4). This is supported by an earlier study indicating that the bacteria dominating in the two products have a similar spoilage potential (Hansen et al., 2023). Acceptance seemed to decline more rapidly with increasing bacterial numbers for products from Hungary (Fig. 4). Although differences in consumer sensitivity cannot be excluded, the observed differences may reflect a higher spoilage potential of *Photobacterium* spp. in the Hungarian products. In fish, *Photobacterium* has been reported to cause spoilage at lower cell densities than *Shewanella* (Dalgaard, 1995).

3.5. Methodology for shelf-life testing

As far as we know, this study is the first combining a full consumer survival analysis with microbiota analysis attempting to define acceptable levels of bacteria after storage. The approach in the present study is similar to the QMSRA that has been suggested earlier (Tsaloumi et al., 2023), but using a consumer study aligned with methodology recommended to assess the sensory shelf life of products, and incorporating culture independent analysis to monitor the spoilage microbiota together with TVC (instead of selective agars). Although the present approach could be used to model acceptance using single spoilage bacteria as predictors, the TVCs were used since known spoilage bacteria were dominating at the end of the sensory shelf life in all three products. However, using models with known spoilage bacteria may be relevant for other products. Table 3 compares the declared shelf life with shelf life estimated using different criteria: 10%, 25% or 50% of consumer rejection (based on odour), and TVC threshold of log 7 cfu/g. Under temperature abuse, the criteria closest to declared shelf life across all three chicken products were 50% and 25% rejection. This aligns with the recommended threshold levels from (Hough and Garitta, 2012). Using a predetermined rejection criterium may help producers set a consumer-anchored shelf life. Assuming that the shelf life set by the chicken producers in the present study is based on long-term experience with their products, using 25%-50% rejection rates at abuse temperature could be a benchmark criterium for shelf-life assessments.

If a universal TVC threshold of 7 log cfu/g had been used to set the shelf life, it would yield large differences in acceptance at the declared date across products (50% to >90%, Fig. 4). Using a criterium such as 25% rejection across both temperature scenarios would lead to corresponding microbial thresholds of 6.4, 7.4 and 7.3 log cfu/g in Hungary, Norway and Portugal. Rather than a universal threshold of 7 log cfu/gram, the consumer-anchored criterium can define a microbial threshold to be used in quality control.

It should be emphasized that the approach proposed here is a methodological approach, that may be applied by producers and researchers using their own consumer and microbial data, rather than a



**Fig. 4.** Fraction of consumers that accept (claim they would prepare the sample of chicken fillet for dinner based on its odour) in Hungary, Norway and Portugal. The fraction of consumers is plotted against the bacterial numbers (log cfu/g). 25% and 50% rejection rates shown as horizontal lines.

predictive model based on the numerical thresholds derived in this study. The product-specific thresholds reported in Table 3 are illustrative outputs of applying the approach to these three products and are not intended as universal values to be adopted directly nor as a causal attribution of the differences to any single factor.

The results support recommendations to complement traditional cultivation with culture-independent microbiota analyses in duration tests (Everis, 2019). Standard microbial analyses may underestimate bacterial numbers by not supporting the growth of all bacteria that could spoil the product. For example, *Photobacterium* spp. may have been overlooked for years because of strict growth requirements, while analyses not requiring cultivation can reveal presence of such bacteria (Dourou et al., 2021; Hilgarth et al., 2018). Also, it is recommended to perform duration tests on at least three batches to cover variations in contamination levels. A companion year-long study of two of the present producers (Moen et al., 2026b) indicates that the dominant spoilage community at the end of shelf life is stable across batches and seasons within a producer, whereas initial load, growth dynamics and thus the time to reach spoilage vary considerably between batches and seasons. This suggests that the consumer-anchored microbial threshold may be relatively transferable across batches, but that translating it into a declared shelf life in calendar days requires replication spanning seasons rather than batches produced under similar conditions alone. In the duration tests, both TVC and microbiota composition should be included. If quality control detects sensory deviations not explained by total counts, shifts in microbiota are plausible drivers and should be identified. Such shifts are more likely to be detected by analysis not requiring cultivation than using pre-defined selective media.

In the present study, a temperature abuse of 8 °C was selected based on the recommendation to utilize the 95% percentile and refrigerator temperature data from Norway and Portugal (no data from Hungary) (Bonanno et al., 2024; Dumitraşcu et al., 2020). Reasonably foreseeable temperatures should take into account the target group, and varies between 7 and 12 °C in different guidelines (Koutsoumanis et al., 2020; Everis, 2019; Food Standards Scotland, 2025; MPI, 2016). No statistical difference in the relationship between acceptance and TVC for the two temperatures tested was found in the present study. Still, both optimal and abuse temperatures should be used in duration tests.

### 3.6. Study limitations and future research

The aim of the current study was to address the relationship between microbial development and consumer decisions to prepare or discard raw chicken. To capture microbial diversity, products from three countries (Hungary, Norway, Portugal) packed with different gas compositions and with variation in declared shelf-life were selected. The study design was not targeted to identify underlying causes for the differences found (e.g., hygiene in the food factory, packaging method) or to study

differences between consumer acceptance or products from different countries. Because the products differ simultaneously in country, processor, packaging and consumer group, these factors are confounded and the design does not support attributing the observed differences to any single one of them. Accordingly, the product-specific microbial thresholds reported (Table 3) should be regarded as illustrative outputs of applying the methodology to each product individually, rather than as a finding that any single factor determines the threshold. The methodology itself is applied to each product separately and does not require these between-product differences to be explained. Nevertheless, the results suggest that the great variation in microbiota of spoiled chicken reported in the literature reflects real differences in microbiota of the products, not only artifacts due to differences in methodologies. To identify the spoilage potential of different bacteria or set threshold levels for specific spoilage bacteria, future research could focus on challenge tests with different spoilage bacteria and relate to sensory profiles or consumer acceptance. To describe taxon-specific development, TVC measured on general media was combined with relative abundance from microbiota profiling. This approach has important limitations. Relative abundance from 16S rRNA gene amplicon sequencing is affected by primer bias, PCR amplification bias, and variation in DNA extraction efficiency, and it does not directly represent absolute abundance. Combining such relative abundance estimates with culture-based TVC therefore compounds uncertainty. Accordingly, the resulting taxon-specific counts should be interpreted only as approximate indicators of microbial dynamics, and conclusions about the spoilage contribution of individual taxa should be treated with caution. An alternative could be to use selective media, but reliable selective media are not available for all spoilage bacteria, so this approach cannot be used universally.



A limitation of the experimental design was that consumers got small samples to evaluate and that the evaluation was performed in laboratory environments. Arranging the evaluation in consumer kitchens and/or using whole product packages to obtain a more realistic evaluation, would have required storing and distributing to consumers >1200 packages of chicken breasts at each study location and was not possible for practical reasons. Consumer decisions to eat or discard food in real-life situations may be influenced by several contextual factors such as date labelling, product appearance, knowledge of the storage history, and individual attitudes toward food safety and waste. These factors were not addressed in the present experimental setting and may affect consumer rejection behaviour. This design was intentional as including such factors would have introduced confounders that obscure the microbiological signal of interest. Also, the methodology is consistent with established methodology for sensory shelf-life studies based on controlled consumer panels (Hough, 2010; Hough and Garitta, 2012).

Similarly, only odour of raw meat was evaluated, although appearance of the raw product, taste and odour of cooked product may also affect acceptance and discarding. Odour was deliberately chosen because earlier research has concluded that the most important factor related to microbial spoilage and rejection is the intensity of the odour, although the appearance or texture may also affect the perceived quality (Dold et al., 2022; Franke et al., 2017; Katiyo et al., 2020). Moreover, the literature recommends not including more than 8–10 samples in a single session of sensory evaluations for reliable results (Lawless and Heymann, 2010), especially with untrained assessors. Therefore, we limited the consumer experiment to 11 raw fillet samples, slightly over the maximum boundary.

Future studies may explore consumer decision-making in more realistic contexts, where more products and situational cues are available, but with a narrower number of storage scenarios. In the context of minimizing food waste, more knowledge about shopping and consumer storage practices are needed to estimate how many consumers will keep the food products until or beyond the end of the declared shelf life, and if so, if they would discard the product due to the date and/or odour.

The practical applicability of the proposed methodology varies considerably depending on producer scale and resources. The full methodology using large consumer panels ( $\geq 120$  participants), microbiota profiling by 16S rRNA amplicon sequencing, and survival analysis linking TVC to acceptance requires expertise and resources that are unlikely to be available to small and medium-sized producers. For such producers, the microbiota profiling component is most critical when the dominant spoilage community is unknown or when standard TVC media may underestimate specific organisms, as demonstrated here with *Photobacterium* spp. For high-volume products at large manufacturers, the full methodology is most relevant, and the investment in a robust consumer study and microbiota analysis is proportionate to the scale of production and the consequences of misaligned shelf-life declarations. Future work could explore whether simplified versions of the methodology, for example using smaller trained panels or targeted qPCR for known spoilage taxa could make the approach more accessible to smaller operations.

A further opportunity is to integrate the consumer-anchored approach with predictive microbiology. The microbial level corresponding to a predefined rejection rate provides a consumer-meaningful spoilage endpoint that growth models could be built to target. Robust prediction may, require early indicators of the level of the spoilage microbiota rather than TVC alone, since the latter has been shown to predict batch-specific shelf life poorly under MAP (Moen et al., 2026b).

#### 4. Conclusion

Based on the results in the present work we propose and demonstrate a consumer-driven methodological approach to shelf-life determination. Rather than providing transferable numerical thresholds, the approach defines a structured procedure that producers can apply to their own

products. It (i) defines a target acceptance level at end of life (e.g.,  $\leq 50\%$  reject for reasonable foreseen conditions), (ii) uses consumer panels and survival analysis to assess accept over time, (iii) supplements microbial cultivation techniques with culture-independent profiling to detect important genera that may be overlooked by standard cultivation techniques, and iv) combine consumer acceptance with microbial data to set microbial thresholds. It should however be noted that using large consumer panels is unrealistic for many food producers, so the approach is most relevant for high volume products and large manufacturers of food.

Information about the relationship between microbial development and consumer acceptance provides a relevant background to set a shelf life, critical limits and corrective actions in case of deviations. This could potentially decrease consumer complaints and enhance trust. Guidelines for the industry should be updated to propose scientifically sound sensory analysis and modern methodologies for microbial analysis. Preparation of guidelines should involve inter-disciplinary teams with experienced members not only from the area of food safety but also from sensory and consumer sciences, and across industry as well as academia.

By combining microbial monitoring with consumer acceptance data, the proposed approach provides new insight into how microbial dynamics translate into consumer rejection during product storage. This integrated perspective may support more informed shelf-life determination and contribute to improved quality management practices in the food supply chain. This methodological approach may therefore contribute to more realistic shelf-life determination by aligning microbiological criteria with consumer perception of product quality.

#### CRedit authorship contribution statement

**Solveig Langsrud:** Conceptualization, Funding acquisition, Investigation, Project administration, Writing – original draft, Writing – review & editing. **Aira Joy Aquino:** Investigation, Methodology, Validation, Writing – original draft, Writing – review & editing. **Beatriz Nunes Silva:** Investigation, Methodology, Validation, Writing – original draft, Writing – review & editing. **Ingunn Berget:** Conceptualization, Formal analysis, Methodology, Visualization, Writing – original draft, Writing – review & editing. **Paula Teixeira:** Conceptualization, Investigation, Methodology, Writing – original draft, Writing – review & editing. **Maria João P. Monteiro:** Investigation, Methodology, Writing – original draft, Writing – review & editing. **Dávid Szakos:** Investigation, Methodology, Writing – original draft, Writing – review & editing. **Gyula Kasza:** Investigation, Methodology, Writing – original draft, Writing – review & editing. **Mari Øvrum Garder:** Conceptualization, Investigation, Methodology, Writing – review & editing. **Annette Fagerlund:** Funding acquisition, Investigation, Methodology, Writing – original draft, Writing – review & editing. **Merete Rusås Jensen:** Investigation, Methodology, Writing – original draft, Writing – review & editing. **Ingrid Måge:** Investigation, Visualization, Writing – review & editing. **Valerie Lengard Almlil:** Methodology, Writing – original draft, Writing – review & editing. **Birgitte Moen:** Conceptualization, Formal analysis, Investigation, Methodology, Visualization, Writing – original draft, Writing – review & editing.

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.fm.2026.105226>.

## Data availability

Data has been shared in Zenodo.

Using consumer acceptance to guide microbial thresholds for the shelf life of fresh chicken breasts (Original data) (Zenodo)

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