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Sodium reduction in cooked meat products without compromising microbiological safety: A predictive microbiology framework for food safety assessment

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ABSTRACT

Sodium plays a critical role in the microbiological stability of cooked meat products, yet regulatory and consumer pressure to reduce sodium intake creates formulation challenges where safety margins are poorly defined. This study quantified the effect of sodium concentration (196–980 mg/100 g), pH, and water activity on the growth dynamics of lactic acid bacteria (*Leuconostoc* sp.) and *Salmonella enterica* serovar Typhimurium in a cooked meat analogue — designed to allow independent control of physicochemical variables — and integrated these relationships into a predictive microbiology framework for reduced-sodium formulation design. Primary modeling using the Baranyi–Roberts model best described experimental growth curves, and secondary modeling via a modified Norrish equation quantified the effect of environmental factors on maximum specific growth rate (μ_{max}). Global sensitivity analysis identified μ_{max} and maximum population density as the dominant sources of variability in model predictions. Increasing sodium concentration significantly reduced μ_{max} in both organisms, with LAB exhibiting greater tolerance than *Salmonella*. Multi-criteria optimization identified ~500 mg sodium/100 g as the microbiologically safe optimum — representing a ~ 41% reduction relative to internationally comparable regulatory limits for cooked ham (846 mg/100 g)— while preserving inhibitory conditions against pathogens. Independent validation in cooked ham confirmed model accuracy with deviations below 5% for growth rate and 2% for shelf-life, predicting a shelf-life of ~18 days. This framework provides an organism-specific, quantitative basis for designing reduced-sodium cooked meat products with defined microbiological safety margins, directly applicable to reformulation strategies in industry and regulatory contexts.

1. Introduction

The food industry faces the challenge of balancing sensory quality, food safety, and consumer health in processed foods (Aaslyng et al., 2014; Calderón, 2021; Wang et al., 2023). Sodium is a key ingredient in processed meats, enhancing flavor and acting as a preservative (Barcenilla et al., 2022; Galvão et al., 2014; Matthews and Strong, 2005). However, excessive sodium intake is associated with public health concerns such as hypertension and cardiovascular diseases (Aaslyng et al., 2014; Barcenilla et al., 2022; Galvão et al., 2014; Matthews and Strong, 2005; Wang et al., 2023).

As a result, international and national regulations have progressively limited sodium concentrations in processed foods. The World Health Organization recommends reducing population sodium intake to below 2000 mg/day ("FDA," 2021; World Health Organization, 2012), and regulatory agencies in the United States and the European Union have implemented voluntary and mandatory sodium reduction targets for processed meat products (FDA [WWW Document], 2021). In Colombia, the Ministry of Health and Social Protection, through Resolution 2013 of 2020, established maximum sodium levels for certain processed products by 2024, including ham (846 mg/100 g), mortadella (912 mg/100 g), and sausage (857 mg/100 g) (Colombia. Ministerio de Salud y Protección Social, 2020) - limits comparable to those adopted in other regulatory frameworks worldwide. These requirements challenge the meat industry globally to ensure microbiological safety and product quality while meeting sodium reduction targets (Barcenilla et al., 2022; Calderón, 2021; FDA [WWW Document], 2021).

Sodium reduction in cooked meat products poses a complex

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technological challenge, as sodium not only influences organoleptic properties but also acts as a barrier against spoilage and pathogenic microorganisms (Calderón, 2021; Chen et al., 2023; De Oliveira et al., 2015; Doyle and Glass, 2010; Galvão et al., 2014; Rodríguez et al., 2022; Solomando et al., 2023; Wang et al., 2023). Lactic acid bacteria (LAB) and *Salmonella* spp. are particularly relevant for ensuring both product safety and shelf-life. LAB dominates meat matrices across technological processes and storage conditions, and their versatility allows growth under diverse environments, including variations in solutes, pH, and temperature (Li et al., 2023; Yang et al., 2020; Yang et al., 2018). While LAB contribute to biopreservation through acidification and antimicrobial compound production, they can also act as significant spoilage organisms in meat products. However, the dominant spoilage microbiota depends on product formulation, storage atmosphere, and packaging conditions — for instance, *Pseudomonas* spp. typically prevail under aerobic storage, whereas LAB often dominate in vacuum- or modified-atmosphere-packaged meats (Kokkosi et al., 2024; Rood et al., 2025; Xu et al., 2021).

At the same time, controlling *Salmonella* in meat products is critical because it represents a significant public health risk (ICONTEC, 2008; da Silva et al., 2022; Soro et al., 2023). While *Listeria monocytogenes* is also a relevant pathogen, particularly in ready-to-eat and refrigerated foods, *Salmonella* remains the leading cause of foodborne outbreaks associated with raw and processed meats worldwide. These pathogens can withstand adverse conditions, including hyperosmotic stress, highlighting the importance of understanding their response to sodium reduction in meat products (Bamidele et al., 2023; Barcenilla et al., 2022; Calle et al., 2021; Thomas et al., 2020; Werlang et al., 2021).

Characterizing microbial growth under different sodium conditions enables the implementation of preventive and corrective measures that ensure product safety and quality (Gutiérrez-Chocoza et al., 2023; Hansen et al., 2021; Huang, 2017; Huang, 2013). In this context, predictive microbiology has emerged as a valuable tool for forecasting LAB and *Salmonella* population dynamics in response to sodium variation in cooked meat products (Antunes-Rohling et al., 2019; Gutiérrez-Chocoza et al., 2023; Hansen et al., 2021; Huang, 2017; Huang, 2013; Werlang et al., 2021). Predictive mathematical models have been successfully applied to study bacterial viability in meat products and its relationship with key physicochemical parameters such as temperature, humidity, pH, water activity (a_w), and other growth-limiting factors, including oxygen, solutes, acidity, and pressure. These models allow correlations between physicochemical parameters and microbial stability during shelf-life (Engstrom et al., 2020; Gutiérrez-Chocoza et al., 2023; Hansen et al., 2021; Koseki et al., 2021).

This study aimed to characterize the growth dynamics of LAB and *Salmonella* under different sodium concentrations in a cooked meat analogue, using predictive microbiology tools to describe and compare their behavior. The modeling framework was applied to identify sodium levels that ensure microbial safety while meeting regulatory and technological requirements and was further validated in cooked ham.

2. Materials and methods

All abbreviations and mathematical symbols are defined in Supplementary File S1. The experimental and modeling approach comprised three stages: (i) data generation, including strain selection, inoculum preparation, and cooked meat analogue formulation; (ii) model development, involving primary, secondary, and dynamic modeling with parameter estimation and sensitivity analysis; and (iii) application and validation, including sodium concentration optimization and shelf-life estimation in cooked ham.

2.1. Growth matrices, microbial strains and inoculum preparation

Lactic acid bacteria (LAB) and *Salmonella* spp. were used. LAB: *Leuconostoc* sp., donated by Zenú® (Medellín, Antioquia, Colombia), isolated from cooked meat products at the end of shelf-life and identified using the automated Vitek® system (Biomérieux). *Salmonella enterica* subsp. enterica serovar Typhimurium ATCC 14028™ is a well-characterized international reference strain widely used in predictive microbiology, food safety, and challenge-test studies due to its reproducibility and physiological stability under controlled experimental conditions. Strains were stored at −18 °C: *Salmonella* in BHI broth (Merck) with 70% BHI and 30% glycerol; LAB in MRS broth (Merck) with 70% MRS and 30% glycerol.

For inoculum preparation, activated strains were plated on TSA (*Salmonella*) or MRS agar (LAB). Colonies were suspended in 0.1% sterile peptone water to 0.5 McFarland (~10^7 CFU/mL). Successive dilutions were performed to reach a final inoculum of ~10^2 CFU/mL, corresponding to 1% inoculation of the samples. The inoculum was aseptically added to the samples and homogenized under sterile conditions prior to storage.

Importantly, inocula were intentionally prepared from agar-grown cultures and inoculated at low cell densities to represent low-level initial contamination and microbial populations developing during refrigerated storage of cooked meat products, rather than optimal exponential-phase laboratory cultures. Consequently, the initial physiological state of the cells reflects an adaptation phase to the food matrix and storage conditions, consistent with applied food systems.

The reproducibility of the inoculum preparation procedure was assessed statistically for both microorganisms (See Supplementary File S2).

2.2. Cooked meat analogue preparation

A cooked meat analogue was formulated in the laboratory using a mixture of glucose (20 g/L), casein (10–15 g/L), meat extract (10 g/L), meat peptone (5 g/L), sodium chloride (5 g/L), yeast extract (4 g/L), triammonium citrate (2 g/L), magnesium sulfate (0.2 g/L), and agar-agar (15 g/L). The composition was then adjusted to match the protein, moisture, and total solids content specified in the Colombian Technical Standard NTC 1325:2008, while excluding preservatives and high sodium levels (Bradford, 1976; ICONTEC, 2009). This simplified formulation was intentionally designed to isolate the effect of sodium concentration on microbial behavior; therefore, the predictive framework should be interpreted within the context of preservative-free cooked meat systems (ISO 20976-1) (ISO, 2019).

2.3. Experimental procedure

A categorical factorial design (triplicate) was applied using meat analogue formulations containing 196, 740, and 980 mg sodium/100 g. These concentrations were selected to represent low-, intermediate-, and high-sodium processed meat conditions, using sufficiently separated sodium levels to facilitate discrimination of microbial kinetic responses across the evaluated range. Samples were sterilized at 121 °C for 15 min and inoculated independently with LAB and *Salmonella* at 10^2 CFU/g.

Products were stored at 0–6 °C under refrigerated conditions representative of commercial cold storage, with sampling every three days to enumerate microbial counts on TSA for *Salmonella* (ISO 6579:2017) (ISO, 2017a) and MRS for LAB (ISO 15214:1998) (ISO, 1998) (Buelvas Salgado, 2013; Castro et al., 2008). In parallel with microbiological sampling, pH was measured in triplicate at each time point using a calibrated penetration pH electrode (ISO 2917:1999) (ISO, 1999). Water activity (aw) was determined using a dew-point water activity meter (AquaLab Series 4, Decagon Devices, USA) at 25 °C ± 0.5 °C in triplicate, in accordance with ISO 18787:2017 (ISO, 2017b). The influence of sodium concentration on apparent maximum specific growth rate (μ_{max}), pH, and a_w was assessed using statistical models (Statgraphics Centurion XIX, Statgraphics Technologies, USA).

2.4. Model validation in cooked meat products

Two batches of traditional cooked ham were prepared by Zenú® (Medellín, Antioquia, Colombia) following NTC 1325:2008 (ICONTEC, 2008) at the sodium concentration predicted as optimal. Upon receipt, samples were inoculated with LAB and *Salmonella* as described in the inoculum preparation section. Samples were stored under refrigeration (0–6 °C), and microbial counts were determined in triplicate every three days. Model predictions were compared with experimental data using the bias factor (Bf) and accuracy factor (Af), with values close to 1 indicating good predictive performance (Baranyi and Roberts, 1995).

$$Af = 10^{\sum \frac{|pred-obs|}{n}} \quad (1)$$

$$Bf = 10^{\sum \frac{pred-obs}{n}} \quad (2)$$

Here, *obs* is the observed value, *pred* the predicted value, and *n* the number of observations.

Shelf-life was further estimated using the primary optimized model, based on the predicted maximum specific growth rate (μ_{max}) (Baranyi and Roberts, 1995). This approach provided a quantitative estimate of the time required for microbial counts to reach the spoilage threshold under the tested storage conditions.

2.5. Predictive modeling

2.5.1. Primary growth models

Five primary microbial growth models were evaluated to describe LAB and *Salmonella* kinetics under different sodium concentrations. The mathematical expressions of the models are summarized in Table 1.

Equations adapted from (Baranyi and Roberts, 1995; Buelvas Salgado, 2013; Castro et al., 2008; González et al., 2022; Mai et al., 2022).

Here, $P(t)$ is the bacterial population density (log CFU/g) at time t, P_0 is the initial population density, P_{max} is the maximum population density (log CFU/g), λ (hours) is the lag phase, μ_{max} is the maximum specific growth rate (h^{-1}), and the remaining parameters define model-specific shape and transition functions. In the Baranyi–Roberts model, parameters such as m and q_0 are associated with the smoothness of phase transitions and the physiological adaptation state of the microbial population during the lag-to-exponential growth transition. A complete description of abbreviations, mathematical notation, and model parameters is provided in Supplementary File S1. Model selection was based on goodness-of-fit criteria and the capacity of the model to adequately reproduce the different microbial growth phases under the evaluated experimental conditions.

Table 1
Primary microbial growth models to describe the kinetics of LAB and *Salmonella* under different sodium concentrations.

Model	Equation
Modified Gompertz	$P(t) = P_0 + (P_{max} - P_0)\exp\left\{ -exp\left[\frac{\mu_{max}{}^{*}e}{P_{max} - P_0}(\lambda - t) + 1\right]\right\}$ (3)
Logistic	$P(t) = P_0 + \frac{(P_{max} - P_0)}{1 + exp\,(\mu_{max}(t-\lambda))}$ (4)
Stannard	$P(t) = P_0(1 + exp[-(\mu_{max} + K)(t - P)])$ (5)
Richards	$P(t) = P_0 + \frac{P_{max} - P_0}{(1 + V\,exp[-\mu_{max}(t-\lambda)])^{\frac{1}{V}}}$ (6)
Baranyi-Roberts	$P(t) = P_0 + \mu_{max}\,A(t) - \frac{1}{m}ln\left\{1 + exp[m\,\mu_{max}(t - \lambda)]\frac{P_{max} - P_0}{exp(m)}\right\}$ with $A(t) = t + \frac{1}{V}ln\left(\frac{exp(-Vt) + q_0}{1 + q_0}\right)$ (7)

2.5.2. Secondary models

To describe the effect of sodium on the maximum microbial growth rate (μ_{max}), an exponential decay model proposed by Norrish (Norrish, 1966) was used. The Norrish model was selected because it provides a parsimonious yet biologically meaningful description of the inhibitory effect of solutes such as sodium on microbial growth. This exponential decay function has been widely applied in predictive microbiology to represent the relationship between water activity, solute concentration, and μ_{max}, providing both empirical accuracy and ease of integration into secondary models.

$$Zp(pH, a_w, \mu_{max}) = A\,exp\left(-B\,[Na^+]^2\right) \quad (8)$$

Here, *Zp* represents the experimentally estimated response variable modelled as a function of sodium concentration, specifically the maximum specific growth rate (μ_{max}), pH, or water activity (a_w). The predicted μmax values obtained from the secondary model were subsequently incorporated into the optimization framework and dynamic population simulations. A is a scale parameter, B is a stability constant, and [Na^+] represents sodium concentration (mg/100 g).

A structural modification was applied by normalizing the independent variable using a scale-range transformation (Rodríguez González and Ugalde Saborio, 2021) and incorporating an asymptotic term (C) to allow the function to approach a non-zero value at extreme sodium concentrations (Amrane, 2001).

$$Z_p[\mu_{max}] = A\,exp\left(-B\frac{[Na^+]^2 - [Na^+]^2 min}{[Na^+]^2 max - [Na^+]^2 min}\right) + C \quad (9)$$

2.5.3. Dynamic population model

To explore potential population-level interaction dynamics in reduced-sodium cooked meat systems, we implemented a dynamic population model integrating Lotka–Volterra interaction terms into a primary growth framework. Kinetic parameters were experimentally estimated from independent monoculture experiments and subsequently incorporated into the dynamic simulations. This approach extends beyond single-population primary models by enabling exploratory evaluation of potential interspecific effects, such as competitive pressure and population displacement, at the simulation level (Caballero et al., 2024). Therefore, the model was intended as an exploratory interaction framework rather than as a mechanistic coculture model directly parameterized from mixed-culture experiments.

Microbial growth and interactions between lactic acid bacteria (LAB) and *Salmonella* spp. were described as follows:

$$\frac{dP_i}{dt} = \mu_{maxi}\frac{P_i \cdot q_i}{(1 + q_i)}\left(1 - \frac{P_i + \beta_i \cdot P_i}{P_{max,i}}\right) - K_i(P_i - Pres_i) \quad (10)$$

$$\frac{dq_i}{dt} = \mu_{maxi} q_i \quad (11)$$

Where, i indicates the microbial population (LAB or *Salmonella*), P is the bacterial population density (log CFU/g), P_{max} is the maximum population density (log CFU/g), P_{res} is the residual population density (log CFU/g), K is the death rate (h^{-1}), μ_{max} is the maximum growth rate (h^{-1}), q is the physiological state variable associated with microbial adaptation, and β represents the interaction coefficient between microbial populations.

2.6. Model fitting and validation

Model parameters were estimated in Python 3.12 using NumPy and SciPy. For primary models, the *least_squares* function with Trust Region or Levenberg–Marquardt algorithms were applied, while secondary models were fitted with *curve_fit*. The best-fitting model was selected according to the lowest mean squared error (MSE) and highest coefficient of determination (R^2):

$$MSE = \sum_{i=1}^{n} \frac{(obs - pred)^2}{n - q} \tag{12}$$

$$R^2 = \frac{\Sigma\,(pred - obs^*)^2}{\Sigma\,(obs - obs^*)^2} \tag{13}$$

Here, *obs* = observed value, *obs** = average observed value, *pred* = predicted value, *n* = number of observations, *q* = number of parameters.

2.7. Sensitivity analysis

Global sensitivity was evaluated using the Morris method (Wang et al., 2023) to quantify the influence of model parameters. Each input was perturbed by ±10% of its nominal value, and the resulting elementary effects (Δ) were computed as the change in the model output divided by the perturbation step. Sensitivity was summarized with two:

$$\mu^* = \frac{1}{r}\sum_{j=1}^{r} |\Delta_j| \tag{14}$$

$$\sigma = \sqrt{\frac{1}{r-1}\sum_{j=1}^{r} (\Delta_j - \mu)^2} \tag{15}$$

where Δ_j denotes the *j*-th elementary effect and *r* the number of perturbations. The mean of the absolute effects (μ^*) reflects the overall importance of each parameter, whereas the standard deviation (σ) captures interactions or non-linear effects.

This formulation justifies the use of dispersion plots (μ^*, σ), which allow a visual classification of parameters according to their relative influence and interaction strength. The analysis was implemented in Python™ 3.12 using the SALib library (*sample.morris* function).

2.8. Optimization of sodium concentration

The minimum sodium concentration that simultaneously reduced the estimated growth potential of LAB and *Salmonella* was determined using a multivariable optimization framework based on desirability functions. Python™ 3.12.0 was employed with the *scipy.optimize* library, using the *minimize_scalar* function with the Brent method (Barcenilla et al., 2022), and numpy.*searchsorted* algorithm for grid adjustment.

For each microorganism, an individual desirability function (d_i) was constructed under minimization criteria:

$$d_i\left(\mu_{max,i}(x)\right) = \begin{cases} 1, & \text{if } \mu_{max,i(x)} \le L_i \\ \left(\dfrac{U_i - \mu_{max,i}(x)}{U_i - L_i}\right)^{s_i}, & \text{if } L_i < \mu_{max,i(x)} < U_i \\ 0, & \text{if } \mu_{max,i(x)} \ge U_i \end{cases} \tag{16}$$

Here, $\mu_{max,i}(x)$ is the estimated maximum specific growth rate at sodium concentration *x*, L_i and U_i are the experimental lower and upper bounds, and s_i is a shape parameter.

The global desirability was then calculated as the geometric mean of the individual desirabilities:

$$D(x) = \left(\prod_{i=1}^{n} d_i\left(\mu_{max,i}(x)\right)\right)^{1/n}, n = 2 \tag{17}$$

where $n = 2$ corresponds to LAB and *Salmonella*. The optimal sodium concentration was identified as:

$$x^* = arg_x \max D(x) \tag{18}$$

Maximization of *D(x)* was restricted to the experimental sodium concentration range (Veatch, 2021). When the optimum did not coincide with an experimental grid point, the *numpy.searchsorted* function was applied to assign the nearest sodium level.

3. Results and discussion

3.1. Physicochemical characterization of the meat analogue

The physicochemical characterization of the laboratory-prepared meat analogue is summarized in Supplementary File S3. Protein content (11.4% *w*/w) complied with the Colombian technical standard (NTC 1325:2008, ≥11%), while moisture (93.8% w/w) slightly exceeded the standard limit (≤92.0%). The baseline sodium concentration (196 mg/100 g) was substantially lower than the maximum values established by the Colombian Ministry of Health (Resolution 2013 of 2020; e.g., 846 mg/100 g for ham)- a limit comparable to those established in other regulatory frameworks worldwide, consistent with the objective of evaluating sodium-reduced cooked meat matrices.

Despite the slight deviation in moisture, the analogue provided a controlled and reproducible experimental matrix in which sodium concentration could be manipulated as an independent variable while holding other compositional factors constant. This design choice is critical for isolating the specific contribution of sodium to microbial growth dynamics, as confounding effects from protein, fat, or carbohydrate variation are minimized. Similar approaches have been reported by (Wang et al., 2022) and (Fraqueza et al., 2021), who documented significant shifts in meat product microbiota associated with variations in salt concentration, supporting the use of standardized analogues as a valid platform for predictive microbiology studies in reduced-sodium systems.

3.2. Effects of sodium concentration and microorganism type

Analysis of variance revealed that sodium concentration significantly affected all measured response variables: maximum growth rate (μ_{max}, $p = 0.002$), pH ($p < 0.0001$), and water activity (a_w, $p = 0.0001$). Microorganism type also influenced μ_{max} ($p = 0.011$) and pH ($p < 0.0001$), but not a_w ($p = 0.059$), and the interaction between sodium concentration and microorganism type was significant for all three variables. Full ANOVA tables and interaction plots are provided in Supplementary File S4 (Tables S4.1 and S4.2).

The maximum specific growth rate (μ_{max}) decreased progressively as sodium concentration increased from 196 to 980 mg/100 g, with LAB exhibiting slightly higher values than *Salmonella*, suggesting greater osmotic tolerance in LAB. This differential response is consistent with (González et al., 2022) and (Xing et al., 2023a), who described contrasting salt stress responses across bacteria with different metabolic strategies, and with (Li et al., 2021), who reported alterations in lag phase and exponential growth of *E. coli* BW25113 under NaCl stress. (Yang et al., 2018) and (Werlang et al., 2021) further highlighted that sodium effects are influenced by interactions with environmental and physiological variables, supporting the concept that osmotic stress responses are species-dependent. These findings also support future studies extending the proposed predictive framework to relevant ready-to-eat meat pathogens such as *Listeria monocytogenes*, particularly under reduced-sodium refrigerated storage conditions.

pH was primarily determined by microorganism type rather than sodium concentration. LAB acidified the medium substantially, reaching average values near pH 5.7, while *Salmonella* maintained a less acidic environment near pH 7.3. This contrast reflects the fermentative metabolism of LAB and its associated organic acid production, as opposed to the respiratory metabolism of *Salmonella* (Díaz-Montes, 2023; Kim et al., 2021; Yang et al., 2018). The pronounced acidification by LAB at reduced sodium concentrations is particularly relevant from a food safety perspective, as it suggests a potential bioprotective effect that could partially compensate for the loss of sodium as a hurdle. However, excessive acidification may also influence sensory attributes such as flavor and overall product acceptability, which should be evaluated in future studies.

Water activity decreased progressively from 0.985 to 0.958 as

sodium concentration increased from 196 to 980 mg/100 g, irrespective of microorganism type, consistent with the well-established role of NaCl as a water activity depressant (Díaz-Montes, 2023; Koutsoumanis et al., 2004; Werlang et al., 2021). This reduction in free water availability creates a progressively more restrictive environment for microbial growth, particularly for organisms with limited osmotolerance. Taken together, these results indicate that sodium concentration modulates microbial growth both directly — through osmotic stress on μ_{max} — and indirectly, through its effect on the physicochemical environment in which microbial activity occurs. However, the persistence of microbial growth at the highest sodium concentration evaluated suggests that sodium reduction strategies may require combination with additional preservation hurdles to ensure effective microbial control.

3.3. Primary modeling of microbial growth

The growth of LAB and *Salmonella* under sodium concentrations of 196, 740, and 980 mg/100 g was fitted to five primary models: modified Gompertz, Logistic, Stannard, Richards, and Baranyi-Roberts. Model performance was evaluated using R^2 and MSE (Supplementary Tables S5.1 and S5.2 in File S5).

The Baranyi-Roberts model provided the best fit across all sodium concentrations and both organisms. For LAB, it yielded $R^2 > 0.98$ and MSE < 0.07, outperforming the Gompertz and Logistic models ($R^2 \leq 0.99$ and ≤ 0.86; MSE ≥ 0.01 and ≥ 0.36, respectively). For *Salmonella*, Baranyi-Roberts also achieved the highest accuracy ($R^2 > 0.94$, MSE ≤ 0.03). This superior performance reflects the model's capacity to capture all four growth phases — lag, exponential, stationary, and decline — including the transition dynamics that simpler models such as Gompertz and Logistic tend to misrepresent (Huang et al., 2023; Juneja et al., 2025). Similar findings have been reported in complex food systems by (Huang et al., 2023) and (Juneja et al., 2025), confirming the broad applicability of Baranyi-Roberts for describing microbial kinetics under variable environmental conditions.

Kinetic parameters estimated by the Baranyi-Roberts model are presented in Table 2. In both organisms, maximum growth rate (μ_{max}) decreased as sodium concentration increased, confirming stronger growth inhibition at higher salinity and consistent with the osmotic stress physiology described by (Li et al., 2021). For LAB, the parameter *m* increased with sodium concentration, indicating a progressively smoother — and longer — transition from the adaptation to the exponential phase under high-salt conditions. At 196 mg/100 g sodium, both LAB and *Salmonella* exhibited rapid growth and short lag phases, whereas at 740–980 mg/100 g, growth rates decreased markedly and lag phases were extended (Fig. 1). However, residual Salmonella growth was still observed at 980 mg/100 g sodium, suggesting that sodium concentration alone may not be sufficient for complete microbial inhibition and that additional preservation hurdles could be required in reduced-sodium cooked meat systems.

LAB consistently exhibited higher μ_{max} and shorter lag phases than *Salmonella* across all sodium levels, suggesting greater physiological adaptability to osmotic gradients. This is consistent with differential osmotic adaptation strategies reported for these organisms in other matrices (Buelvas Salgado, 2013; González et al., 2022; Xing et al., 2023b).

3.4. Secondary modeling of microbial growth

The effect of sodium concentration on μ_{max}, pH, and water activity was described using a modified Norrish equation. The evaluated sodium concentrations provided broad separation across the reduced-sodium range, allowing clearer discrimination of sodium-dependent kinetic responses. Although the secondary models were constructed from three sodium levels, the fitted equations showed high goodness-of-fit values ($R^2 \geq 0.9890$, MSE ≈ 0) within the evaluated range, supporting the suitability of the proposed formulation for describing the observed experimental trends. Estimated parameters for each organism are summarized in Table 3.

Differences in the scale parameter (*A*) and stability coefficient (*B*) between organisms reflected distinct physiological responses to sodium concentration (Fig. 2). For both LAB and *Salmonella*, μ_{max} (panel a) and a_w (panel b) decreased progressively with increasing sodium concentration, consistent with the well-established inhibitory role of sodium on microbial growth through osmotic stress and reduction of free water availability. LAB displayed higher *A* values and stronger response gradients across the evaluated sodium range, suggesting differential adaptive responses to physicochemical variation. In contrast, *Salmonella* showed more stable parameter values, indicating a more consistent but less flexible inhibitory response.

pH responses diverged markedly between microorganisms (panel c): in LAB, pH increased with sodium concentration, possibly reflecting changes in acid production under osmotic stress conditions; in *Salmonella*, pH decreased, indicating a contrasting physiological response. This divergence reinforces the observation from Section 3.2 that pH dynamics in cooked meat systems are influenced predominantly by microorganism type rather than sodium concentration alone and underscores the importance of incorporating organism-specific secondary models when predicting microbial behavior in reduced-sodium formulations.

3.5. Dynamic population model

The dynamic population model integrated the kinetic parameters estimated from the primary and secondary modeling stages into a population-level interaction framework applicable to cooked meat systems. In this hierarchical approach, microbial growth kinetics were first described using the Baranyi–Roberts model, after which the influence of sodium concentration on microbial behavior was incorporated through the secondary model and subsequently used in the dynamic simulations. The physiological state variable q, inherited from the Baranyi–Roberts formulation, was retained in the coupled dynamic framework to account for microbial adaptation and lag-phase behavior during the initial stages of growth. Because the interaction framework was constructed from monoculture-derived kinetic parameters, lag-associated dynamics were independently estimated for each population and were not interpreted as coculture-specific adaptation effects.

An integrated Baranyi–Roberts/Lotka–Volterra framework (Caballero et al., 2024) was used to simulate the population dynamics of LAB and *Salmonella* in cooked meat analogue under sodium concentrations of 196, 740, and 980 mg/100 g. This coupled approach allowed simultaneous description of individual growth kinetics — maximum

Table 2
Kinetic parameters estimated by the Baranyi–Roberts model for LAB and *Salmonella* under different sodium concentrations. P_0: initial population density (log CFU/g); P_{max}: maximum population density (log CFU/g); μ_{max}: maximum specific growth rate (h^{-1}); q_0: physiological adaptation parameter.

Microorganism	$[Na^+]$ (mg/100 g)	P_0	P_{max}	μ_{max}	V	m	q_0
LAB	196	1.3085	9.1723	0.8498	0.0091	0.0115	0.0500
	740	1.5008	7.0669	0.0132	0.0445	0.7724	0.0368
	980	1.5646	5.2766	0.0049	0.0412	2.4136	0.0001
Salmonella	196	1.8294	8.9247	0.0228	0.0147	0.3240	0.0006
	740	1.5432	7.3718	0.0109	0.0120	0.7247	0.0031
	980	1.5509	4.9636	0.0099	0.0072	0.1653	0.8984

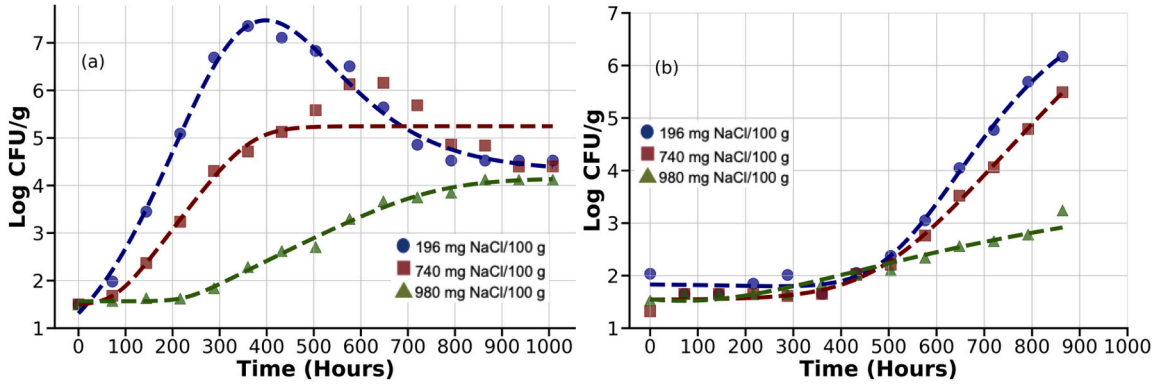

Fig. 1. Growth kinetics of lactic acid bacteria (LAB) and *Salmonella* spp. in a cooked meat analogue at different sodium concentrations (196, 740, and 980 mg/100 g), fitted with the Baranyi–Roberts model. (a) Lactic acid bacteria (LAB). b) *Salmonella* spp. Symbols represent experimental data, and dashed lines correspond to model predictions.

Table 3
Estimated parameters of the modified Norrish equation for LAB and *Salmonella* and statistical indices (R^2, MSE). A, B, and C correspond to fitted coefficients of the modified Norrish equation associated with scale, stability, and offset terms, respectively.

Microorganism	Variables	*A*	*B*	*C*	R^2	MSE
LAB	μ_{max}	0.8500	8.3272	−0.0002	1.0000	0.0000
	a_w	206.9689	0.00020	-4.7902×10^3	0.9899	0.0000
	pH	4.8837	2.4115	1.0985	1.0000	0.0000
Salmonella	μ_{max}	0.0235	2.9007	−0.0581	1.0000	0.0000
	a_w	0.9879	2.0498	−0.1477	1.0000	0.0000
	pH	7.4524	2.7359	−0.0693	1.0000	0.0000

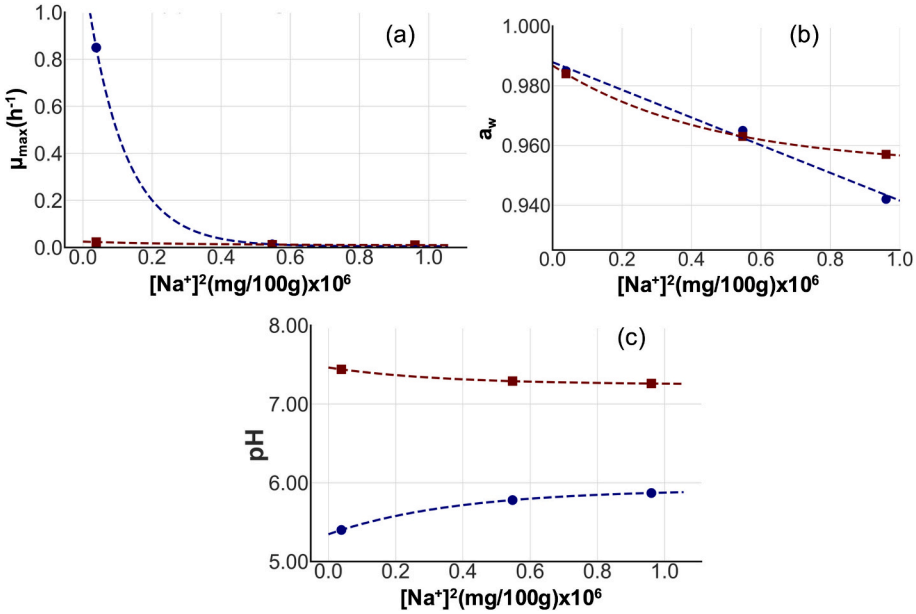

Fig. 2. Secondary models describing the effect of sodium concentration on microbial growth parameters using the modified Norrish equation. Main effect plots for (a) maximum growth rate (μ_{max}), (b) water activity (a_w), and (c) pH as a function of normalized sodium concentration $[Na^+]^2$ (mg/100 g). Dashed lines represent model predictions; symbols denote experimental data (● LAB; ■ *Salmonella*).

population density (P_{max}), residual population density (P_{res}), and maximum specific growth rate (μ_{max}) — together with exploratory interaction coefficients (β) and environmental inactivation rates (K). All kinetic parameters were expressed in h^{-1} units throughout the modeling framework.

Adjusted parameter estimates are summarized in Table 4. LAB exhibited a marked sodium-dependent response across all fitted parameters. LAB exhibited a marked sodium-dependent response: both P_{max} and μ_{max} decreased progressively with sodium concentration (Table 4), and a post-maximum decline phase was resolved at 196 and 740 mg/100 g but not at 980 mg/100 g, where growth was strongly suppressed throughout the experimental period.

For *Salmonella*, μ_{max} also decreased with increasing sodium concentration (0.01925, 0.01779, and 0.00728 h^{-1}), confirming a dose-dependent inhibitory effect of sodium on pathogen growth. However, P_{max} values approached the imposed biological upper bound (~9.0 log

Table 4
Adjusted parameters of the dynamic population model for LAB and *Salmonella* in cooked meat analogue. P_{max}: maximum population density (log CFU/g); P_{res}: residual population density (log CFU/g); μ_{max}: maximum specific growth rate (h^{-1}); K: death rate (h^{-1}) and β the interaction coefficient between microbial populations.

Microorganism	[Na^+] (mg/100 g)	P_{max}	P_{res}	μ_{max}	β	K	R^2	MSE
LAB	196	7.400	3.977	0.0525	2.50	0.0759	0.9956	0.0128
	740	6.270	4.037	0.0232	1.14	0.0415	0.9871	0.0267
	980	6.000	–	0.00821	5.00	–	0.9594	0.0417
Salmonella	196	8.995	–	0.01925	0.003	–	0.9638	0.0878
	740	9.000	–	0.01793	0.005	–	0.9861	0.0244
	980	8.965	–	0.007275	5.00	–	0.9844	0.0042

CFU/g) under all sodium conditions, indicating limited parameter identifiability due to the absence of a fully developed stationary phase during the experimental period. No death phase was detected for *Salmonella* under any condition. Model performance was satisfactory overall, with R^2 values ranging from 0.96 to 1.00 and MSE values between 0.004 and 0.088 log CFU/g.

The interaction component of the coupled framework was used as an exploratory simulation tool to evaluate potential population-level competition scenarios under reduced-sodium conditions. Because the present study was based exclusively on monoculture experiments, the β coefficients were not interpreted as experimentally validated measures of microbial competition. he asymmetric β estimates (LAB: 2.50 and 1.14; *Salmonella*: 0.003 and 0.005 at 196 and 740 mg/100 g) suggest a directional influence within the model structure, though both β values reached the upper bound at 980 mg/100 g, likely reflecting compensatory fitting under strong growth suppression. All β coefficients should be interpreted as exploratory parameters, not experimentally validated competition measures.

Sodium concentration modulated population dynamics in a dose-dependent manner for both organisms (Fig. 3), with LAB showing a progressive shift from active growth-and-decline to full suppression, and Salmonella sustaining growth across all conditions at progressively reduced rates. These simulations suggest that sodium stress differentially influences microbial persistence and hypothetical interaction dynamics in cooked meat systems. From a food safety perspective, LAB-associated population pressure could potentially contribute as an additional hurdle in reduced-sodium formulations, complementing the direct inhibitory effect of sodium on pathogen growth. However, because the interaction structure was not experimentally parameterized from coculture observations, the simulated interaction patterns should be interpreted as exploratory hypotheses requiring validation through controlled coculture experiments.

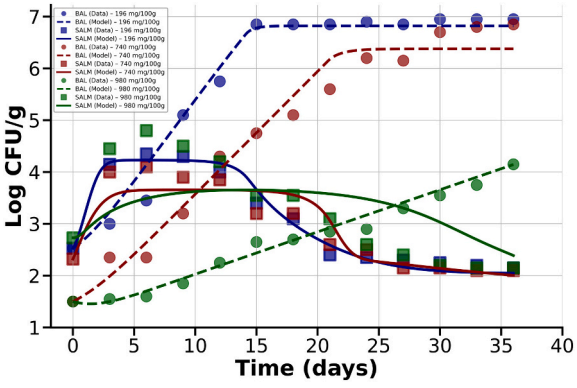

Fig. 3. Simulated population dynamics of lactic acid bacteria (LAB) and *Salmonella* spp. in a cooked meat system stored under refrigerated conditions (0–6 °C) using an integrated Baranyi–Roberts/Lotka–Volterra model. Dashed lines represent LAB model predictions; solid lines represent *Salmonella* model predictions. Symbols indicate experimental monoculture data at sodium concentrations of 196, 740, and 980 mg/100 g.

3.6. Sensitivity analysis

3.6.1. Primary model: Baranyi-Roberts

Global sensitivity analysis of the Baranyi-Roberts model was performed using the Morris method, evaluating the effect of ±10% perturbations on each parameter. Results are summarized in Fig. 4, showing deviations in predicted population *P(t)* and Morris plots of mean elementary effect (μ*) versus standard deviation (σ) for LAB (panels a, b) and *Salmonella* (panels c, d).

For LAB, P_{max}, P_0, and μ_{max} exerted the strongest influence on model predictions and showed evidence of nonlinear interactions with other parameters, consistent with their direct biological roles in defining population ceiling, initial inoculum response, and metabolic rate (Baranyi and Roberts, 1995; González et al., 2022). For *Salmonella*, the most influential parameters were P_{max}, μ_{max}, and m, with P_{max} and μ_{max} also exhibiting interaction effects. In both organisms, V and q_0 displayed negligible sensitivity and no interaction patterns, confirming their role as numerical adjustment parameters without primary biological interpretation (Baranyi and Roberts, 1995; González et al., 2022). These findings are consistent with (Latimer et al., 2002) and (Caballero et al., 2024), who similarly identified growth-related parameters as the dominant sources of variability in microbial predictive models. Practically, these results indicate that accurate estimation of μ_{max} and P_{max} is critical for reliable predictions in reduced-sodium cooked meat systems, and these parameters were therefore prioritized in subsequent secondary modeling and optimization steps.

3.6.2. Secondary model: Modified Norrish

Fig. 5 shows the sensitivity analysis of the modified Norrish equation, parameters A (scale) and B (sodium stability) were the strongest determinants of predicted outputs (μ_{max}, a_w, and pH) for both LAB and *Salmonella*, with both parameters displaying interaction effects. Parameter C exhibited low sensitivity and no interaction patterns, indicating a marginal role that simplifies model calibration without compromising predictive accuracy (Chuang and Toledo, 1976; Maneffa et al., 2017).

Parameter comparisons between organisms revealed higher A and B values in LAB, consistent with greater osmotic stability and adaptive capacity relative to *Salmonella* — a finding that reinforces the differential responses documented in Sections 3.2 and 3.4. High model performance ($R^2 > 0.989$, MSE < 0.0001) confirmed that the modified Norrish equation accurately captured the effect of sodium on both microbial growth and the surrounding physicochemical environment in a species-specific manner. Similar results were reported by (Buelvas Salgado, 2013), who described the combined effects of sodium and temperature on LAB growth in cooked meats and highlighted synergistic interactions between sodium and environmental conditions, reinforcing the utility of this formulation for predicting microbial responses in reformulated food systems (Alfaro-Vives and Más-Diego, 2022; Díaz-Montes, 2023; Koutsoumanis et al., 2004; Wang et al., 2023; Werlang et al., 2021).

3.6.3. Population dynamic model

Sensitivity analysis of the integrated Baranyi–Roberts/Lotka–Volterra model identified $P_{max,LAB}$, β_{LAB}, $P_{max,SALM}$, $\mu_{max,LAB}$, K_{LAB}, and $P_{res,SALM}$ as the parameters with the greatest influence on predicted

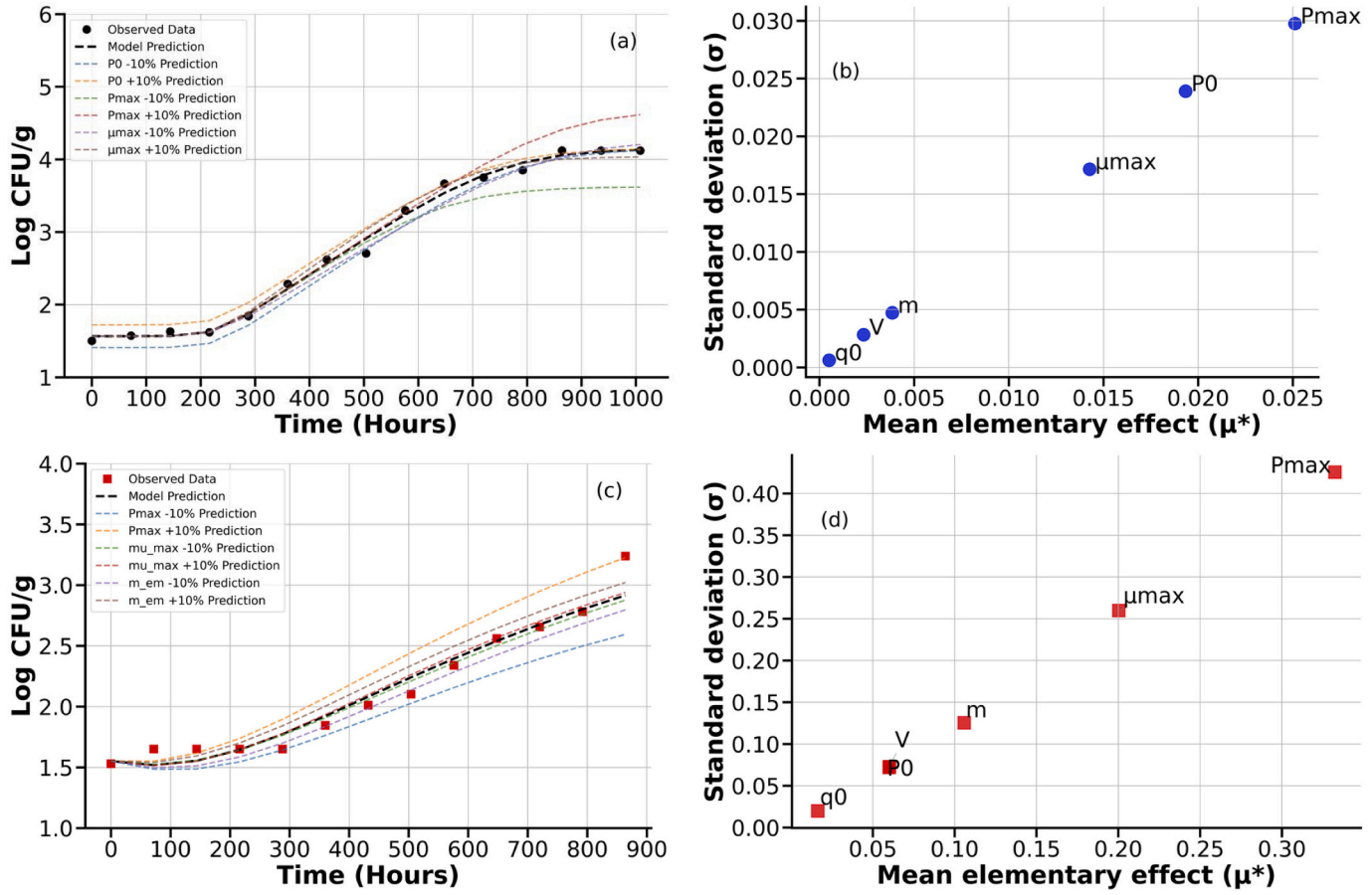

Fig. 4. Global sensitivity analysis of the Baranyi–Roberts primary growth model. Sensitivity analysis of the Baranyi–Roberts model. Relative changes in model output $P(t)$ after ±10% perturbations of optimized parameters, and Morris plots of mean elementary effect (μ^*) versus standard deviation (σ) for LAB (a, b) and *Salmonella* (c, d).

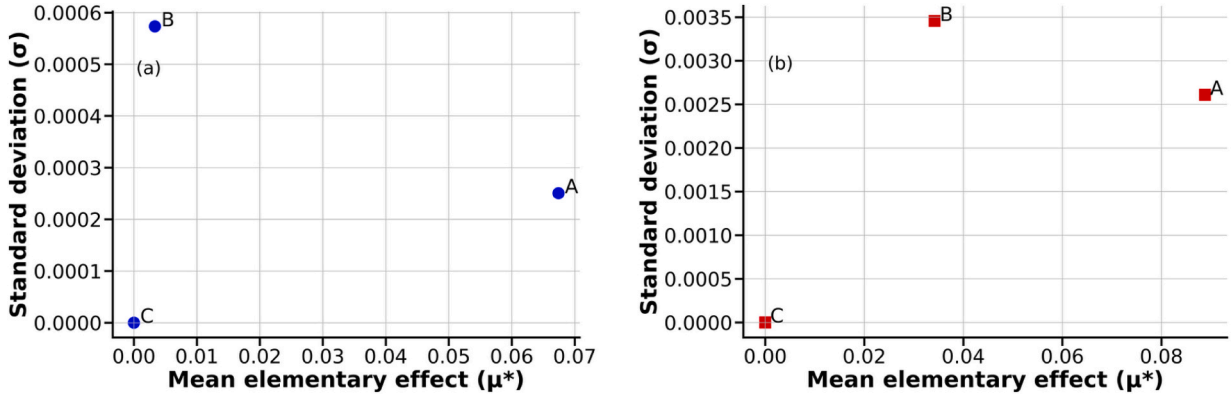

Fig. 5. Global sensitivity analysis of the modified Norrish secondary model. Mean elementary effect (μ^*) versus standard deviation (σ) for LAB (a) and *Salmonella* (b).

bacterial populations over time (Fig. 6). These parameters also demonstrated nonlinear effects and interactions, particularly within the *Salmonella*-associated variables, reflecting the complex feedback between competitive pressure and pathogen persistence. In contrast, $P_{res,LAB}$ showed minimal influence and no interactions, consistent with the near-zero residual populations observed for LAB across all sodium conditions (Table 4).

These results match those of (Caballero et al., 2024), who identified maximum population density and competition-related parameters as key determinants of LAB–*Salmonella* interactions, and collectively confirm that carrying capacity, growth rates, and interaction coefficients are the primary drivers of population dynamics in reduced-sodium cooked meat systems. Together, the sensitivity analyses across all three model levels provide a coherent picture: μ_{max} and population ceiling parameters consistently dominate model behavior, while adjustment parameters such as V, q_0, and C contribute minimally — a finding that streamlines future model recalibration efforts when applying this framework to other sodium concentrations or meat formulations.

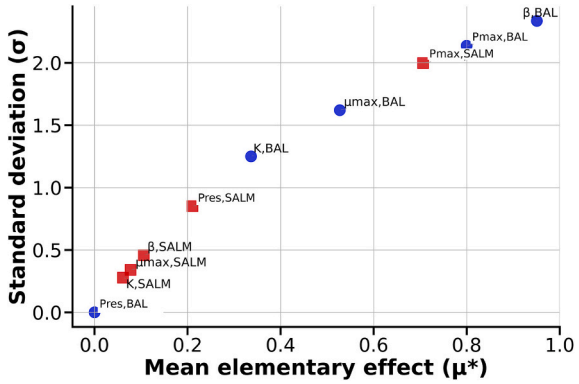

Fig. 6. Sensitivity analysis of the population dynamic model for LAB and *Salmonella*. Mean elementary effect (μ^*) versus standard deviation (σ) of the evaluated parameters. Symbol legend: Blue circle (●) LAB; Red square (■) *Salmonella*.

3.7. Optimization of sodium concentration and validation of the model for shelf-life estimation in cooked meat products

Multi-criteria optimization using the desirability function identified 470.7 mg/100 g as the optimal sodium concentration for simultaneous minimization of LAB and *Salmonella* growth, calculated as the geometric mean of individual desirability functions within the experimental sodium range (Fig. 7). For practical implementation, this value was standardized to 500 mg/100 g — representing a ~ 41% reduction relative to the Colombian regulatory limit for cooked ham (846 mg/100 g, Resolution 2013 of 2020) — a level that maintains microbiological control efficiency while facilitating industrial processing.

Model validation was conducted using two batches of cooked ham formulated at 500 mg/100 g sodium. Predicted μ_{max} values from the modified Norrish model showed high agreement with experimental observations, with percentage differences of 3.39% for LAB and 4.14% for *Salmonella* (Table 5), confirming the transferability of the secondary model from the meat analogue to a real product matrix. These results are comparable to those reported by (Buelvas Salgado, 2013), who validated predictive models for *Leuconostoc mesenteroides* growth in vacuum-packed ham and obtained similar levels of predictive accuracy.

Comparison between experimental data from two batches and the model-simulated curve for LAB and *Salmonella*; Relationship between predicted and observed values for LAB (block a) and *Salmonella* (block b), respectively.

Predicted growth rates were subsequently used as inputs for the Baranyi–Roberts and dynamic population models. For LAB, adjustment

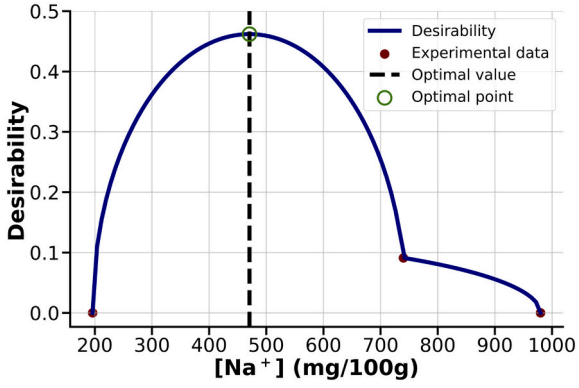

Fig. 7. Desirability-based optimization of sodium concentration in cooked meat products. Global desirability as a function of sodium concentration ([Na^+], mg/100 g). The dashed line indicates the optimal sodium concentration, and black points represent the experimental sodium levels evaluated.

Table 5
Maximum growth rate (μ_{max}) estimation of LAB and *Salmonella* in cooked ham with sodium adjusted to 500 mg/100 g.

Microorganism	μ_{max} observed (h^{-1})	μ_{max} predicted (h^{-1})	% Difference
LAB	0.1149	0.1110	3.39
Salmonella	0.0169	0.0162	4.14

factors of $Af = 1.3813$ and $Bf = 0.9936$ were obtained; for *Salmonella*, $Af = 1.4443$ and $Bf = 1.0212$ (Fig. 8). Bf values close to 1 indicate unbiased predictions, while Af values reflect the average spread of predictions around observations — values below 2.0 are generally considered acceptable for predictive models in food systems (Perez-Rodriguez and Valero, 2013; Rood et al., 2025). These results confirm the model's capacity to reproduce microbial growth dynamics under the sodium conditions tested.

Shelf-life estimation based on a LAB spoilage threshold of 6.5 Log CFU/g — a microbiological criterion frequently associated with sensory deterioration in refrigerated meat products (Casaburi et al., 2015; Nychas et al., 2008) — predicted 437.1 h (18.2 days), compared with the experimentally observed 432 h (18.0 days), a deviation of only 1.19%. This level of accuracy demonstrates that the framework reliably predicts microbial shelf-life trends in sodium-reduced cooked ham under the evaluated experimental conditions without requiring additional empirical calibration for each formulation.

3.8. Study limitations and future directions

The present framework was developed under controlled experimental conditions designed to isolate the effect of sodium concentration on microbial growth dynamics in cooked meat systems. Therefore, some limitations should be considered when interpreting the applicability of the model under industrial conditions.

The microbiological evaluation included a single reference strain of *Salmonella enterica* subsp. *enterica* serovar Typhimurium ATCC 14028™ and one representative LAB isolate (*Leuconostoc* sp.). Although suitable for controlled predictive microbiology studies, additional strains, strain cocktails, and other relevant microorganisms such as *Listeria monocytogenes* or *Pseudomonas* spp. should be incorporated in future studies to better represent industrial microbial diversity.

In addition, the experimental system was based on a laboratory-prepared cooked meat analogue formulated to independently evaluate sodium concentration while minimizing confounding compositional effects. Although this approach enabled robust parameter estimation, the analogue may not fully reproduce the physicochemical complexity of industrial meat products, including the presence of additional preservation hurdles such as lactates, sorbates, nitrites, organic acids, or modified-atmosphere packaging systems.

The framework was also evaluated under relatively stable refrigerated conditions (0–6 °C), whereas real distribution chains frequently involve temperature fluctuations and abuse scenarios. Future studies should therefore incorporate dynamic temperature conditions, stochastic modeling approaches, coculture validation experiments, and sensory evaluation to further expand the industrial applicability of the proposed framework.

4. Conclusions

This study demonstrates that predictive microbiology provides a feasible and quantitatively robust framework for designing reduced-sodium cooked meat products without compromising microbiological safety. The Baranyi–Roberts model outperformed alternative primary models in describing LAB and *Salmonella* growth kinetics, and its integration with the modified Norrish secondary model accurately captured the physicochemical drivers of microbial behavior. Multi-criteria optimization identified 500 mg sodium/100 g as a practical

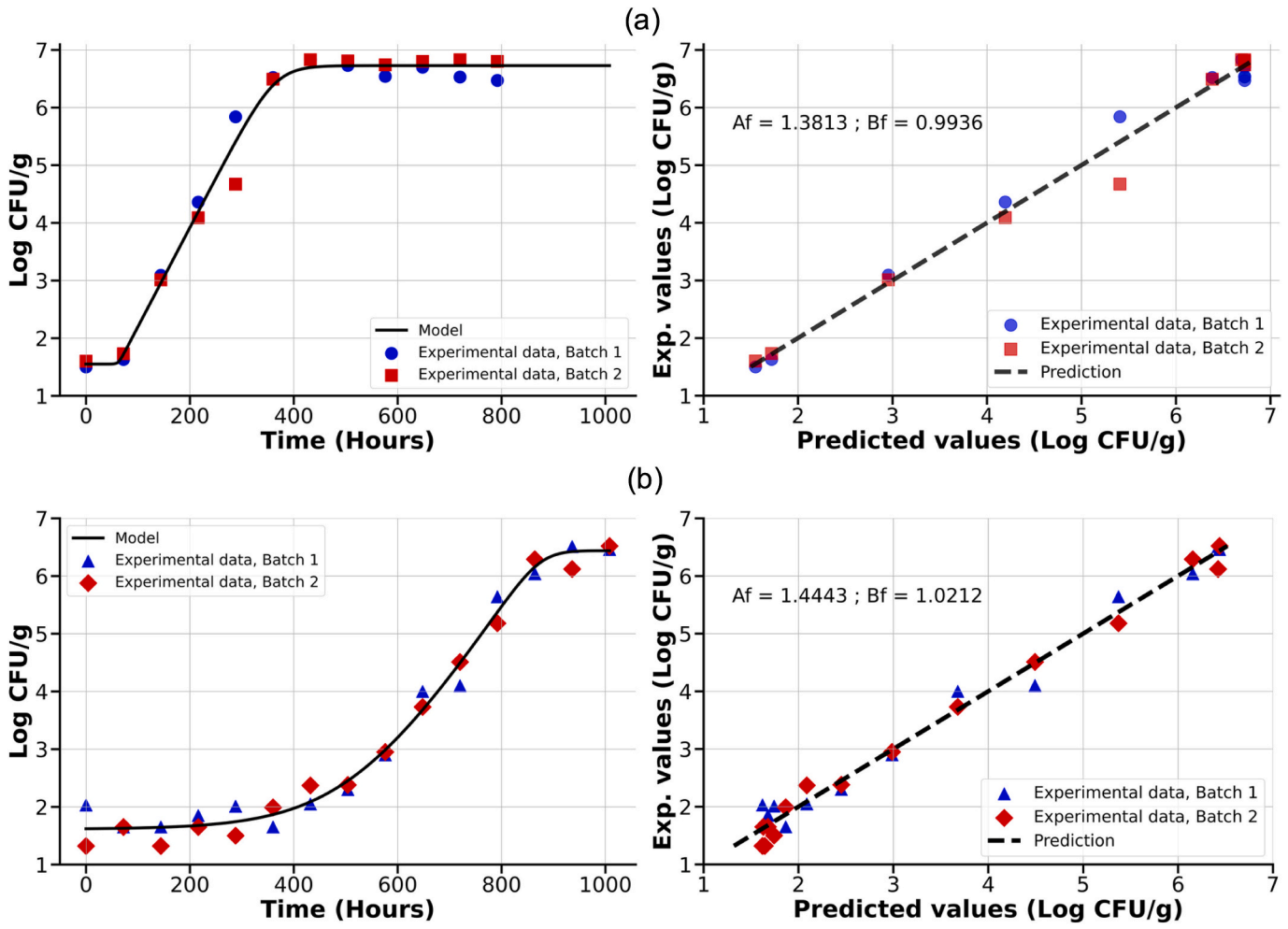

Fig. 8. Validation of model predictions in cooked ham formulated with 500 mg/100 g sodium.

optimum (~41% reduction relative to the Colombian regulatory limit), which was successfully validated in cooked ham with deviations below 5% for growth rate and 2% for shelf-life prediction.

Global sensitivity analysis identified μ_{max} and maximum population density parameters as the dominant sources of model variability. In addition, the dynamic modeling framework suggested sodium-dependent population interaction patterns between LAB and *Salmonella*, with LAB showing greater persistence under reduced-sodium conditions, potentially relevant for future bioprotective strategies in cooked meat systems.

Overall, the proposed framework provides a quantitative and transferable basis for sodium-reduction strategies in processed meat products and may support future developments in predictive microbiology and food safety optimization under industrial conditions.

Declaration of generative AI and AI-assisted technologies in the manuscript preparation process

During the preparation of this work the author(s) used Claude (Anthropic, claude.ai) in order to support programming and script development, code review, and text editing assistance during manuscript preparation. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the published article.

CRediT authorship contribution statement

Juan David Velasquez-Florez: Writing – original draft, Software, Methodology, Investigation, Formal analysis, Conceptualization. **Catalina Quevedo-Ospina:** Writing – original draft, Methodology, Investigation, Formal analysis, Conceptualization. **Leon F. Toro-Navarro:** Writing – review & editing, Writing – original draft, Supervision, Investigation, Funding acquisition, Formal analysis, Conceptualization.

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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.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ijfoodmicro.2026.111902.

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