Hindawi
Journal of Food Quality
Volume 2022, Article ID 9135887, 12 pages
https://doi.org/10.1155/2022/9135887

Research Article

Comparative Study of ε-Polylysine or Nisin Inhibition Kinetics of *Lactococcus lactis* and Spoilage Microorganisms in Fresh *Flammulina velutipes* Fruiting Bodies

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Received 21 November 2021; Revised 23 January 2022; Accepted 10 February 2022; Published 1 March 2022

Academic Editor: Efstathios Giaouris

Flammulina velutipes is one of the most important edible mushrooms, which quickly decays with a short shelf life. However, little is known about the effect of ε-polylysine (ε-PL) or nisin on the survival of *Lactococcus lactis* (*L. lactis*) during the storage at constant temperatures. The objective of this study was to investigate the effect of ε-PL or nisin on the growth of *L. lactis* and background (BK) microorganisms in fresh *Flammulina velutipes* fruiting bodies (FVFB) and develop mathematical models to predict their growth behavior. The effect of ε-PL (0.15 and 0.30 g/kg) or nisin (0.10 and 0.20 g/kg) on the growth of *L. lactis* and BK microorganisms in FVFB was analyzed at 4, 16, and 20°C. The lag phase of *L. lactis* was extended, and the specific growth rate was decreased by increasing concentrations of ε-PL or nisin and lowering the temperature. The results showed that ε-PL or nisin could control the growth of *L. lactis* in FVFB. However, the growth of BK microorganisms was not affected by ε-PL or nisin. The growth of *L. lactis* and BK microorganisms could be successfully described by the reparameterized Gompertz and no lag phase models, respectively. Additionally, ε-PL or nisin could maintain the quality of FVFB by preventing weight loss, color-changing, and decreasing soluble solid content in FVFB at 4°C. These results suggest that ε-PL or nisin in combination with low temperature may inhibit the growth of *L. lactis* in FVFB and prevent the decrease in the quality of FVFB.

1. Introduction

Flammulina velutipes fruiting body (FVFB), known as golden needle mushroom or winter mushroom, is one of the most important edible mushrooms widely cultivated and consumed in China, Japan, and Korea [1]. Compared to other commercial edible mushrooms, such as *Agaricus bisporus*, *Pleurotus eryngii*, *Pleurotus geesteranus*, and *Volvariella volvacea*, FVFB requires a simple cultivation technique and has unique advantages of fast fruiting and high yield [2, 3]. FVFB has been one of the major mushroom species employed in factory cultivation in many countries. Additionally, FVFB contains a variety of bioactive compounds, such as polysaccharides, essential amino acids, glycoproteins, and sesquiterpenoids. Several studies demonstrated that FVFB had potential bioactivity of hepatoprotection, antitumor, antihyperlipidemia, and immune regulation [4–6]. Because of the aforementioned high nutritional value, medicinal properties, and favorable taste, an increasing number of FVFB have been widely consumed. However, as fresh FVFB is nutritious and contains high moisture content, it favors microorganisms (covering pathogenic and spoilage bacteria) propagating, leading to its decay and short shelf life (1–3

days) under ambient conditions [7]. Niu et al. [8] indicated that *Pseudomonas* spp. served as the main native bacteria presented in FVFB.

Lactococcus lactis (*L. lactis*) has generally been recognized as a safe status by the United States Food and Drug Administration [9, 10]. A previous study showed that *L. lactis* was the dominant microflora in FVFB [11]. Zhao et al. [12] reported that the water-soaked and sunken lesions on the mushroom stipes were related to *Lactococcus lactis* subsp. Therefore, *L. lactis* might cause the decay of the mushroom. It is worth noting that some species of *Lactococcus* could be opportunistic pathogens [13, 14], posing potential risks for human health. Therefore, it is vital to prevent the growth of *L. lactis*.

Hurdle technology is a valuable method to ensure food safety by eliminating or controlling the growth of pathogens and extend the shelf life of food products [15]. These hurdles may include temperature, water activity, pH, redox potential, and preservatives. ε-Polylysine (ε-PL) was approved as a natural antimicrobial food additive by FDA and applied in many commercial food products [16, 17]. ε-PL exhibited broad inhibitory activity against microorganisms. ε-PL can be decomposed into lysine in the human body without any side effects and serves as a kind of lysine source. Fukutome et al. [18] reported that there was no obvious pathological tissue change and no possible carcinogenicity was found in rats by dietary administration with 20 mg/kg of ε-PL. Hiraki et al. [19] reported that ε-PL was practically nontoxic in an acute oral toxicity study in rats, with no mortality up to 5 g/kg, and was not mutagenic in bacterial reversion assays. Nisin is a natural food preservative. It can be digested into amino acids by proteolytic enzymes in the digestive system after consumption. Ucar et al. [20] demonstrated that nisin could inhibit the food-borne pathogen growth, thereby reducing biogenic amine production in sea bass fillets. In addition, with the treatment of nisin, the shelf life of sea bass fillets could also be extended at 4°C [21]. These results suggested that nisin could be applied to avoid pathogen contamination, preserve the organoleptic quality, and extend the shelf life of sea bass fillets [22]. However, no studies have reported the impacts of ε-PL and nisin on the growth of *L. lactis* and native microorganisms in FVFB. Besides, temperature plays a crucial role in microbial growth. However, limited studies systematically investigated the combined effect of temperature and ε-PL or nisin on the survival of *L. lactis* in FVFB.

Therefore, this study aims to (1) investigate the influences of ε-PL or nisin on the growth of *L. lactis* and native background microorganisms in FVFB at various isothermal temperatures and (2) develop mathematical models to describe and predict the growth behavior of *L. lactis* and background microorganisms. Furthermore, the quality characteristics (including weight loss, color, and soluble solid content) of FVFB were evaluated with the presence of ε-PL or nisin during 12 days of storage at 4°C. The results obtained in this study will be helpful for the food industry to extend the shelf life of FVFB.

2. Materials and Methods

2.1. Bacterial Cultures and Preparation. The culture of *L. lactis* was isolated from FVFB and identified by 16S RNA gene amplification. The isolated *L. lactis* was transferred to 10 mL of Luria–Bertani broth (Huankai Microbial Co., Ltd., Guangzhou, China) and incubated at 37°C for 20 h. After incubation, the bacteria pellet was harvested by a refrigerated centrifuge (4°C; Sigma-Aldrich Co., MO) at 4500 rpm for 15 min and washed three times using 10 mL of 0.1% sterile peptone water (PW; Huankai Microbial Co., Ltd., Guangzhou, China). Subsequently, the bacterial culture pellet was resuspended in 10 mL of 0.1% sterile PW. Serial dilutions of *L. lactis* were conducted to achieve approximately 10^{5-6} CFU/mL of the bacterial suspension as a working culture.

2.2. Sample Preparation and Bacterial Growth. FVFB was purchased from a local grocery store in Fuzhou, China, and sterilized by irradiating at a dose of 5 kGy (Rice Research Institute, Fujian Academy of Agricultural Sciences, Fuzhou, China). Both ε-PL and nisin were purchased from Zhejiang Silver Elephant Bio-Engineering Co., Ltd. (Zhejiang, China). The preservative solutions of ε-PL (0.15 and 0.30 g/kg) and nisin (0.10 and 0.20 g/kg) were prepared using sterile phosphate-buffered saline (PBS; Huankai Microbial Co., Ltd., Guangzhou, China) and thoroughly mixed by using a stomacher (BagMixer 400W, Interscience Co., Ltd., France) at the maximum speed (10 strokes/s) for 1 min before use. 25 g of FVFB were, respectively, submerged in 200 mL of 0.15 g/kg ε-PL, 0.30 g/kg ε-PL, 0.10 g/kg nisin, or 0.20 g/kg nisin for 2 min. PBS was used as a control. After the treatment, FVFB was immediately removed and dried in a biohood for 1 h. Subsequently, the FVFB samples were inoculated with 0.1 mL of *L. lactis* working culture, individually transferred into a sterile plastic filter bag (20 × 22 cm, Bkmam Co., Ltd., Changde, China) and then stored at different isothermal temperatures (4, 16, and 20°C). The samples were periodically retrieved from the incubators to enumerate the concentration of *L. lactis*. Each sample was homogenized with 225 mL of 0.1% sterile PW in the stomacher at 10 strokes/s for 20 s. 0.1/1.0 mL of homogenized samples with or without serial dilution were plated onto de Man–Rogosa–Sharpe agar (MRS; Huankai Microbial Co., Ltd., Guangzhou, China) plates. After incubation for 24 h at 37°C, the bacterial colonies on MRS plates were counted and converted to the logarithm of base 10 or natural base, recorded as Log_{10} CFU/g or Ln CFU/g.

To study the effect of ε-PL or nisin on native background (BK) microorganisms in FVFB, fresh FVFB samples that were not sterilized and without inoculation of *L. lactis* were taken. The FVFB samples were treated by ε-PL (0.30 g/kg) or nisin (0.20 g/kg), incubated under the temperature conditions, and removed from incubators using the aforementioned procedures and parameters. The concentrations of BK microorganisms in FVFB samples were counted on tryptic soy agar (Huankai Microbial Co., Ltd., Guangzhou, China) plates after being incubated at 37°C for 24 h. The

bacterial concentrations were also converted to the logarithm of base 10 or natural base, recorded as Log_{10} CFU/g or Ln CFU/g.

2.3. Reparameterized Gompertz Model. According to our previous study [13], the reparameterized Gompertz model was more suitable to describe the growth of *L. lactis* in FVFB than the Huang model and Baranyi model. Therefore, in this study, the reparameterized Gompertz model (equation (1)) was developed for predicting the growth of *L. lactis* in FVFB [23]. The no lag phase model (equation (2)) was employed to describe the growth of BK microorganisms [24].

The reparameterized Gompertz model is expressed as

$$Y(t) = Y_0 + (Y_{\max} - Y_0)\exp\left\{-\exp\left[\frac{\mu_{\max} e}{Y_{\max} - Y_0}(\lambda - t) + 1\right]\right\}, \quad (1)$$

where λ represents the lag phase duration (h), $Y(t)$ is the natural logarithm of bacterial count (Ln CFU/g), Y_0 is the natural logarithm of initial bacterial count (Ln CFU/g), $Y_{\max}$ is the natural logarithm of stationary phase bacterial count (Ln CFU/g), and $\mu_{\max}$ is the maximum specific growth rate (h^{-1}).

The no lag phase model is expressed as

$$Y(t) = Y_0 + Y_{\max} - \ln\left[e^{Y_0} + \left(e^{Y_{\max}} - e^{Y_0}\right)e^{-\mu_{\max} t}\right], \quad (2)$$

where Y_0, $Y_{\max}$, $Y(t)$, and $\mu_{\max}$ are identical to those used in the reparameterized Gompertz model.

2.4. Modeling and Statistical Analysis. The Integrated Pathogen Modeling Program (IPMP) developed by United States Department of Agriculture (USDA) was used to analyze the growth curves of *L. lactis* and BK in FVFB at different temperatures [23]. The lag phase duration, specific growth rates, maximum bacterial concentration, confidence intervals, and data analysis were obtained from IPMP analysis.

2.5. Quality Characteristics Analysis. FVFB with similar size, color, and without mechanical damage was selected for the experiment. FVFB was submerged in 200 mL of 0.30 g/kg ε-PL or 0.20 g/kg nisin solution for 2 min. PBS treatment was set as control (CK). The treated FVFB was dried in a biohood for 1 h, dispensed in polystyrene trays, and then stored at 4°C. During the storage, FVFB quality characteristics covering color, weight loss, and soluble solid content were monitored. The color of FVFB was measured by using a colorimeter (Chen Taike Instrument Technology Inc., Beijing, China), and the L^* (light/dark) value of each FVFB was recorded for 12 days. Weight loss analysis was conducted as described by Gao et al. [25]. The soluble solid content was determined according to the method reported by Nasiri et al. [26]. In brief, FVFB juice was yielded by grounding using a pestle. Then, the soluble solid content of the juice was analyzed using a refractometer (TH-S9, Tuohe Co., Ltd., Shanghai, China). Each experiment was repeated three times.

3. Results and Discussion

3.1. Growth of L. lactis and Background Microorganisms in FVFB. Though *L. lactis* was generally recognized as a safe status and widely used in the food industry, it has been reported that *L. lactis* could be associated with the postharvest decay of mushrooms [27]. Moreover, *L. lactis* is a homofermentative bacteria, meaning it can produce lactic acid. The lactic acid could lower the pH value and create suitable conditions for vegetable decay [28]. With the population of *L. lactis* increasing, lactic acid gradually accumulates, causing browning, softening, and deterioration. Furthermore, FVFB typically contains native bacteria, contributing to FVFB's decay and short shelf life. Therefore, ε-PL or nisin was used to control the growth of *L. lactis* and BK microorganisms in FVFB at different storage temperatures (4, 16, and 20°C). The growth data of *L. lactis* and BK microorganisms in FVFB treated with ε-PL or nisin at different isothermal storage temperatures were collected and fitted to the reparameterized Gompertz model and no lag phase model, respectively.

Figure 1 shows the growth curves of *L. lactis* in FVFB samples treated with ε-PL at 0, 0.15, and 0.30 g/kg (Figure 1(a)) and nisin at 0, 0.1, ad 0.2 g/kg (Figure 1(b)) at 4, 16, and 20°C. Throughout storage at 4, 16, and 20°C, the concentration of *L. lactis* increased for all treatments exhibiting three phases of growth: lag phase, exponential phase, and stationary phase. The duration of the lag phase and specific growth rate were affected by the incubation temperature, with the lag phase decreasing and the growth rate increasing with the temperature rising. As shown in Figure 1, nisin or ε-PL appeared to interfere with the growth of *L. lactis*. This result was consistent with the findings reported by Bortolotto et al. [29] and Georgnaras et al. [30]. It was also observed that lower temperature and higher concentrations enhanced inhibition efficacy of nisin or ε-PL on the growth of *L. lactis* synergistically. The reason might be that bacterial cells are more sensitive to nisin or ε-PL at a lower temperature [31]. Additionally, increasing nisin concentrations could promote an increase of cytoplasmic compounds (such as potassium and phosphate ion) leakage, leading to the rise of killed cells [32, 33].

Figure 2 displays the growth curves of BK microorganisms in FVFB treated by different concentrations of ε-PL and nisin at temperatures of 4, 16, and 20°C. It indicates that the BK microorganisms could grow well without the lag phase. As the BK microorganisms naturally exist, they have already adapted to the FVFB. Like *L. lactis*, decreasing temperature could reduce the growth of BK microorganisms. However, the growth behaviors of BK microorganisms in FVFB treated by nisin are similar to those without nisin treatment, suggesting that ε-PL or nisin has no effect on BK microorganisms. The reason might be that the BK microorganisms in FVFB are formed by Gram-negative bacteria (such as *Pseudomonas aeruginosa*) [8]. Nisin has no or little

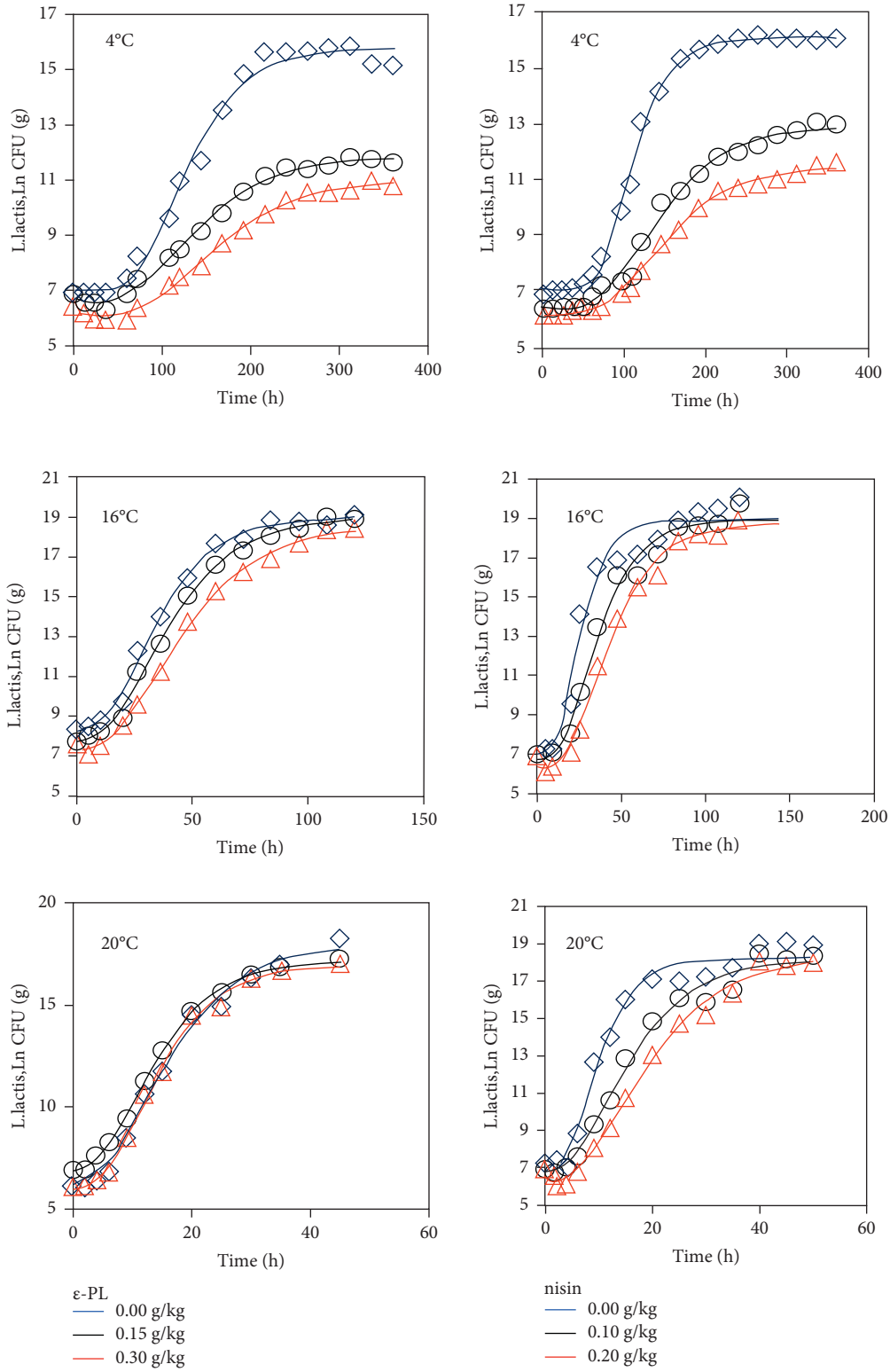

Figure 1: Growth of *L. lactis* in FVFB and curve-fitting at 4, 16, and 20°C: (a) ε-PL treated and (b) nisin treated. Solid line: reparameterized Gompertz model; symbol: observed growth data.

inhibitory effect on Gram-negative bacteria [34, 35]. It has been widely reported that ε-PL has an effective lethal effect on Gram-positive and Gram-negative bacteria [36]. However, in our study, ε-PL exhibited no impact on BK microorganisms, which is inconsistent with those previously reported by other researchers [16, 37]. BK microorganisms might contain some species of Gram-negative bacteria which could resist ε-PL.

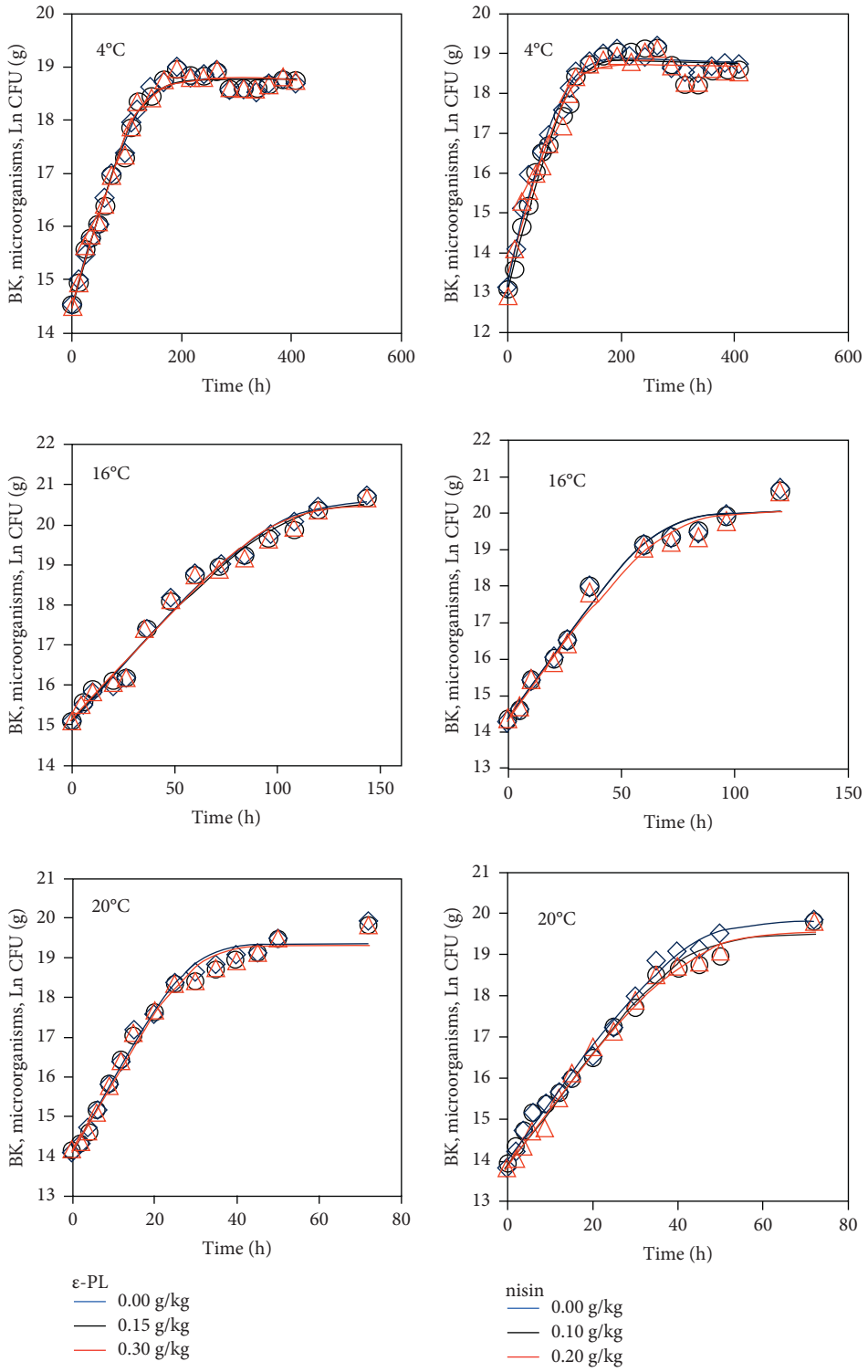

Figure 2: Growth of BK microorganisms in FVFB and curve-fitting at 4, 16, and 20°C: (a) ε-PL treated and (b) nisin treated. Solid line: no lag phase model; symbol: observed growth data.

3.2. Mathematical Modeling. As depicted in Figure 1, the reparameterized Gompertz model shows good fits to the observed growth curves of *L. lactis* in FVFB with or without nisin or ε-PL treatment. The growth kinetic parameters of *L. lactis* estimated by the reparameterized Gompertz model are listed in Table 1. At 4, 16, and 20°C, the lag phases of *L. lactis* were extended from 69.240, 11.563, and 3.474 h, in the absence of ε-PL to 76.904, 13.309, and 4.703 h, when 0.30 g/kg of ε-PL were used. Similarly, as the concentration of nisin varied from 0 to 0.20 g/kg, the bacterial lag phases were increased from 67.693, 11.159, and 3.549 to 76.816, 16.732, and 5.419 h. By inspection of Table 1 and Figure 1, it

Table 1: Growth parameters of *L. lactis* estimated by the reparameterized Gompertz model.

Temperature	4°C			16°C			20°C				4°C			16°C			20°C		
ε-Polylysine (g/kg)	0.00	0.15	0.30	0.00	0.15	0.30	0.00	0.15	0.30	Nisin (g/kg)	0.00	0.10	0.20	0.00	0.10	0.20	0.00	0.10	0.20
Y_0 (ln CFU/g)	7.010	6.614	6.111	8.228	7.635	7.137	5.597	6.740	5.901	Y_0 (ln CFU/g)	7.131	6.458	6.231	6.941	6.605	6.280	6.915	6.423	6.099
L95CI	6.588	6.389	5.920	7.571	7.009	6.603	4.391	6.439	5.289	L95CI	6.912	6.210	6.112	5.325	5.473	5.541	5.524	5.045	5.144
U95CI	7.531	6.839	6.302	8.884	8.261	7.671	6.802	7.040	6.513	U95CI	7.351	6.707	6.350	8.557	7.738	7.020	8.306	7.800	7.053
Y_{max} (ln CFU/g)	15.791	11.883	11.043	18.963	19.007	18.604	17.971	17.176	16.968	Y_{max} (ln CFU/g)	16.107	12.959	11.545	18.880	18.933	18.746	18.224	18.120	18.284
L95CI	15.360	11.605	10.724	18.499	18.507	18.054	16.898	16.935	16.394	L95CI	15.913	12.626	11.364	17.916	18.061	18.099	17.531	17.237	17.297
U95CI	16.222	12.161	11.363	19.428	19.507	19.153	19.045	17.417	17.541	U95CI	16.301	13.292	11.725	19.844	19.806	19.394	18.917	19.004	19.272
μ_{max} (h^{-1})	0.074	0.034	0.029	0.237	0.208	0.184	0.535	0.536	0.594	μ_{max} (h^{-1})	0.106	0.044	0.035	0.409	0.270	0.238	0.894	0.523	0.465
L95CI	0.058	0.028	0.024	0.197	0.180	0.163	0.432	0.502	0.505	L95CI	0.093	0.037	0.032	0.205	0.197	0.195	0.615	0.401	0.376
U95CI	0.090	0.039	0.034	0.278	0.236	0.206	0.637	0.571	0.683	U95CI	0.118	0.051	0.038	0.614	0.343	0.281	1.173	0.645	0.553
λ (h)	69.240	63.435	76.904	11.563	11.450	13.309	3.474	3.748	4.703	λ (h)	67.693	70.487	76.816	11.159	12.809	16.732	3.549	3.648	5.419
L95CI	53.915	47.852	61.112	6.136	5.877	7.866	0.330	2.747	2.707	L95CI	61.586	57.334	68.704	1.745	4.401	10.332	0.576	0.836	1.631
U95CI	84.566	79.018	92.696	16.989	17.024	18.752	7.279	4.748	6.699	U95CI	73.801	83.639	84.928	20.574	21.218	23.132	8.306	8.132	9.207

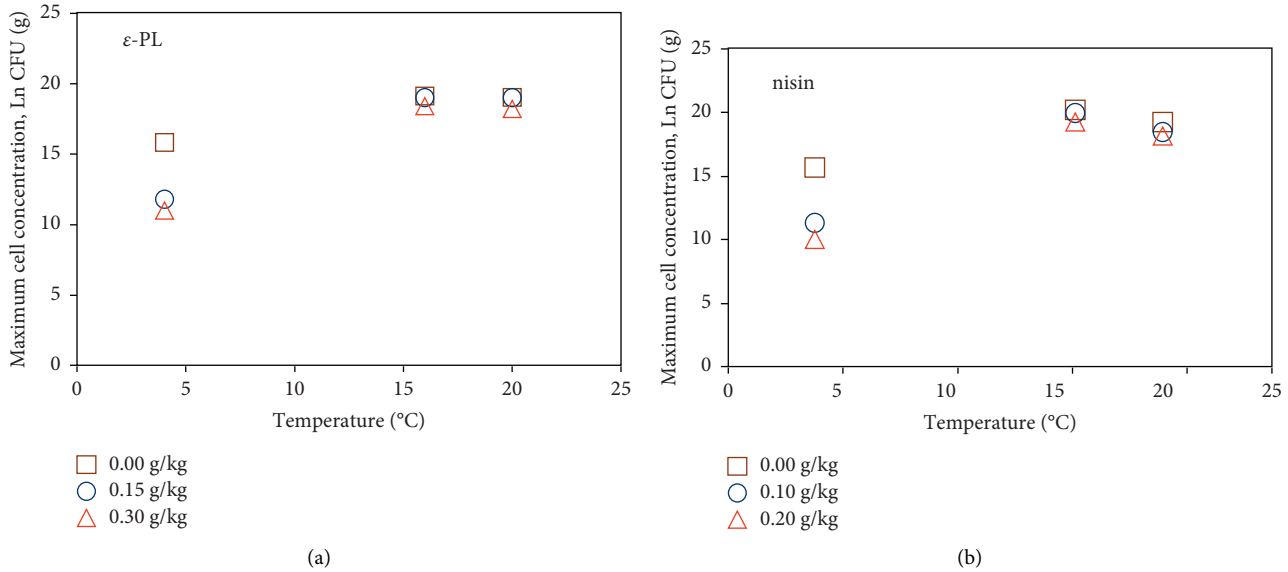

Figure 3: The maximum population of *L. lactis* in FVFB: (a) ε-PL treated and (b) nisin treated.

appeared that *L. lactis* in FVFB samples treated with ε-PL or nisin exhibited longer lag times than those in control samples (CK) at the same temperatures, suggesting that the addition of ε-PL or nisin to FVFB could delay the growth of *L. lactis* in FVFB. It has been previously reported and verified that nisin is effective in the lag phase extension of bacteria [38, 39]. In our previous study, ε-PL also exhibited an influence on the increase of bacterial lag phase [40]. A similar result was also reported by Qian et al. [41]. In addition, temperature also played an essential role in increasing the lag phase of *L. lactis*. The lower the temperature was, the longer the lag phase was. As bacterial cells could be damaged by ε-PL, nisin, or temperature [42], a longer time was needed to recover, leading to an increase of lag phase. The specific growth rates (μ_{max}) of *L. lactis* in CK were 0.074, 0.208, and 0.535 h^{-1} at 4, 16, and 20°C, which were 0.045, 0.053, and 0.059 h^{-1} higher than those in 0.30 g/kg ε-PL-treated FVFB. When nisin concentration was enhanced to 0.20 g/kg, μ_{max} of *L. lactis* were decreased to 0.035, 0.238, and 0.465 h^{-1} at 4, 16, and 20°C, respectively. The results indicated that nisin played a role in hindering bacterial growth rate. Similar phenomena were found by Qian et al. [41] and Prado-Acosta et al. [43]. Moreover, ε-PL or nisin could decrease μ_{max} of *L. lactis* in FVFB with the decrease in temperature, suggesting that ε-PL or nisin could control the growth of *L. lactis* in FVFB. The antibacterial effect of ε-PL or nisin against *L. lactis* in FVFB at 4°C was more substantial than 16 and 20°C. These results demonstrated that both ε-PL and nisin could cooperate with temperature, exhibiting effectiveness against *L. lactis* by increasing lag phase and decreasing growth rate.

Figure 3 shows the maximum bacterial populations of *L. lactis* in FVFB. The estimated maximum concentrations of *L. lactis* ranged between 11.043 and 19.007 Ln CFU/g (Table 1 and Figure 3). Overall, the maximum concentrations of *L. lactis* showed a light declining trend for all treatments as the storage temperature decreased. At high temperatures (16 and 20°C), the maximum concentrations of *L. lactis* were not affected by nisin or ε-PL. However, at a low temperature (4°C), in comparison with control, the maximum populations of *L. lactis* were reduced by around 3.908 and 4.748 Ln CFU/g in samples with 0.15 and 0.30 g/kg of ε-PL (Figure 3(a)) and by 3.148 and 4.562 Ln CFU/g in samples with 0.10 and 0.20 g/kg of nisin (Figure 3(b)). These results indicated that the maximum bacterial populations of *L. lactis* in FVFB were associated with storage temperatures and the concentrations of ε-PL and nisin. ε-PL and nisin, at their highest levels of 0.3 and 0.2 g/kg, exhibited the strongest antibacterial activities at 4°C among all storage temperatures.

The experimental growth data of BK microorganisms were fitted to the no lag phase model, and the estimated bacterial growth parameters are presented in Table 2. As shown in Figure 2, the no lag phase model was suitable for describing the growth behavior of BK microorganisms in FVFB with/without ε-PL or nisin addition at 4, 16, and 20°C. As shown in Table 2 and Figure 2, the specific growth rate of BK microorganisms was not affected by ε-PL or nisin. BK microorganisms could reach maximum population densities of around 18.702–20.601 Ln CFU/g at 4–20°C (Table 1). Under the same temperature condition, no difference of maximum bacterial populations was observed among samples with all treatments (Figure 4). These results indicated that ε-PL or nisin has no effect on inhibiting the growth of BK microorganisms in FVFB.

3.3. Analysis of Quality Characteristics. The quality characteristics of FVFB, including weight loss, color, and soluble solid content, are illustrated in Figure 5. As fresh FVFB is high in moisture content (approximately 90%), weight loss is perceived as one of the main factors of FVFB degradation during storage. The loss of weight could be relative to the tissue shrank in FVFB. Nasiri et al. [26] reported that weight

Table 2: Growth parameters of BK microorganisms estimated by the no lag phase model.

Temperature	4°C			16°C			20°C				4°C			16°C			20°C		
ε-Polylysine (g/kg)	0.00	0.15	0.30	0.00	0.15	0.30	0.00	0.15	0.30	Nisin (g/kg)	0.00	0.10	0.20	0.00	0.10	0.20	0.00	0.10	0.20
Y_0 (ln CFU/g)	14.515	14.517	14.553	15.155	15.194	15.161	14.071	14.107	14.123	Y_0 (ln CFU/g)	13.593	13.250	13.542	14.391	14.363	14.469	14.033	13.797	14.163
L95CI	14.328	14.321	14.354	14.849	14.885	14.861	13.768	13.784	13.809	L95CI	13.238	12.847	13.111	13.899	13.885	13.975	13.838	13.594	13.966
U95CI	14.701	14.713	14.753	15.460	15.504	15.462	14.374	14.430	14.438	U95CI	13.948	13.653	13.973	14.883	14.842	14.962	14.229	13.999	14.360
Y_{max} (ln CFU/g)	18.775	18.763	18.768	20.601	20.506	20.510	19.371	19.305	19.322	Y_{max} (ln CFU/g)	18.805	18.727	18.702	20.070	20.049	20.067	19.826	19.511	19.574
L95CI	18.678	18.660	18.663	20.128	20.032	20.043	19.097	19.008	19.030	L95CI	18.642	18.538	18.496	19.553	19.553	19.486	19.506	19.199	19.225
U95CI	18.872	18.866	18.873	21.073	20.980	20.978	19.645	19.602	19.613	U95CI	18.968	18.916	18.908	20.587	20.545	20.648	20.146	19.823	19.923
μ_{max} (h^{-1})	0.036	0.035	0.036	0.056	0.056	0.055	0.188	0.183	0.182	μ_{max} (h^{-1})	0.051	0.052	0.048	0.088	0.089	0.081	0.138	0.141	0.125
L95CI	0.033	0.032	0.032	0.048	0.047	0.048	0.162	0.155	0.155	L95CI	0.044	0.044	0.039	0.067	0.069	0.062	0.126	0.129	0.113
U95CI	0.039	0.039	0.039	0.064	0.064	0.063	0.214	0.211	0.209	U95CI	0.059	0.060	0.056	0.108	0.109	0.101	0.150	0.154	0.137

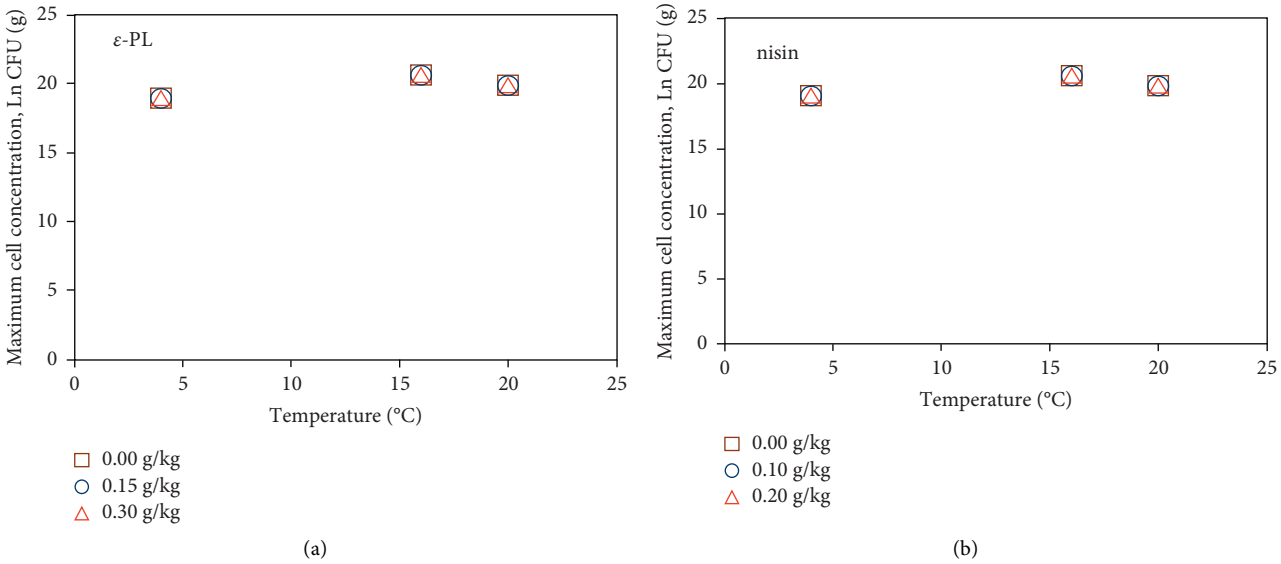

Figure 4: The maximum population of BK microorganisms in FVFB: (a) ε-PL treated and (b) nisin treated.

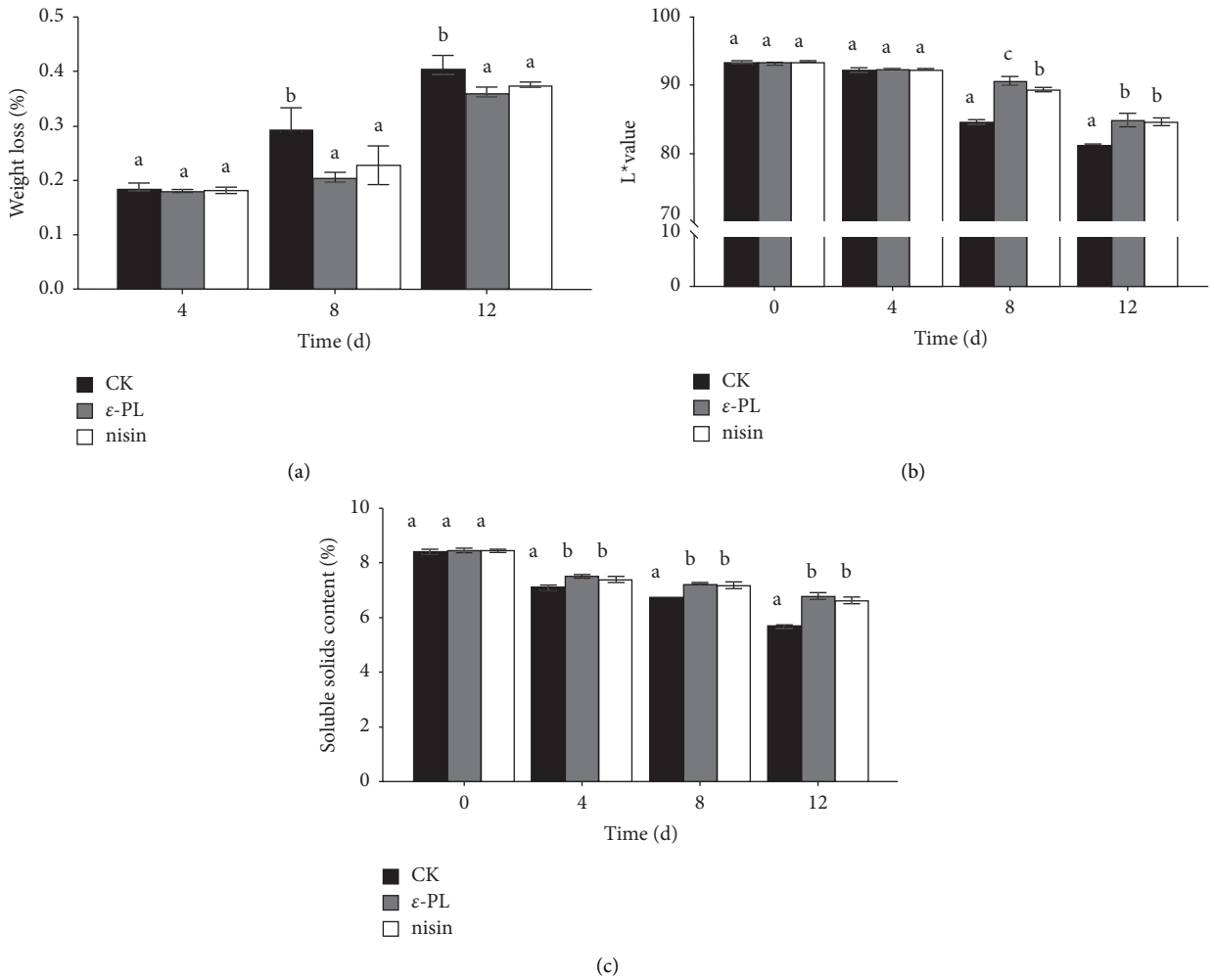

Figure 5: Changes of weight loss, color, and soluble solid content in FVFB. CK: FVFB treated with PBS; ε-PL: FVFB treated with 0.30 g/kg ε-PL; Nisin: FVFB treated with 0.20 g/kg nisin.

loss was associated with the unprotected thin epidermal structure and transpiration rates of *Agaricus bisporus*, which might provide a favorable environment for the growth of microorganisms. Additionally, microorganisms could destroy the fresh mushroom by breaking down the intracellular matrix and reducing the central vacuole, causing partial cell collapse. Figure 5(a) displays the weight loss shift for 12-day storage at 4°C. As shown in Figure 5(a), there was no significant difference in weight loss between ε-PL, nisin, and CK samples during 4-day storage at 4°C. However, in the period of 8–12-day storage at 4°C, the weight loss of FVFB was significantly prevented by ε-PL or nisin ($P < 0.05$), in comparison with CK samples.

The color parameter lightness (L^*) is used as a sign of FVFB darkening. Figure 5(b) shows the L^* value change of FVFB during storage for 0, 4, 8, and 12 days. Compared to CK samples, ε-PL or nisin could significantly increase the L^* value of FVFB ($P < 0.05$) after 8–12-day storage at 4°C. The result suggests that ε-PL or nisin exhibited an impact on preventing the browning processing of FVFB.

Soluble solid content enhancement was gradual during the storage time in ε-PL- or nisin-treated samples (Figure 5(c)). In the absence of ε-PL or nisin, soluble solid content displayed a decreasing tendency during storage. Furthermore, within 4–12 days of storage, the soluble solid content in CK FVFB was much lower than that in ε-PL- or nisin-treated FVFB. It suggests that ε-PL or nisin could prevent the decrease of soluble solid content in FVFB. The high value of soluble solid content was a sign of retarding the senescence process [44], which indicated that ε-PL or nisin was beneficial to extend the shelf life of FVFB.

The above results are in accordance with the findings of Barbosa et al. [45], who indicated that nisin maintained the physicochemical characteristics of mangoes throughout the storage period, and Fan et al. [46], who demonstrated that ε-PL showed a positive effect on preventing weight loss and color change of lettuce.

4. Conclusion

This study was conducted to investigate the effects of ε-PL and nisin on the growth of *L. lactis* and BK microorganisms in FVFB at various constant temperatures (4, 16, and 20°C). It was found that ε-PL or nisin could interact with temperature to inhibit the growth of *L. lactis*. With temperature decreasing and ε-PL or nisin concentration increasing, the inhibitory effect of ε-PL or nisin was strengthened. It was also found that ε-PL (0.15 and 0.30 g/kg) or nisin (0.10 and 0.20 g/kg) could extend the lag phase duration and reduce the specific growth rate. However, the growth of BK microorganisms in FVFB was not affected by ε-PL (0.15 and 0.30 g/kg) or nisin (0.10 and 0.20 g/kg). The developed reparameterized Gompertz model and no lag phase model could accurately predict the growth of *L. lactis* and BK in FVFB with or without ε-PL or nisin treatment, respectively. Moreover, the ε-PL or nisin could effectively restrain the decrease of weight loss, soluble solid content, and L^* value of FVFB during the storage period at 4°C compared with the control of untreated FVFB.

Data Availability

The data used to support the findings of this study are included within the article.

Additional Points

Applications. The results obtained in this study will be helpful for the food industry to extend the shelf life of *Flammulina velutipes* fruiting bodies.

Ethical Approval

The study does not involve any human or animal testing.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Acknowledgments

This project was supported by the Key Cultivating Program of Ningde Normal University (Grant no. 2020ZDK03).

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