

Production of Probiotic Kiwifruit Juice Containing *Lacticaseibacillus paracasei* B31-2: Investigation of Probiotic Viability, Physicochemical Properties, and AI Predictive Insights

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ABSTRACT

This study examined the viability, physicochemical characteristics, and sensory qualities of kiwifruit juice containing *Lacticaseibacillus paracasei* B31-2. To analyze the data, Gaussian Process Regression (GPR) and Multi-Layer Perceptron (MLP) models were used to predict various factors, including pH, acidity, viable cell counts of *L. paracasei* B31-2, color differences (ΔE), and overall acceptance. Probiotic *L. paracasei* B31-2 was added to the kiwifruit juice at different concentrations (0, 1 and 2%) and stored at 4°C. The probiotic juices showed fewer changes in pH, acidity, and color compared to the control juice during storage at room temperature. The sample with a 2% probiotic concentration exhibited the highest viable cell count (7.98 log CFU mL⁻¹) and received the most sensory scores among the tested samples. A strong correlation between the predictions made by the GPR model and the actual observed data further validated its effectiveness in similar experimental contexts. This suggests that GPR could offer strategic benefits by lowering laboratory costs and improving analytical efficiency. The GPR model's precision in closely matching real-world data demonstrates its potential as a cost-effective and expedited tool for scientific inquiries. Overall, these findings indicate that kiwifruit juice serves as a promising substrate for carrier of *L. paracasei* B31-2.

Keywords: Gaussian process regression, Microbial load, Multi-layer perceptron, Sensory analysis.

INTRODUCTION

Fruit juices have been widely acknowledged for their nutritional benefits and health-promoting qualities (Xiong *et al.*, 2022a). Packed with essential vitamins, minerals, antioxidants, and phytochemicals, fruit juices have become a popular choice for individuals looking for a refreshing and flavorful alternative to traditional drinks

(Salehi, 2020). In recent years, plant juices, notably fruit juices, have been gaining attention as a more nutritious option to milk drinks that are high in calories, saturated fat, and cholesterol (Gao *et al.*, 2022). As the focus on plant-based diets increases lactose intolerance problem, the demand for new alternatives like probiotic fruit juices has also been on the rise (Naseem *et al.*, 2023; Pérez-Rodríguez *et al.*, 2023). The rising

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demand for functional foods has spurred increased interest in probiotic applications for fruit juice processing. Recent studies have focused on fermenting Kiwifruit Pulp (KP) (Chen *et al.*, 2022; Zhu *et al.*, 2024) and Kiwifruit Juice (KJ) (Wang *et al.*, 2022) using lactic acid bacteria (LAB). Zhu *et al.* (2020) and Chen *et al.* (2022) employed three LAB strains to ferment KP, assessing the transformation of phenolic compounds and *in vitro* digestibility. Separately, Wang *et al.* (2022) investigated the fermentation of kiwifruit juices using *Lactiplantibacillus plantarum* and *Lactobacillus acidophilus*, respectively, monitoring changes in bioactive components and volatile aroma profiles. Collectively, these studies indicate that LAB fermentation improves the sensory properties of KP and KJ while boosting bioactive compound levels and antioxidant capacity. However, the outcomes varied significantly depending on the LAB strain and kiwifruit cultivar. However, incorporating probiotics into fruit juices presents unique challenges, including low pH, high sugar content, and the presence of antimicrobial compounds, which can reduce bacterial viability. Recent studies, such as the fermentation of kiwifruit juice by probiotic bacteria, highlight its potential to enhance bioactive phenolics, antioxidant activities, and flavor volatiles while maintaining probiotic viability (Wang *et al.*, 2022).

Kiwifruit (*Actinidia deliciosa*), is a nutrient-dense fruit rich in ascorbic acid, dietary fiber, carotenoids, polyphenols, and minerals. Its high phenolic content has been linked to immune system enhancement and prevention of chronic diseases. However, fresh kiwifruit is highly perishable due to its thin peel and climacteric nature, necessitating innovative processing methods to extend shelf life and diversify product applications. Fermentation with probiotics offers a promising solution to address these limitations while amplifying health benefits (Xiong *et al.*, 2022b; Zhang *et al.*, 2021; Sanz *et al.*, 2021; Wang *et al.*, 2021). Additionally, kiwifruit contains actinidin, an

enzyme that aids in digestion and may help promote gut health (Zeng *et al.*, 2024; Kaur *et al.*, 2022). Given its unique combination of nutrients and functional properties, kiwifruit has the potential to be a valuable addition to probiotic juices and beverages.

Probiotic juices have become increasingly popular due to their potential health benefits, specifically for digestive health (Kardooni *et al.*, 2023; Mojikon *et al.*, 2022). These beverages often contain various probiotic strains, such as *Lactobacillus* and *Bifidobacterium*, known to support digestive health, enhance the immune system, and improve overall well-being (Gomes and Malcata, 1999). Probiotics are live microorganisms that, when consumed in sufficient quantities, can offer a range of health advantages by improving the balance of beneficial bacteria in the gut (FAO/WHO, 2002). By combining the probiotic properties of these beneficial microorganisms with the nutritional benefits of fruit juices, probiotic juices provide a convenient and delicious way to promote gut health and overall wellness (Pimentel *et al.*, 2019).

LAB are a group of beneficial bacteria that play a crucial role in the fermentation process of beverages. As Gram-positive, non-spore-forming microbes, LAB are uniquely adapted to thrive in low-pH and anaerobic environments, making them ideal for food fermentation (Alebooye *et al.*, 2023). Their primary metabolic pathway—homolactic or heterolactic fermentation enables them to convert sugars (e.g., glucose, fructose) into lactic acid, acetic acid, and other bioactive compounds. This acid production not only helps preserve the beverage by inhibiting pathogenic bacteria but also creates a tangy flavor profile characteristic of fermented products. Additionally, LAB exhibit strain-specific probiotic properties, including bile salt hydrolase activity, adhesion to intestinal epithelium, and immunomodulation (Ghazanfari *et al.*, 2024; Wuyts *et al.*, 2018). One specific LAB strain that has shown promise for beverage fortification is

Lactocaseibacillus paracasei, previously known as *Lactobacillus paracasei*. This probiotic bacteria strain has been extensively researched for its ability to survive the acidic stomach environment, colonize the intestines, and provide a variety of health benefits, including immune support, improved digestion, and reduced inflammation (Bengoa *et al.*, 2021). By incorporating *L. paracasei* into fruit juices, beverage manufacturers can enhance the functional properties of their products and provide consumers with a flavorful and nutritious way to support their gut health (Mantzourani *et al.*, 2023).

Artificial Neural Networks (ANNs) are revolutionizing the field of food science and technology by providing sophisticated tools for complex problem-solving (Chen *et al.*, 2025). ANNs, inspired by the neural processing of the human brain (Sui *et al.*, 2014) are capable of learning from data, identifying patterns, and making predictions with remarkable accuracy (Chu *et al.*, 2025). In food science, ANNs are being applied to a myriad of tasks, such as predicting the sensory properties of food products, optimizing processing parameters, and ensuring food safety (Neto *et al.*, 2021). For instance, ANNs have been utilized to model microbial growth, thereby aiding in the prediction of food safety and shelf life (Deng *et al.*, 2019). They also play a crucial role in interpreting spectroscopic data, which is essential for assessing the quality and authenticity of food products (Feng *et al.*, 2025a). Moreover, ANNs contribute to the understanding of the complex relationships between food components and their functional, chemical, and physical properties during processing and distribution. In the realm of nutrition, ANNs are instrumental in dietary assessment and in analyzing the gut micro-biome profile, which is pivotal for personalized nutrition plans (Feng *et al.*, 2025b). They also assist in the toxicity prediction of food ingredients, ensuring consumer safety and compliance with regulatory standards (Gao *et al.*, 2025).

The integration of ANNs with other technologies, such as electronic noses, electronic tongues, computer vision systems (Sui *et al.*, 2013), and Near-Infrared Spectroscopy (NIR) further enhances their utility in the food industry (He *et al.*, 2025). These combined systems can accurately classify foods, determine quality, and control food processing tools, thereby streamlining operations and improving product consistency (Liu *et al.*, 2023).

As the demand for food increases with the growing global population, ANNs are set to become an indispensable asset in the food industry (Li *et al.*, 2019). Their ability to handle large datasets and complex calculations makes them ideal for addressing the challenges of food production, safety, and quality assurance (Lu *et al.*, 2024). The future of food science and technology is bright with the continued development and application of ANNs, promising enhanced efficiency, sustainability, and innovation in the field (Shi *et al.*, 2025).

Zhao *et al.* (2024) applied ANNs to assess the interdependence of information in today's digital landscape. Through model path analysis, it was discovered that both anticipatory confirmation and perceived usefulness significantly contributed to explaining the variance in financial sustainability, at 61 and 74%, respectively. The deployment of a deep ANN with a multilayer perceptron architecture enhanced the predictive precision for perceived usefulness, achieving a training accuracy of 87.54% and a testing accuracy of 90.34%. This advancement has enriched the comprehension of the dynamics involved in utilizing green food safety data. Furthermore, a separate investigation conducted by Wang *et al.* (2023) employed ANNs to determine the key elements influencing the online live sale of fresh produce, guided by a push-pull framework. The findings indicated that the neural network model predicted live sale outcomes with an 83.76% accuracy rate. The insights gained from these studies offer a solid theoretical foundation for forecasting



consumer behavior in purchasing fresh food via live-streaming platforms, considering both consumer and supply chain perspectives. Additionally, these findings suggest strategies for crafting live broadcast content and enhancing the overall user experience in e-commerce environments.

In this study, we aimed to produce probiotic kiwifruit juice as an innovative beverage option for individuals looking to enhance their digestive health and overall well-being. Moreover, the effect of probiotic *L. paracasei* incorporation in kiwifruit juice on its physiochemical and sensory characteristics was investigated. In the next step, the study employed Gaussian Process Regression (GPR) alongside Multi-Layer Perceptron (MLP) to forecast a range of variables: pH, acidity, viable cells of *L. paracasei* B31-2, ΔE , and overall acceptance.

MATERIALS AND METHODS

Kiwifruit Juice Formulation

Fresh kiwifruits were obtained from a local market in Mollasani (Khuzestan Province, Iran). The fruits were peeled and juiced using a mechanical juicer. The resulting pulp was then centrifuged (6,000×g, 15 minutes, 4°C) to obtain clarified kiwifruit juice. The juice was standardized to an initial pH of 4.0 using a 50 mg/mL food-grade Na₂CO₃ solution and adjusted to 12.0 °Brix with potable water. Prior to fermentation, the juice was pasteurized at 60°C for 30 minutes (Wang *et al.*, 2022). To create the inoculum, the stock culture of *L. paracasei* B31-2 (previously isolated from yogurt samples in our earlier study (Alizadeh Behbahani *et al.*, 2024b) was activated by inoculating 100 µL of glycerol stock into 100 mL sterile de Man, Rogosa and Sharpe (MRS) broth (Merck, Germany) in a 250 mL Erlenmeyer flask. The bacterial cells were grown at 37°C for 18 h under static conditions until reaching late-log phase (OD₅₉₀ ≈ 0.60, ~9.0 log CFU

mL⁻¹), as measured using a Jenway 7305 spectrophotometer (England). Prior to inoculation, the cultured cells were aseptically centrifuged (3,000×g, 10 minutes, 4°C) to pellet the bacteria. The supernatant (MRS broth) was carefully decanted, and the cell pellet was washed twice with sterile phosphate-buffered saline (PBS, pH 7.2) to remove residual medium components that might affect juice fermentation. The final pellet was resuspended in 10 mL of fresh PBS to standardize the cell density. Different concentrations of this standardized inoculum (0, 1, and 2% v/v, respectively) were aseptically added to pasteurized kiwifruit juices. The probiotic juices were manually stirred to ensure homogeneous distribution of cells, and incubated for 21 days at 4 (refrigeration) and 25°C (room temperature) to simulate different storage conditions (Kardooni *et al.*, 2023).

Changes in pH Value

The pH meter (Metrohm, Switzerland) was used to determine the pH of the probiotic juice samples that had been stored at both 4 and 25°C for 21 days (Vasiee *et al.*, 2025; Han *et al.*, 2015).

Acidity Analysis

Titrateable acidity was determined by titrating each sample mixture (6 g 50 mL⁻¹ water) with 0.1 N NaOH (Merck, Germany) until a pH of 8.2 was reached, and the result was expressed as a percentage of citric acid (% acid) (Falah *et al.*, 2021, Yang *et al.*, 2025).

Change in Microbial Load

To determine viable cell counts, samples were serially diluted with sterile peptone water. Following that, 0.1 mL portions of the dilution were plated in triplicate on MRS

agar plates. The plates were subsequently incubated at 37°C for 72 hours. Then, plates with 20–350 colonies were enumerated, and the outcomes were documented as Colony-Forming Units (CFU) per millilitre of solution, in accordance with the methodology outlined by Pereira *et al.*, (2011).

Color Analysis

The colorimeter (Minolta CR300, Japan) was used to assessment the color of the probiotic juice. The instrument underwent calibration with illuminated D65, and color readings were acquired through an 8-mm port/viewing area. The colorimeter outputted three color metrics: Lightness (L^*), redness (a^*), and yellowness (b^*). Subsequently, the numerical values of L^* , a^* , and b^* underwent transformation into total color difference (ΔE) through Equation (1) (Costa *et al.*, 2013).

$$\Delta E = \sqrt{\Delta L^{*2} + \Delta b^{*2} + \Delta a^{*2}} \quad (1)$$

Sensory Evaluation

A group of twenty-five individuals, (15 women and 10 men, 25-35 years old), from the Food Science and Technology Department at Agricultural Sciences and Natural Resources University of Khuzestan in Mollasani, Iran, assessed the samples based on their odour, color, texture, and overall acceptability. A nine-point hedonic scale was utilized to assess the sensory characteristics of probiotic juices, including odour, color, and overall acceptability, with 1 indicating strong dislike and 9 indicating strong likeness. The samples, labelled with three-digit numbers, were randomly served in acrylic cups at 10°C. (Alizadeh Behbahani *et al.*, 2024a).

Statistics

The experiments were conducted thrice, and the data was analyzed using Minitab software (version 16) with a completely randomized factorial design. The average values were grouped based on the Tukey post-test, with a significance level established at $P < 0.05$.

Gaussian Process Regression (GPR) Model

The GPR emerges as a formidable statistical instrument adept at deciphering complex data configurations and offering predictions, even when faced with data that is fragmented or laden with noise (Ni *et al.*, 2025). This technique operates on the principle of a preliminary probabilistic model over a range of plausible functions that may correspond to the observed data, progressively honing this model as additional data is introduced (Sui *et al.*, 2010).

The GPR model's objective is to assimilate training data and adeptly extend the output distribution to new input scenarios (Tian *et al.*, 2013). Here, the model considers noise in the output to account for uncertainties stemming from factors beyond the input variable, like measurement errors. The presumption is that this noise is additive, with a zero mean, stationary, and randomly distributed (Tian *et al.*, 2019). This assumption allows for a more accurate representation of the underlying data structure and enhances the model's predictive capabilities. The general GPR can be shown by Equations (2) and (3) (Xiang *et al.*, 2025).

$$y = f(x) + \xi \quad (2)$$

y : The observed output variable

$f(x)$: The underlying latent function that maps input variables to output

x : The input variables

$\xi(x_i)$: Additive noise term representing measurement errors or unmodeled factors affecting the output

$$\xi \sim P(0, s_{\text{noise}}^2) \tag{3}$$

Where, s_{noise}^2 is the variance of the noise. The assumption of a Gaussian prior facilitates the interpretation of the function via the mean (Zhu *et al.*, 2015). Consistent with prevailing academic findings, the configuration of the mean function holds importance, primarily in regions without observations, and is customarily set to zero. This simplification aids in focusing on the covariance structure, which is pivotal in understanding the relationships within the data (Yang *et al.*, 2011).

Multilayer Perceptron (MLP) Neural Network

The MLP, a widely utilized type of neural network, is structured with several layers that include an input layer, one or more hidden layers, and an output layer. Each layer is comprised of neurons, which are the fundamental processing elements (Yao and Chen, 2016; Wang *et al.*, 2025). An example MLP configuration is depicted in Figure 1, showcasing a structure of MLP model with 3 hidden layers. During the network's

operation, each neuron applies a specific bias in its calculations.

The input layer receives n distinct input variables denoted as $X = \{X_1, X_2, X_3, \dots\}$ while the output layer produces m distinct output variables represented by $Y = \{Y_1, Y_2, Y_3, \dots\}$ (Zhiquan *et al.*, 2015). In this research, data normalization precedes the training of the neural network (Yao *et al.*, 2017). The purpose of normalization is to transform the data values to a range between zero and one. This step is crucial for both the training and testing phases of the network, as it ensures the learning algorithm operates effectively (Yao *et al.*, 2014). Without normalization, the network may fail to converge during training, leading to suboptimal outcomes. When employing a sigmoid activation function, the ideal range for data transformation is between 0.9 and 1.0 (Zhu *et al.*, 2024). The method of linear normalization was utilized for this data transformation process, which can be mathematically expressed by Equation (4) (Taki *et al.*, 2018; Zhao *et al.*, 2012).

$$x_n = \frac{x - x_{\min}}{x_{\max} - x_{\min}} \times (r_{\max} - r_{\min}) + r_{\min}$$

x : The original/unprocessed input data value from the raw dataset

x_n : The normalized data value after transformation

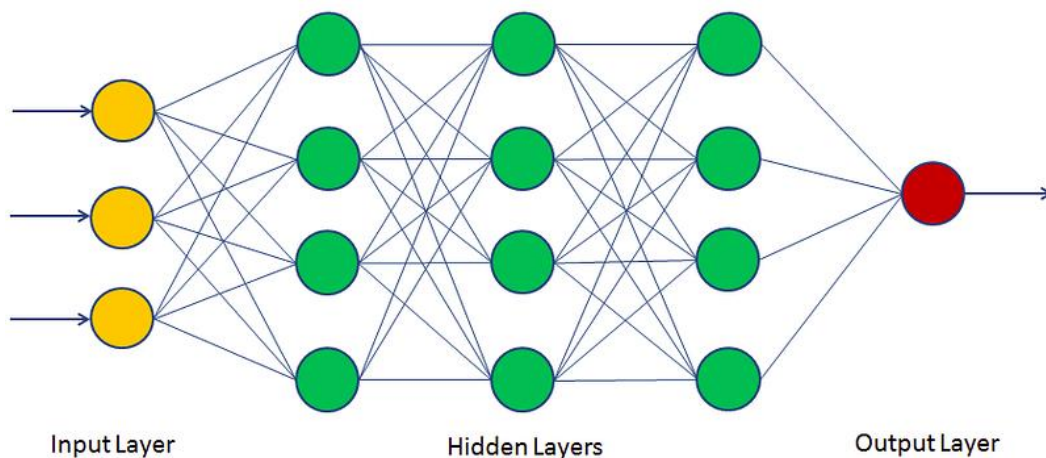


Figure 1. MLP neural network structure with 3 neurons in input, 3 hidden layer and one neuron in output (3-4-4-4-1).

The minimum value observed in the original raw dataset

xmin: The minimum value observed in the original raw dataset

xmax: The maximum value observed in the original raw dataset

rmin: The desired minimum limit of the new normalized data range

rmax: The desired maximum limit of the new normalized data range (4)

Where, x signifies the unprocessed initial dataset, while x_n denotes the data post-normalization. The highest and lowest values within the original dataset are symbolized by x_{max} and x_{min} , respectively (Zheng *et al.*, 2025). Conversely, x_{max} and x_{min} represent the new data range's maximum and minimum limits. The datasets are subjected to a thorough assessment through these normalization procedures, and the most advantageous technique is selected contingent upon the network's efficacy (Zhao *et al.*, 2011).

The research utilized the Levenberg-Marquardt, also known as Trainlm, as the back-propagation algorithm for the training phase (Zheng and Cao, 2024). The neuron count in the hidden layer was varied from 5 to 35, with the optimal number being chosen based on achieving the minimal error rate.

Performance Evaluation Criteria

$$MAPE = \frac{1}{n} \sum_{j=1}^n \left| \frac{T_{d_j} - T_{p_j}}{T_{d_j}} \right| \times 100 \quad (5)$$

$$R^2 = \left[\frac{\sum_{j=1}^n (T_{d_j} - \bar{Td})(T_{p_j} - \bar{Tp})}{\sum_{j=1}^n (T_{d_j} - \bar{Td}) \times \sum_{j=1}^n (T_{p_j} - \bar{Tp})} \right]^2 \quad (6)$$

To assess the performance of both GPR and MLP models, various statistical metrics were employed, including the Root Mean Square Error (RMSE), Mean Absolute Percentage Error (MAPE) and the coefficient of determination R^2 as the Equations (5) and (6) (Taki and Rohani, 2022; Zheng *et al.*, 2024).

Where, n is the experimental data, T_{p_j} is the predicted data by the models, T_{d_j} is the actual data and \bar{Td} and \bar{Tp} are the average values of actual and predicted data.

RESULTS AND DISCUSSION

Changes in pH Value and Acidity Analysis

It's crucial to carefully control the pH and acidity levels in probiotic juices to maintain the viability of the probiotics, preserve product quality, promote health benefits, and enhance taste and flavour. This close monitoring and control by manufacturers are essential in creating a successful probiotic beverage. The variations in pH and acidity levels of kiwifruit juices during storage are illustrated in Figure 2. The pH of the control sample of kiwifruit juices stored at 4°C on the first and 21st days was 3.2 and 3, respectively, while for the control sample stored at 25°C it was 3.2 and 2.9, respectively. The pH of the with 1% *Lactocaseibacillus Paracasei* (LP) ample of fruit juice stored at 4°C on the first and 21st days was 3.2 and 2.85, respectively, while for the control sample stored at 25°C it was 3.11 and 2.65, respectively. The pH of the with 2% LP sample of fruit juice stored at 4°C on the first and 21st days was 3.17 and 2.75, respectively, while for the control sample stored at 25°C it was 3.1 and 2.55, respectively. With an increase in storage time, temperature and probiotic concentration, the pH value significantly decreased by 12, 4, and 7%, respectively ($P < 0.05$) (Figure 2-a). The activity of the probiotic strain and enzymes found in kiwifruit juice may have been enhanced at ambient temperature, resulting in a more noticeable decline in pH. The titratable acidity of kiwifruit juices showed a similar trend in pH, as depicted in Figure 2-b. The observed pH reduction in kiwifruit juice samples correlated with increased acidity, consistent with the known logarithmic



inverse relationship between pH and hydrogen ion concentration. According to research by Tiekling *et al.* (2005), *L.*

composition within the juice. A study conducted by Kumar *et al.* (2015) also noted a similar trend, where mango and sapota

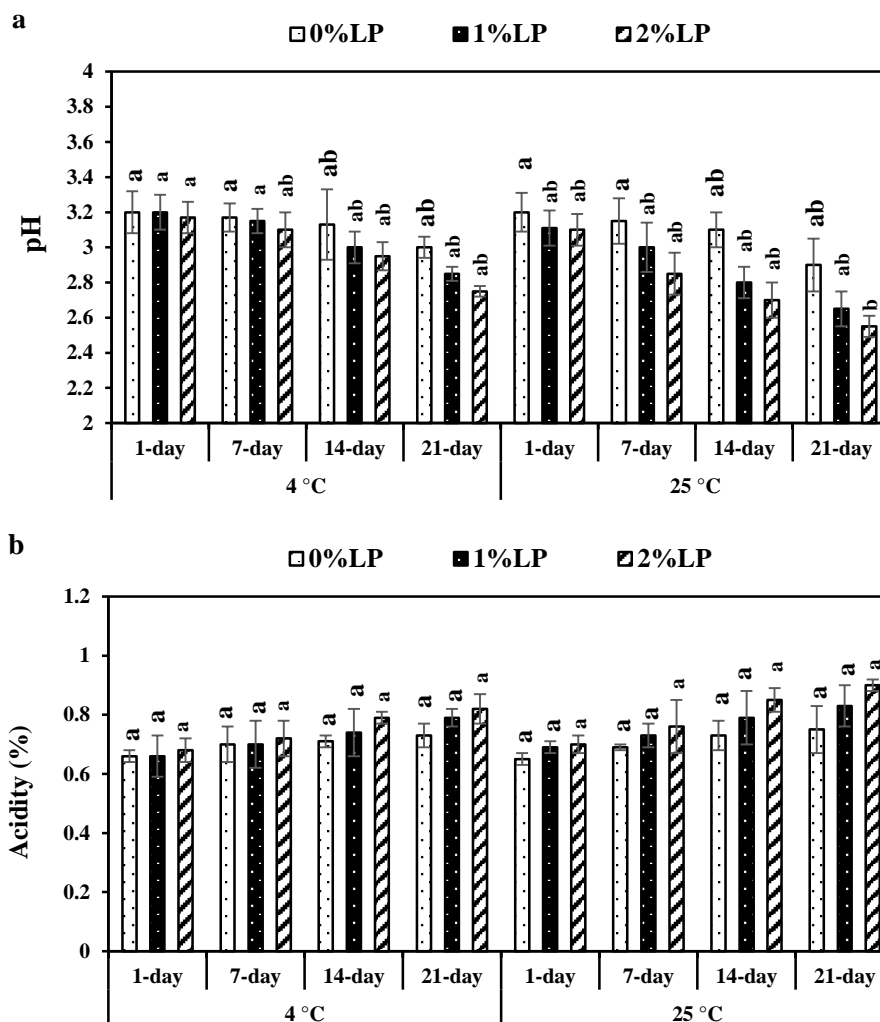


Figure 2. Fluctuations in pH (a) and acidity (b) of kiwifruit juices loaded with *L. paracasei* B31-2 during the storage period. Significant differences ($P < 0.05$) are represented by different superscript small letters.

sanfranciscensis demonstrated the ability to metabolize fructose through microbial processes, leading to a reduction in pH levels and an increase in titratable acidity in the samples. In the juice samples, the rise in acid levels could be attributed to the presence of various by-products resulting from the breakdown of fructose by *Lactobacillus*, which include lactic acid, acetic acid, and carbon dioxide. This collective influence of by-products positively impacts the overall acidic

juices inoculated with *Lactiplantibacillus plantarum* displayed an increment in acidity throughout the incubation time. Additionally, apart from the production of organic acids, an increase in acidity levels may also stem from the hydrolysis of juice sugars facilitated by enzymes released from lysed probiotic cells. This could be credited to the by-products resulting from the fructose breakdown by *Lactobacillus*, including lactic acid, acetic acid, and carbon dioxide (Peterson and Fred, 1920), all

positively influencing the overall acid content in the juice samples.

Color Analysis

During the storage period, a notable increase in the overall color change of the juices was observed (Figure 3). Additionally, it was noted that the samples containing *L. paracasei* B31-2 and stored at 4°C exhibited lower color changes compared to their respective counterparts. These findings align with a study by da Costa *et al.* (2017), who reported that both control and probiotic pineapple juice underwent darkening and became more yellow during storage. Pereira *et al.* (2011) optimized *L. casei* NRRL B-442 cultivation in cashew apple juice, determining ideal conditions (pH 6.4, 30°C, 7.48 Log CFU mL⁻¹ inoculum, 16 hours fermentation) and demonstrating its viability (> 8 Log CFU mL⁻¹) during 42-day refrigerated storage. The study noted increased lightness/yellowness and reduced redness over time, concluding that fermented cashew

juice is a viable probiotic functional food, comparable to dairy for *L. casei* growth. Shah *et al.* (2010) investigated probiotic survival (*Lacticaseibacillus rhamnosus* HN001, *Bifidobacterium lactis* HN001, *L. paracasei* LPC 37) in a model fruit juice with vitamins/antioxidants. While most additives failed to sustain viability (< 10 CFU mL⁻¹ after 6 weeks), vitamin C, grape seed extract, and green tea extract improved survival (4.29–7.41 Log CFU mL⁻¹).

Change in Microbial Load

The changes in the number of viable cells of *L. paracasei* B31-2 in the juice over the storage period are shown in Figure 4. With increased storage time and temperature, the count of viable cells of *L. paracasei* B31-2 decreased significantly by 17 and 5%, respectively ($P < 0.05$). Nevertheless, the samples loaded with 2% LP exhibited the greatest quantity of viable cells (7.98 Log CFU mL⁻¹) in comparison to those with 1% LP (7.59 Log CFU mL⁻¹) ($P < 0.05$). According to Zhu *et al.* (2020), a decrease in

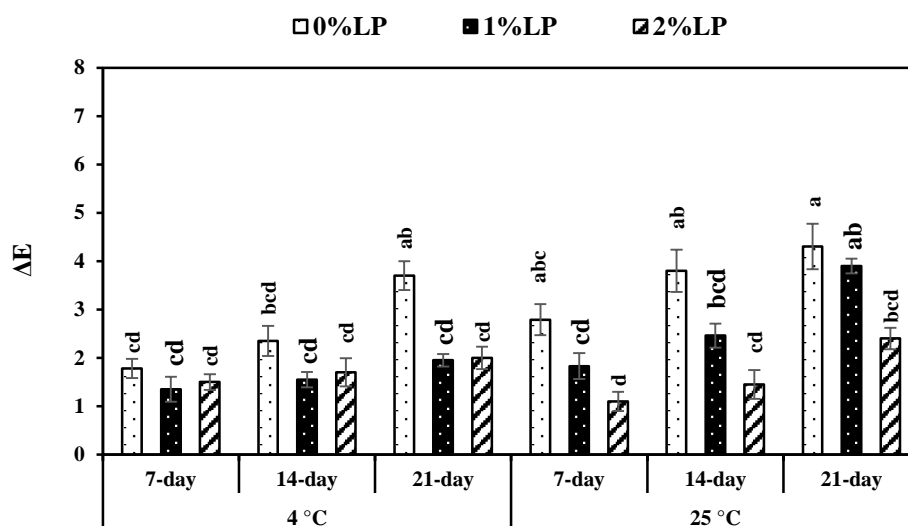


Figure 3. Fluctuations in total color difference (ΔE) of kiwifruit juices loaded with *L. paracasei* B31-2 during the storage period. Significant differences ($P < 0.05$) are represented by different superscript small letters.



probiotics' survival rate in food products has been linked to increased acidity. The improved preservation of probiotic cells may be attributed to the effective regulation of pH and acidity in kiwifruit juices stored at 4°C throughout the storage period (Figure 2). Similarly, the viability of *L. sanfranciscensis* in tomato, orange, and apple juices significantly decreased ($P < 0.05$) after 4 weeks of storage, due to the increasing lactic acid content, which influenced the subsequent viability of the probiotics (Zhu *et al.*, 2020). Although probiotic counts decreased, all juice samples maintained viability above the recommended threshold ($> 10^6$ CFU mL⁻¹) after 3 weeks. However, strain-specific acid tolerance must be considered. In our study, *L. paracasei* B31-2 showed a log reduction of 21 days, aligning with Okina *et al.* (2018), where *L. paracasei* ssp. retained $> 10^9$ CFU 200 mL⁻¹ in grape juice after 21 days. In contrast, Rodrigues *et al.* (2012) reported variable survival for *L. paracasei* L26 in low-pH juices, with counts declining below 10^6 CFU mL⁻¹ in some formulations by day 21. These disparities highlight the need for strain- and matrix-specific evaluations when designing probiotic juices.

Sensory Evaluation

Maintaining the quality of products by adding probiotics is crucial because consumers prefer functional beverages without strange aromas or unpleasant flavors, despite the health benefits (Pimentel *et al.*, 2015). The main effect of storage time on the sensory scores of samples was significant ($P < 0.05$), and the sensory scores for color, odor, and overall acceptance decreased generally from 8.15 to 4.33, 7.76 to 3.75, and 7.93 to 4.07, respectively, as storage time increased from 1 to 21 days (Figure 5). The decline in color scores for the control sample (without probiotics) was likely due to non-enzymatic browning (Maillard reaction) and pigment degradation, which are accelerated in the absence of protective compounds produced by probiotics, such as antioxidants or pH-stabilizing metabolites. Additionally, oxidation of phenolic compounds in the juice matrix may have contributed to undesirable color changes. The sensory scores were also impacted by the storage temperature, as cold storage yielded higher scores in comparison to ambient storage.

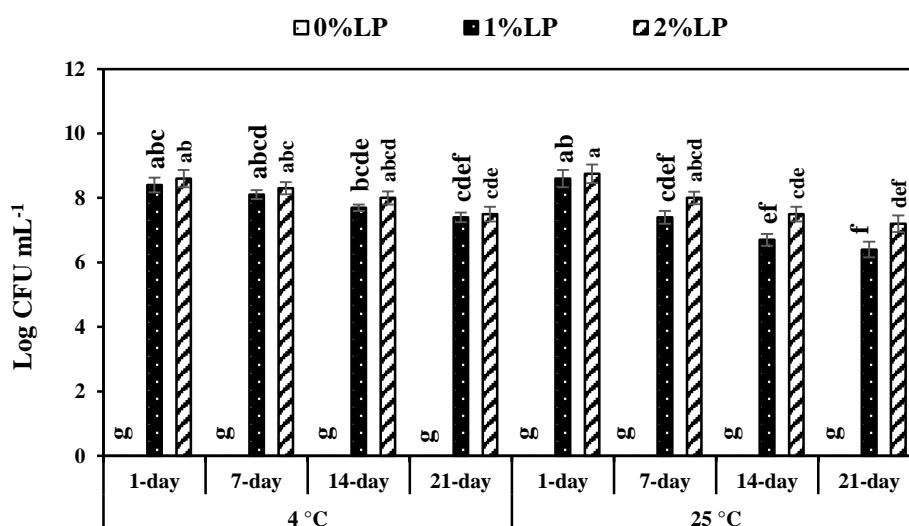


Figure 4. Fluctuations in microbial load of kiwifruit juices loaded with *L. paracasei* B31-2 during the storage period. Significant differences ($P < 0.05$) are represented by different superscript small letters.

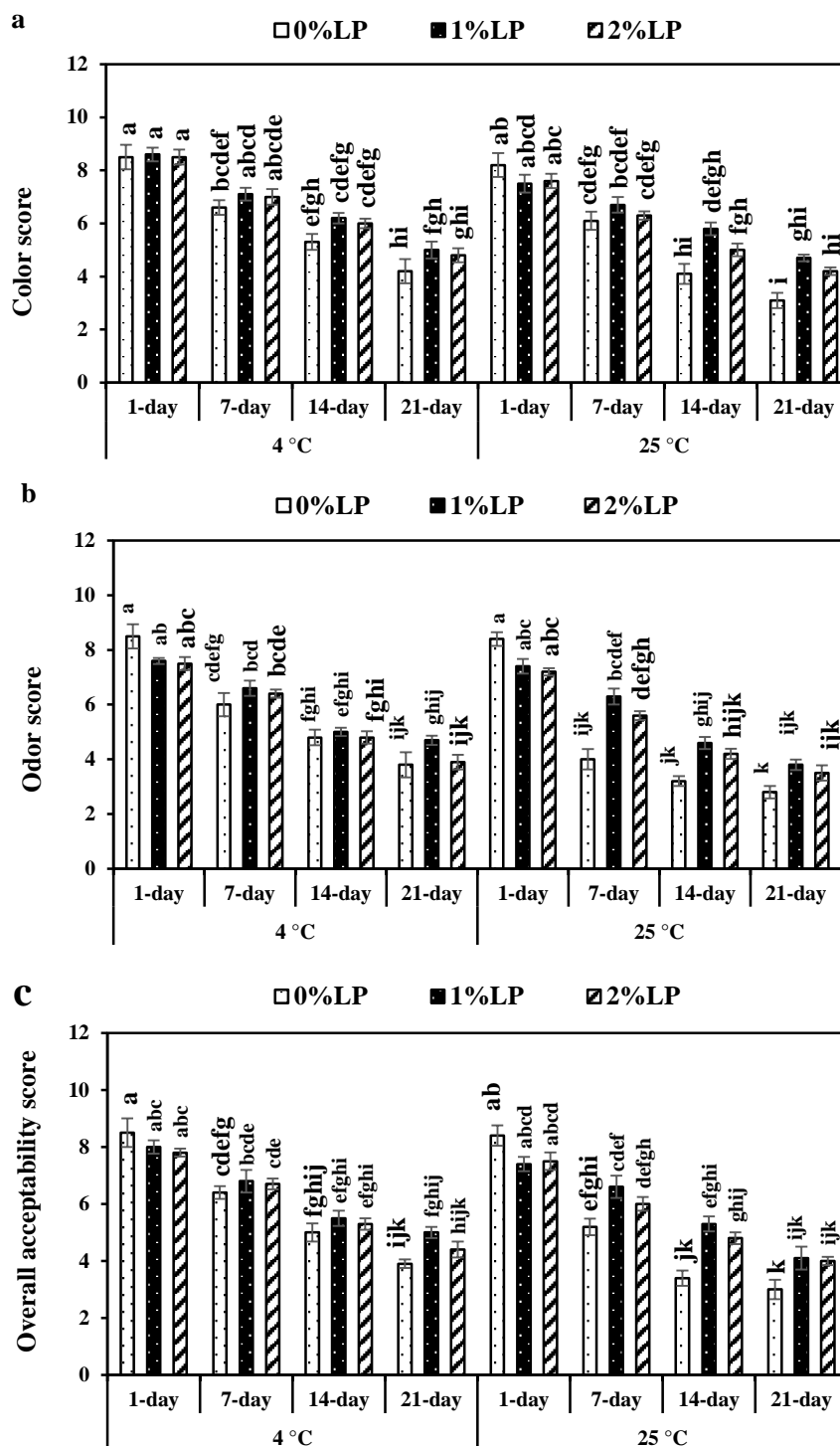


Figure 5. Fluctuations in color (a), color (b), and overall acceptance (c) of kiwifruit juices loaded with *L. paracasei* B31-2 during the storage period. Significant differences ($P < 0.05$) are represented by different superscript small letters.

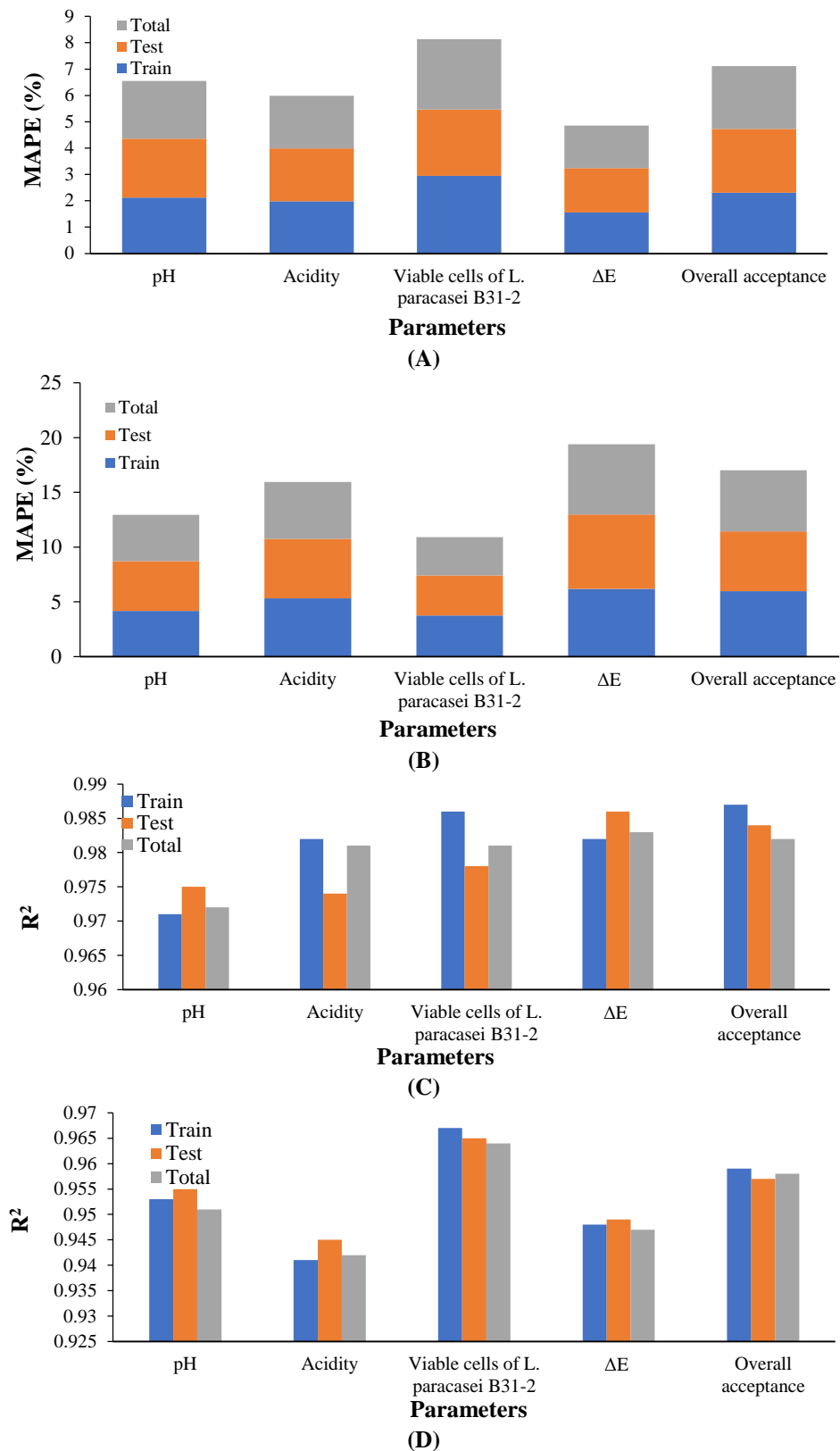


Figure 6. The results of modeling (A and C: using GPR model based on the MAPE and R² factors; B and D: Using MLP model based on the MAPE and R² factors.

Furthermore, the samples with probiotics exhibited significantly elevated sensory scores relative to the control samples, particularly when stored at 4°C. This can be ascribed to the probiotics' ability to stabilize acidity and inhibit oxidative reactions, thereby preserving color and flavour. Additionally, it was stated by Alizadeh Behbahani *et al.* (2024a) that peach juices containing *Levilactobacillus brevis* HL6 and stored at 4°C demonstrated the highest levels of sensory scores.

GPR and MLP Models

The results of Figure 6 show that GPR model outperformed the MLP in predicting pH, acidity, viable cells of *L. paracasei* B31-2, ΔE , and overall acceptance. This superior performance can be attributed to several factors inherent to GPR. Unlike MLP, GPR is a non-parametric approach, which means it does not make strong assumptions about the underlying function form, allowing for greater flexibility and adaptability to the data's intrinsic patterns. GPR's ability to incorporate prior knowledge through kernels enables it to reflect assumptions about the function's behavior, enhancing its predictive power. Furthermore, GPR provides uncertainty quantification for its predictions, a feature not typically available in MLP models, offering valuable insights for risk assessment and decision-making processes. The optimization of GPR's hyper parameters is generally more straightforward, avoiding the complex architecture tuning required by MLPs. Lastly, GPR's probabilistic foundation renders it more robust against overfitting, a common pitfall for MLP, particularly when dealing with limited or noisy datasets. The MAPE values, ranging from 1.62% for ΔE to 2.67% for Log, underscore GPR's precision in output prediction, which is crucial for the reliability and accuracy required in food science and technology applications. These factors

collectively contribute to the GPR model's higher accuracy in this context.

CONCLUSIONS

This particular LAB was tested for its ability to create probiotic kiwifruit juice. When stored at 4 °C, the probiotic juice retained its pH and acidity without any detrimental alterations. Moreover, it exhibited minimal color deterioration and sustained the highest viable cells at the conclusion of the storage duration. Additionally, the probiotic kiwifruit juice garnered the most favourable sensory scores. Based on the findings of this study, it can be concluded that *L. paracasei* B31-2 could be utilized as a probiotic culture for producing a healthy beverage from kiwifruit, which would be suitable for vegetarians or individuals allergic to lactose found in probiotic dairy products. In the conducted research, the efficacy of GPR was compared with MLP in forecasting a set of laboratory parameters such as pH, acidity, viable cells of *L. paracasei* B31-2, ΔE , and the overall acceptance. The findings revealed that GPR stands out as an exceptionally precise method for output prediction. This is particularly notable in situations where the dataset is limited in size. The robust analytical capabilities of GPR ensure that even with a smaller pool of data the model can deliver reliable and accurate predictions, making it a valuable tool for laboratory and research applications where data scarcity is a common challenge.

ACKNOWLEDGEMENTS

The authors would like to express their sincere gratitude to the Vice-chancellor for Research and Technology of Agricultural Sciences and Natural Resources University of Khuzestan for supporting this study as a project number 1.411.863.



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**تولید آبمیوه کیوی پروبیوتیک حاوی لاکتیکازنیاسیلوس پارانازی B31-2:
بررسی زنده مان‌ی پروبیوتیک، ویژگی‌های فیزی‌کوشیمیایی و بینش‌های پیش
بینی‌کننده مبتنی بر هوش مصنوعی**

بهرز علیزاده بهبهانی، حسین جوینده، مرتضی تکی، و فرشته فلاح

چکیده

این مطالعه به بررسی زنده مان‌ی، ویژگی‌های فیزی‌کوشیمیایی و ویژگی‌های حسی آبمیوه کیوی حاوی لاکتیکازنیاسیلوس پارانازی B31-2 پرداخت. برای تحلیل داده‌ها، از مدل‌های رگرسیون فرایند گاوسی (GPR) و پرسپترون چندلایه (MLP) به منظور پیش‌بینی عوامل مختلفی از جمله pH، اسیدیته، تعداد سلول‌های زنده *L. paracasei* B31-2، اختلاف رنگ (ΔE) و پذیرش کلی استفاده شد. پروبیوتیک *L. paracasei* B31-2 در غلظت‌های مختلف (صفر، ۱ و ۲ درصد) به آبمیوه کیوی افزوده شد و نمونه‌ها در دمای ۴ درجه سلسیوس نگهداری شدند. آبمیوه‌های پروبیوتیک در مقایسه با آبمیوه شاهد (کنترل) در

طول نگهداری در دمای اتاق، تغییرات کمتری در pH، اسیدیته و رنگ نشان دادند. نمونه های با غلظت ۲ درصد پروبیوتیک بیشترین تعداد سلول زنده ($\log \text{CFU mL}^{-1} 7.98$) را نشان داد و بالاترین امتیازات حسی را در میان نمونه های مورد بررسی کسب کرد. همبستگی قوی بین پیش بینی های مدل GPR و داده های واقعی مشاهده شده، کارایی این مدل را در زمینه های تجربی مشابه تأیید کرد. این امر نشان می دهد که GPR می تواند با کاهش هزینه های آزمایشگاهی و بهبود کارایی تحلیلی، مزایای راهبردی ارائه دهد. دقت مدل GPR در تطابق نزدیک با داده های واقعی، پتانسیل آن را به عنوان ابزاری مقرون به صرفه و سریع برای پژوهش های علمی نشان داد. در مجموع، این یافته ها حاکی از آن است که آمیوه کیوی بستر مناسبی برای حامل *L. paracasei* B31-2 محسوب می شود.