

Development of a Novel Growth Model Based on the Central Limit Theorem for the Determination of Beef Spoilage

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Abstract

Background and Objective: Currently, no published studies are available that compare central limit theorem model with traditionally used growth models in predictive food microbiology to describe bacterial growth behaviors of *Pseudomonas* spp. in beefs. The major objectives of the present study were to develop a novel growth model based on the central limit theorem and compare the prediction capability of the model with those of various growth models (modified Gompertz, logistic, Baranyi and Huang models) commonly used in predictive food microbiology.

Material and Methods: Bacterial growth data for *Pseudomonas* spp. were collected from previously published studies on beefs stored at isothermal storage temperatures (0, 4, 7, 10, 15 and 20 °C). Temperature dependent kinetic parameters (maximum specific growth rate ' μ_{max} ' and lag phase duration ' λ ') collected from various primary models were described as functions of storage temperatures using Ratkowsky model. Fitting capability of the novel growth model based on the central limit theorem was compared with other growth models using mean square error and coefficient of determination.

Results and Conclusion: The novel growth model developed in this study provided mean square errors less than 0.104 and coefficients of determination greater than 0.962. No significant differences ($p > 0.05$) were seen between the statistical indices of this developed model and traditionally used growth models. Results have shown that the novel growth model based on the central limit theorem can be used to describe the growth behaviors of microorganisms as alternative to traditionally used growth models of modified Gompertz, logistic, Baranyi and Huang models in predictive food microbiology. Furthermore, this novel model can be used for the prediction of shelf-life of beefs as a function of temperature since spoilage of beefs is directly linked to the load of *Pseudomonas* spp.

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1. Introduction

Microbial loads can be assessed using traditional microbiology quantifying methods. However, use of these methods in food products is time-consuming and expensive. Furthermore, results of these methods provide only information about particular time and situation. In fact, the growth performance of microorganisms depends on changing environmental conditions. Therefore, traditional quantifying methods are not effectively practical. Due to these disadvantages, mathematical microbial models have increasingly been used to assess the growth behaviors of microorganisms in foods during processing and storage [1]. Predictive microbiology is the integration of traditional

microbiology knowledge with disciplines of mathematics and statistics to describe microbial behaviors under various environmental conditions [1,2]. This mathematical knowledge enables researches to suggest behaviors of pathogens and spoilage microorganisms in foods; for which, there are no microbial data [2]. Therefore, the major purpose of predictive microbiology is to predict microbial behaviors, which can prevent food spoilage as well as food-borne illnesses using mathematical models.

The two-step modelling approach is an approach; in which, the primary and secondary models commonly used in predictive food microbiology are separately fitted to the growth data and kinetic parameters, respectively. This

approach is the most popular modelling approach in predictive food microbiology [3]. For the primary models, modified Gompertz, logistic, Baranyi and Huang models are the most popular approaches, describing microbial growth data as a function of time at constant environmental conditions. The central limit theorem states that when the number of random variables increases towards infinity, distribution of the sum (or mean) of those variables approximates the normal distribution asymptotically [4]. Alternatively, Robazza et al. [5] have suggested a novel growth model based on the central limit theorem (CLT) to describe the growth behaviors of *Pseudomonas* spp. in fish meats and reported that this model resulted in a better fitting performance than that the other models widely used as primary models in predictive food microbiology did. The secondary models indicate how achieved parameters from the primary models change due to one or more environmental or cultural factors (e.g. gas composition, pH, temperature and salt level). Temperature is one of the most important environmental factors directly affecting the growth behaviors of microorganisms in foods, and its effects are widely simulated using Ratkowsky model [6]. Currently, no published studies are available that compare the CLT model with traditionally used growth models in predictive food microbiology to describe bacterial growth behaviors of *Pseudomonas* spp. in beefs. Meat provides natural high quality proteins, including all essential amino acids, minerals and vitamins. Therefore, meat products are favourable growth media for microorganisms [7-9]. In this study, bacterial growth data of *Pseudomonas* spp. in beefs stored at isothermal storage temperatures (0, 4, 7, 10, 15 and 20 °C) were simulated using two-step modelling approaches with various primary models. A novel growth model based on the CLT was developed and used for the statistical comparison of the CLT with traditional growth models.

2. Materials and Methods

2.1. Data collection

Bacterial growth data of *Pseudomonas* spp. were collected from a published study for beefs stored at isothermal temperatures of 0-20 °C [10]. Briefly, the beef was arbitrarily separated into three sets of three pieces each. Tendons were removed aseptically and the beef was cut perpendicular to the muscle fibers into 25-cm² pieces of nearly 1-2 cm thickness. Beef samples were overwrapped in low-density storage bags and stored aerobically at 0, 4, 7, 10, 15 and 20 °C for 320 h maximally. Samples were plated on CFC agar (Oxoid, UK) and incubated aerobically at 25 °C for 2 days to count the *Pseudomonas* population. Each assessment was repeated at least three times. In this study, 71 growth data sets were extracted from the growth curves using GetData Graph Digitizer Software v.2.26 (www.get-data-graph-digitizer.com); by which, growth data points

could be assessed precisely with one decimal accuracy for the storage temperature of 0-20 °C.

2.2. Modelling

2.2.1. Primary modelling

Four primary models of modified Gompertz, logistic, Baranyi and Huang models were used in two-step and one-step modelling approaches since they were the most popular sigmoid functions that described the bacterial growth behaviors. The modified Gompertz and logistic models at constant environmental conditions were described using Eqs. (1) and (2), respectively [11]:

$$x(t) = x_0 + (x_{max} - x_0) \cdot \exp \left\{ -\exp \left[\frac{r_{max} \cdot e}{(x_{max} - x_0)} \cdot (\lambda - t) + 1 \right] \right\} \quad \text{Eq.1}$$

$$x(t) = x_0 + \frac{(x_{max} - x_0)}{\left\{ 1 + \exp \left[\frac{4 \cdot r_{max}}{(x_{max} - x_0)} \cdot (\lambda - t) + 2 \right] \right\}} \quad \text{Eq.2}$$

Where, t was the time (h), $x(t)$ was the concentration of bacterial populations (log CFU g⁻¹) at time t , x_0 was the initial concentration of bacterial populations (log CFU g⁻¹), x_{max} was the maximum concentration of bacterial populations (log CFU g⁻¹), r_{max} was the maximum bacterial growth rate (log CFU h⁻¹) and λ was the duration of lag phase (h). Baranyi and Huang models were other extensively used primary functions, respectively described by Eqs. (3) and (4) [12,13]:

$$y(t) = y_0 + \mu_{max} F(t) - \ln \left(1 + \frac{e^{\mu_{max} F(t)} - 1}{e^{(y_{max} - y_0)}} \right) \quad \text{Eq.3}$$

$$y(t) = y_0 + y_{max} - \ln(e^{y_0} + [e^{y_{max}} - e^{y_0}] \cdot e^{-\mu_{max} B(t)}) \quad \text{Eq.4}$$

Where, t was the time (h), $y(t)$ was the concentration of bacterial populations (ln CFU g⁻¹) at time t , y_0 was the initial concentration of bacterial populations (ln CFU g⁻¹), y_{max} was the maximum concentration of bacterial populations (ln CFU g⁻¹), μ_{max} was the maximum specific bacterial growth rate (1/h), λ was the duration of lag phase (h), $F(t)$ and $B(t)$ were the adjustment functions, respectively described by Baranyi and Roberts [12] and Huang [13]. Additionally, a novel growth model based on the central limit theorem was suggested by Robazza et al. [5]: Where, t was the time (h), $y(t)$ was the concentration of bacterial populations (ln CFU g⁻¹) at time t , y_0 was the initial concentration of bacterial populations (ln CFU g⁻¹), m was linked to the physiological state of cells, t^* was the time (h) of inflection point, erf was the Gaussian error function, μ_{max} was the maximum specific

bacterial growth rate (1/h) and λ was the duration of lag phase (h).

$$y(t) = y_0 + \mu_{max} \sqrt{\frac{\pi}{2\sqrt{m}}} (\lambda - t^*) \left\{ \operatorname{erf} \left[\sqrt{2} \left(\frac{t^* - t}{t^* - \lambda} \right) \right] - \operatorname{erf} \left[\sqrt{2} \left(\frac{t^*}{t^* - \lambda} \right) \right] \right\} \quad \text{Eq.5}$$

Since the primary models used various scales for the count of microorganism populations, the growth rates (r_{max}) from the Modified Gompertz and logistic models after fitting were converted to the maximum specific growth rates (μ_{max}) multiplying $\ln(10)$.

2.2.2. Secondary modelling

Ratkowsky model [6] was used for the assessment of relationships between the storage temperature and μ_{max} using Eq. (6):

$$\sqrt{\mu_{max}} = b_1(T - T_0) \quad \text{Eq.6}$$

Where, T was the storage temperature ($^{\circ}\text{C}$), T_0 was the theoretical lowest bacterial growth temperature ($^{\circ}\text{C}$), μ_{max} was the maximum specific bacterial growth rate (1/h) and b_1 was the regression coefficient. Moreover, λ was described as a function of μ_{max} based on the temperature using Eq (7) [14]:

$$\lambda = \frac{b_2}{\mu_{max}(T)} \quad \text{Eq.7}$$

Where, b_2 was the regression coefficient and $\mu_{max}(T)$ was the function of temperature; by which, λ was described as a function of storage temperature. For the two-step and one-step modelling approaches, each parameter (x_0/y_0 , x_{max}/y_{max} , b_1 , b_2 and T_0) was calculated using NonLinearModel command, which used Levenberg Marquardt algorithm in Matlab Software v.8.3.0.532 (R2014a) (MathWorks, Natick, MA, USA). Assessment of the appropriate starting values in nonlinear regression procedures is a necessary step to estimate accurate parameters. The starting values for the parameters, x_0/y_0 and x_{max}/y_{max} , were selected as the minimum and the maximum concentrations of bacterial populations, including the complete temperature range, respectively. Randomly choosing starting points for the parameters, b_1 , b_2 and T_0 , might lead the estimated parameters to possible local optimal points around global one, especially for one-step modelling approach. Therefore, starting points of these parameters were selected using ga command, which used genetic algorithm in Global Optimization Toolbox of Matlab Software for two-step and one-step modelling approaches. Following successful iteration process for the nonlinear regression procedure, the global optimum values of the parameters were achieved.

2.2.3. Comparison of the goodness of fit of the global models

Comparison of the global model estimation capabilities was carried out using the mean square error (MSE) and coefficient of determination (R^2) values in Eqs. (8) and (9), respectively:

$$\text{MSE} = \sum_{i=1}^n \frac{(\text{observed}_i - \text{fitted}_i)^2}{n - s} \quad \text{Eq.8}$$

$$R^2 = 1 - \left(\frac{\text{SSE}}{\text{SST}} \right) \quad \text{Eq.9}$$

Where, the observed_i was the experimental bacterial growth, n was the number of experiments, s was the number of parameters of the model, SSE was the sum of squares of errors and SST was the total sum of squares. Since the primary models used various scales for the count of microorganism populations, MSE values of the modified Gompertz and logistic models could not directly be compared with MSE values of the Baranyi and Huang models. Therefore, conversion from $\ln$ to $\log$ scales was carried out to compare the MSE values of all primary models.

2.2.4. Shelf-life prediction

The *Pseudomonas* spp. counts in beefs stored at 0-20 $^{\circ}\text{C}$ could be predicted as a function of time and storage temperature using Eq. (10) by Buchanan et al. [15]:

$$\begin{aligned} x(t) &= x_0, \text{ for } t \leq \lambda \\ x(t) &= x_0 + r_{max}(t - \lambda), \text{ for } \lambda < t < t_{max} \\ x(t) &= x_{max}, \text{ for } t \geq t_{max} \end{aligned} \quad \text{Eq.10}$$

Where, t was the time (h), $x(t)$ was the concentration of bacterial populations ($\log \text{CFU g}^{-1}$) at time t , x_0 was the initial concentration of bacterial populations ($\log \text{CFU g}^{-1}$), t_{max} was the time (h) when the *Pseudomonas* counts reached their maximum rates, x_{max} was the maximum concentration of bacterial populations ($\log \text{CFU g}^{-1}$), r_{max} was the fitted maximum bacterial growth rate ($\log \text{CFU h}^{-1}$) at the storage temperature T ($^{\circ}\text{C}$) and λ was the duration of lag phase (h). Furthermore, Eq. (10) could be used to assess the microbial spoilage and predict the product shelf-life. In this study, shelf-life of beefs was described as a time; at which, the *Pseudomonas* counts in beefs reached a threshold value of $8.2 \log \text{CFU g}^{-1}$ [10].

2.2.5. Statistical analysis

In general, MSE and R^2 values from two-step and one-step modelling approaches were subjected to one-way analysis of variance using Matlab Software v.8.3.0.532 (R2014a) (MathWorks, Natick, MA, USA). Statistical differences between the modelling approaches were assessed using post hoc analysis and Tukey's test. Differences between the means were reported as statistically significant if $p \leq 0.05$.

3. Results and Discussion

The experimental *Pseudomonas* counts collected from previously published curves [10] for beefs at storage temperatures of 0, 4, 7, 10, 15 and 20 °C were modelled using two-step modelling approach (Fig. 1). The initial bacterial counts of *Pseudomonas* spp. averagely included 3.4 ± 0.2 log CFU g⁻¹ for all temperatures.

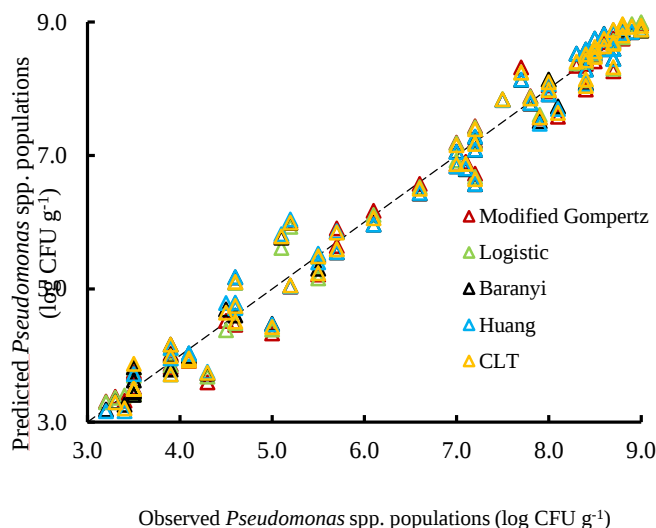


Figure 1. Correlations between the *Pseudomonas* populations on beefs at various isothermal conditions (0, 4, 7, 10, 15 and 20 °C) and the fitted *Pseudomonas* populations with various primary model

The storage time was directly linked to storage temperatures and ranged 320–72 h (~13-3 days) with increases in storage temperatures of 0–20 °C. The *Pseudomonas* counts could reach 8.8 ± 0.5 log CFU g⁻¹ at the end of storage. No

statistical differences ($p > 0.05$) were seen between the final populations of *Pseudomonas* spp. in beefs stored at temperatures of 0–20 °C. The goodness of fit of the five primary models (modified Gompertz, logistic, Baranyi, Huang and central limit theorem-based growth models) were analyzed by comparing the corresponding MSE and R² values for each model (Tables 1-5). At 0 °C, each primary model resulted in the greatest MSE and the minimum R² values to describe the growth of *Pseudomonas* spp. in beefs. However, MSE values less than 0.333 and R² values higher than 0.952 were achieved for each model and all the temperature ranges. Low MSE and high R² values indicated that all primary models could describe the growth behavior of *Pseudomonas* spp. in beefs with no significant differences between their MSE and R² values.

The minimum calculated counts of *Pseudomonas* spp. included averages of 3.4 ± 0.2 , 2.7 ± 0.4 , 3.4 ± 0.1 and 3.4 ± 0.1 for modified Gompertz, logistic, Baranyi and Huang models, respectively (Tables 1-4). The maximum estimated counts of *Pseudomonas* spp. included averages of 9.1 ± 0.3 , 8.9 ± 0.4 , 8.8 ± 0.3 and 8.8 ± 0.3 for modified Gompertz, logistic, Baranyi and Huang models, respectively (Tables 1-4). Although the logistic model detected smaller minimum counts of *Pseudomonas* spp., compared to that the modified Gompertz, Baranyi and Huang models did, no significant differences ($p > 0.05$) were seen between the primary models in minimum calculated counts of *Pseudomonas* spp. in beefs. In the exponential phase; in which, the growth rate reached its maximum values (μ_{max}), changes in organoleptic properties of foods such as color, odor and texture reached their maximum rates as well [16]. These changes were adversely affected due to the high metabolic activity and growth of microorganisms.

Table 1. Kinetic parameters of *Pseudomonas* spp. in beef stored at different isothermal temperatures using modified Gompertz model.

Storage Temperature(°C)	y_0 (log CFU g ⁻¹)	y_{max} (log CFU g ⁻¹)	r_{max} (log CFU h ⁻¹)	λ (h)	MSE	R ²
0	3.3 ± 0.3	8.4 ± 0.6	0.041 ± 0.012	98.7 ± 21.4	0.333	0.952
4	3.4 ± 0.4	9.6 ± 0.3	0.036 ± 0.004	24.9 ± 19.1	0.115	0.982
7	3.7 ± 0.3	9.3 ± 0.3	0.048 ± 0.006	21.8 ± 12.5	0.107	0.982
10	3.4 ± 0.3	9.1 ± 0.2	0.080 ± 0.008	10.2 ± 6.7	0.047	0.991
15	3.3 ± 0.2	9.1 ± 0.1	0.116 ± 0.006	5.0 ± 2.5	0.015	0.998
20	3.1 ± 0.3	8.8 ± 0.1	0.154 ± 0.012	1.5 ± 3.2	0.029	0.995

MSE: mean square error calculated based on log CFU/g, R²: coefficient of determination

Table 2. Kinetic parameters of *Pseudomonas* spp. in beef stored at different isothermal temperatures using logistic model.

Storage Temperature(°C)	y_0 (log CFU g ⁻¹)	y_{max} (log CFU g ⁻¹)	r_{max} (log CFU h ⁻¹)	λ (h)	MSE	R ²
0	3.2 ± 0.3	8.3 ± 0.4	0.044 ± 0.011	106.2 ± 20.2	0.224	0.967
4	2.7 ± 0.8	9.4 ± 0.3	0.034 ± 0.004	3.4 ± 32.8	0.112	0.982
7	3.3 ± 0.5	9.1 ± 0.2	0.048 ± 0.006	15.6 ± 17.5	0.094	0.984
10	2.7 ± 0.6	9.0 ± 0.2	0.076 ± 0.007	0.0 ± 11.6	0.045	0.992
15	2.4 ± 0.4	9.1 ± 0.1	0.108 ± 0.005	0.0 ± 5.1	0.017	0.998
20	2.2 ± 0.6	8.8 ± 0.1	0.147 ± 0.009	0.0 ± 5.1	0.024	0.996

MSE: mean square error calculated based on log CFU g⁻¹, R²: coefficient of determination.

Table 3. Kinetic parameters of *Pseudomonas* spp. in beef stored at different isothermal temperatures using Baranyi model.

Storage Temperature(°C)	$y_0(\log \text{CFU g}^{-1})$	$y_{\max}(\log \text{CFU g}^{-1})$	$r_{\max}(\log \text{CFU h}^{-1})$	$\lambda(\text{h})$	MSE	R ²
0	3.2 ± 0.3	8.1 ± 0.4	0.034 ± 0.007	84.1 ± 19.9	0.226	0.967
4	3.4 ± 0.3	9.2 ± 0.2	0.030 ± 0.003	14.1 ± 17.6	0.129	0.979
7	3.6 ± 0.3	8.9 ± 0.2	0.041 ± 0.004	1.8 ± 11.9	0.109	0.981
10	3.5 ± 0.2	8.8 ± 0.1	0.066 ± 0.005	5.0 ± 5.8	0.048	0.991
15	3.5 ± 0.2	9.0 ± 0.1	0.092 ± 0.005	2.7 ± 2.6	0.024	0.997
20	3.3 ± 0.1	8.6 ± 0.1	0.127 ± 0.008	0.4 ± 1.9	0.022	0.996

MSE: mean square error calculated based on log CFU g⁻¹, R²: coefficient of determination.

Table 4. Kinetic parameters of *Pseudomonas* spp. in beef stored at different isothermal temperatures using Huang model.

Storage Temperature(°C)	$y_0(\log \text{CFU g}^{-1})$	$y_{\max}(\log \text{CFU g}^{-1})$	$r_{\max}(\log \text{CFU h}^{-1})$	$\lambda(\text{h})$	MSE	R ²
0	3.2 ± 0.3	8.1 ± 0.4	0.032 ± 0.006	77.8 ± 17.7	0.253	0.963
4	3.5 ± 0.4	9.2 ± 0.2	0.030 ± 0.002	16.5 ± 14.5	0.123	0.980
7	3.5 ± 0.3	8.9 ± 0.2	0.038 ± 0.003	4.3 ± 10.6	0.115	0.980
10	3.5 ± 0.2	8.8 ± 0.1	0.065 ± 0.004	5.0 ± 4.6	0.046	0.991
15	3.5 ± 0.1	9.0 ± 0.1	0.092 ± 0.004	3.3 ± 2.1	0.020	0.997
20	3.3 ± 0.2	8.6 ± 0.1	0.126 ± 0.007	0.1 ± 3.0	0.023	0.996

MSE: mean square error calculated based on log CFU/g, R²: coefficient of determination.

Table 5. Kinetic parameters of *Pseudomonas* spp. in beef stored at different isothermal temperatures using CLT model.

Storage Temperature(°C)	$r_{\max}(\log \text{CFU h}^{-1})$	$\lambda(\text{h})$	$t^*(\text{h})$	MSE	R ²
0	0.037 ± 0.007	127.221 ± 9.833	154.012 ± 10.273	0.222	0.962
4	0.033 ± 0.003	70.342 ± 12.172	104.859 ± 8.029	0.104	0.981
7	0.045 ± 0.005	45.783 ± 9.478	70.501 ± 6.267	0.092	0.983
10	0.073 ± 0.006	26.451 ± 5.454	41.676 ± 3.609	0.040	0.992
15	0.103 ± 0.005	16.820 ± 2.430	28.167 ± 1.658	0.015	0.998
20	0.143 ± 0.007	25.015 ± 1.227	16.549 ± 1.895	0.020	0.996

MSE: mean square error calculated based on log CFU g⁻¹, R²: coefficient of determination.

In lag phase, organoleptic properties did not change because the number of microorganisms was relatively constant. Therefore, lower $\mu_{\max}$ and higher λ were desired to increase shelf-lives of foods [17]. Temperature is one of the most important factors affecting $\mu_{\max}$ and λ [18]. The $\mu_{\max}$ and λ derived from the modified Gompertz, logistic, Baranyi, Huang and central limit theorem-based growth models were correlated with the temperature using Ratkowsky model for $\mu_{\max}$ and λ . After carrying out the secondary model fitting, calculated $\mu_{\max}$ and λ values were recorded in Figs. 2a and 2b, respectively. The $\mu_{\max}$ increased from 0.061 to 0.354 h⁻¹, 0.060 to 0.349 h⁻¹, 0.051 to 0.289 h⁻¹, 0.048 to 0.289 h⁻¹ and 0.055 to 0.326 h⁻¹ with increases in storage temperatures for modified Gompertz, logistic, Baranyi, Huang and central limit theorem-based growth models, respectively (Fig. 2a). An exact opposite tendency was observed using Eq. 7 for λ , respectively decreasing from 68 to 12 h, 52 to 9 h, 51 to 9 h, 48 to 8 h and 54 to 9 h for modified Gompertz, logistic, Baranyi and Huang models, as temperature increased from 0 to 20 °C (Fig. 2b). These results revealed that $\mu_{\max}$ and λ values were inversely correlated to each other and beefs should be stored at low temperatures to decrease their microbial contamination. The MSE values of Ratkowsky model for $\mu_{\max}$ were less than

6.63×10^{-4} , while the R² values were greater than 0.954 (Table 6).

The high R² and low MSE values indicated that Ratkowsky model could reliably be used to assess effects of temperature on the maximum specific growth rate and lag phase from each of the primary models for *Pseudomonas* spp. in beefs.

Table 6. Parameters of Ratkowsky model for maximum specific growth rate.

Primary model	b_1	T_0	MSE	R ²
Modified Gompertz	0.0175 ± 0.001	-14.1 ± 2.8	5.51 × 10 ⁻⁴	0.963
Logistic	0.0173 ± 0.002	-14.2 ± 3.2	6.63 × 10 ⁻⁴	0.954
Baranyi	0.0156 ± 0.002	-14.5 ± 2.8	3.40 × 10 ⁻⁴	0.965
Huang	0.0160 ± 0.002	-13.8 ± 2.7	3.41 × 10 ⁻⁴	0.966
CLT	0.0168 ± 0.002	-14.0 ± 2.6	3.81 × 10 ⁻⁴	0.970

MSE: mean square error calculated based on maximum specific growth rate (1/h).

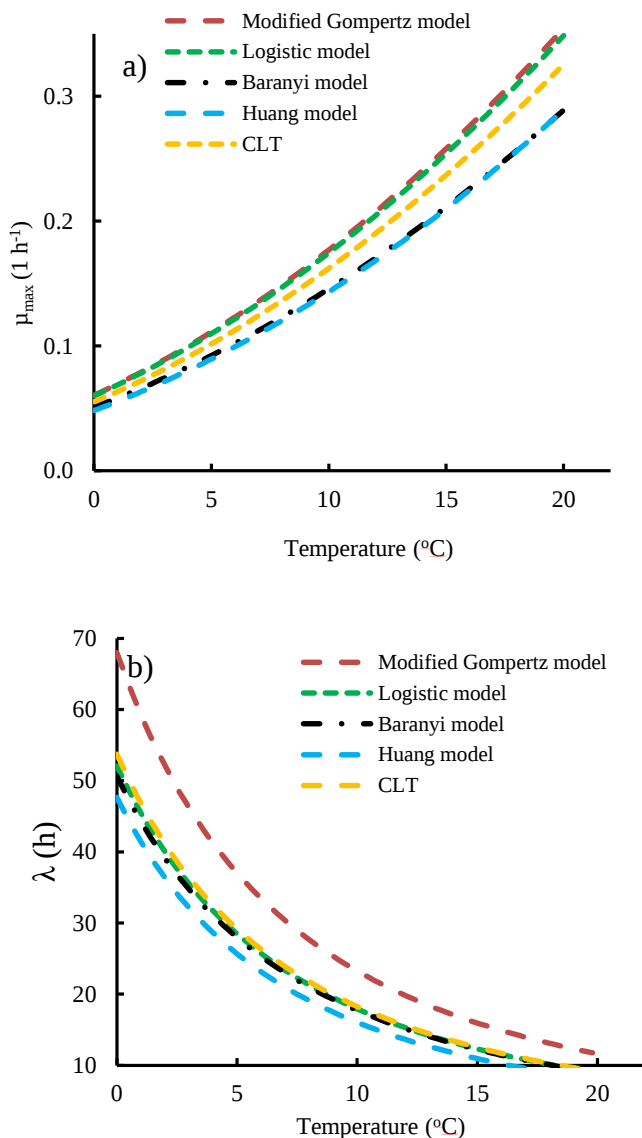


Figure 2. The maximum specific growth rate (a) and lag phase duration (b) described as a function of time

As the beef spoilage is directly linked to *Pseudomonas* concentration, each of the models was used to predict the shelf-life of beefs as a function of storage temperature. Thus, $8.2 \log \text{CFU g}^{-1}$ *Pseudomonas* were considered as the limiting microbial criteria for the beefs, assuming that the initial count of *Pseudomonas* spp. was $3.2 \log \text{CFU g}^{-1}$ [10]. Effects of temperature on microbial spoilage were linked to the shelf-lives of the beefs (Fig. 3). Temperature was identified as a critical factor, affecting shelf-lives of the beefs. As the temperature increased, shelf-lives of the beefs decreased considerably. For example, shelf-lives were calculated as 251, 236, 267, 277 and 254 h at 0 °C for modified Gompertz, logistic, Branyi, Huang and central limit theorem-based growth models, respectively but it was only 43, 41, 47, 46 and 43 h at 20 °C for the modified Gompertz, logistic, Branyi, Huang and central limit theorem-based growth models, respectively.

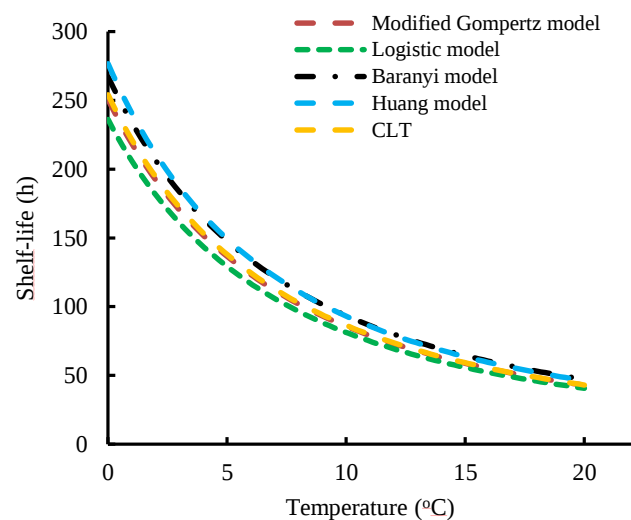


Figure 3. Effects of storage temperatures on the shelf-life of beefs. Shelf-life was predicted using $x_0 = 3.2 \log \text{CFU/g}$ and $x(t) = 8.2 \log \text{CFU g}^{-1}$

The shelf-life was almost six times lower at 20 °C than 0 °C. Therefore, beefs should not be stored at higher temperatures and always must be stored at chilled conditions, preferably less than 0 °C. Statistical indices showed no significant differences between the shelf-lives predicted by various growth models, meaning that the developed growth model based on the central limit theorem could reliably be used to predict the microbial shelf-life of food products.

4. Conclusion

In this study, several growth models such as modified Gompertz, logistic and Baranyi and Huang models commonly used as primary models in predictive food microbiology were used to describe growth behaviors of *Pseudomonas* spp. in beefs at various isothermal storage temperatures (0, 4, 7, 10, 15 and 20 °C). A novel growth model based on the central limit theorem was used as an alternative to these traditional models and the model prediction capability was compared to that of other models using their MSE and R^2 values. Statistical indices revealed that no significant differences between the goodness of fit results of the traditional models and the suggested novel growth model. Each of the global models was used to assess beef spoilage with no significant differences between their predictions. These results have shown that the novel growth model can be used to describe the growth behaviors of microorganisms as alternative to traditionally used growth models, including modified Gompertz, logistic, Baranyi and Huang models in predictive food microbiology.

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6. Conflict of Interest

The authors report no conflicts of interest.

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توسعه یک مدل نوین رشد براساس تئوری حد مرکزی برای تعیین فساد گوشت گاو فاتح تارلک

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چکیده

سابقه و هدف: اخیراً، مطالعه‌ای در مورد مقایسه مدل تئوری حد مرکزی^۱ با مدل‌های سنتی رشد برای پیش‌بینی میکروبی‌شناسی مواد غذایی به منظور توصیف رفتار رشد باکتریایی گونه سودوموناس در گوشت گاو در دسترس نبوده است. اهداف اصلی مطالعه حاضر توسعه مدل نوین رشد بر اساس تئوری حد مرکزی و مقایسه توانایی پیش‌بینی مدل با مدل‌های گوناگون رشد (گومتز اصلاح شده، مدل‌های لاجیستیک، بارانای و هانگ)، که معمولاً برای پیش‌بینی میکروبی‌شناسی مواد غذایی مورد استفاده قرار می‌گیرند، می‌باشد.

مواد و روش‌ها: اطلاعات رشد باکتریایی گونه سودوموناس از مطالعات منتشر شده قبلی بر روی گوشت گاو نگهداری شده در درجه حرارت‌های نگهداری همدم^۲ (۰، ۴، ۷، ۱۰، ۱۵ و ۲۰ درجه سلسیوس) گردآوری شد. پارامترهای سینتیک وابسته به درجه حرارت (بیشینه نرخ رشد ویژه ' μ_{max} ' و طول مدت فاز تاخیری ' λ ') جمع‌آوری شده از مدل‌های اولیه گوناگون به‌عنوان توابع درجه حرارت‌های نگهداری با استفاده از مدل راتکوفسکی توصیف شدند. قابلیت برازش مدل نوین رشد بر اساس درجه تئوری حد مرکزی با سایر مدل‌های رشد با استفاده از میانگین توان دوم خطاها^۳ و ضریب تعیین مقایسه شد.

یافته‌ها و نتیجه‌گیری: توسعه مدل نوین رشد این مطالعه موجب شد که میانگین‌های توان دوم خطاها کمتر از ۰/۱۰۴ و ضرایب تعیین بیشتر از ۰/۹۶۲ باشد. تفاوت معنی‌داری ($p > 0/05$) بین شاخص‌های آماری مدل توسعه‌یافته و سنتی مورد استفاده در مدل‌های رشد مشاهده نشد. نتایج نشان داده است که مدل نوین رشد بر اساس تئوری حد مرکزی می‌تواند برای توصیف رفتارهای رشد میکروارگانیسم‌ها به‌عنوان جایگزین مدل‌های سنتی رشد گومتز اصلاح شده، مدل‌های لاجیستیک، بارانای و هانگ برای پیش‌بینی میکروبی‌شناسی مورد استفاده قرار گیرد. علاوه‌براین، از آنجا که فساد گوشت گاو به‌طور مستقیم به میزان گونه سودوموناس ارتباط دارد، این مدل نوین می‌تواند برای پیش‌بینی عمر انباری گوشت گاو به‌عنوان تابعی از درجه حرارت مورد استفاده قرار گیرد.

تعارض منافع: نویسندگان اعلام می‌کنند که هیچ نوع تعارض منافع مرتبط با انتشار این مقاله ندارند.

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- تئوری حد مرکزی (CLT)
- سینتیک رشد
- کیفیت میکروبی
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^۱ central limit theorem or CLT

^۲ isothermal

or mean square error ^۳ mean squared error