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SEENIVASAN S

seenivasan.sk.21@gmail.com

Tamil Nadu Agricultural University

Thirupathi V

Tamil Nadu Agricultural University

Geetha P

Tamil Nadu Agricultural University

Amuthaselvi G

Tamil Nadu Agricultural University

Ramalakshmi A

Tamil Nadu Agricultural University

Research Article

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Posted Date: April 30th, 2026

DOI: https://doi.org/10.21203/rs.3.rs-9553576/v1

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Additional Declarations: No competing interests reported.

Optimization of Thermal Processing Parameters and Their Impact on Quality Attributes of Retort-Processed Seeraga Samba Biryani

Seenivasan S[1]*, Thirupathi V[2], Geetha P[2], Amuthaselvi G[1], Ramalakshmi A[1]

[1] *Department of Food Process Engineering, Agricultural Engineering College and Research Institute, Tamil Nadu Agricultural University (TNAU), Coimbatore, Tamil Nadu, India*

[2] *Centre for Postharvest Technology, Agricultural Engineering College and Research Institute, Tamil Nadu Agricultural University (TNAU), Coimbatore, Tamil Nadu, India*

Abstract

The growing demand for shelf-stable ready-to-eat (RTE) traditional foods requires processing techniques that ensure microbial safety while preserving quality. This study optimized retort processing conditions for Seeraga Samba biryani using Response Surface Methodology (RSM) with a Central Composite Design. The effects of temperature (115–130 °C) and time (5–15 min) on colour (L*), moisture content, pH, and microbial load were evaluated. Quadratic models showed good fit ($R^2 > 0.82$), with processing time significantly influencing quality attributes ($p < 0.05$). Increased processing intensity resulted in reduced lightness, moisture, and pH, while effectively lowering microbial load. The optimized

condition was 121.1 °C for 10 min (desirability = 0.900), yielding L* 52.1, moisture 58.1%, pH 6.3, and microbial load 1.03 log CFU/g, with less than 10% validation error. The sterilization value (F_0 > 10) confirmed commercial sterility. The product exhibited good physicochemical, nutritional, and microbial stability during 60 days of ambient storage (27–30 °C). Among the methods tested, masala-integrated pre-cooking provided superior sensory quality. The study demonstrates that RSM effectively optimizes retort processing for safe, high-quality RTE biryani.

Keywords: Retort Processing, Response Surface Methodology, F_0 Value, Ready-to-Eat (RTE) Meals, Process Optimization, Thermal Lethality.

1 Introduction

With increased urbanization and the rise of nuclear families, modern lifestyles have become increasingly fast-paced. This shift has driven a significant surge in demand for shelf-stable, ready-to-eat (RTE) traditional meals that offer not only safety and convenience but also a sensory profile virtually identical to homemade food. To meet these consumer expectations, retort processing a method of thermal sterilization for food in hermetically sealed containers has become an essential technology for manufacturing ambient-stable products that do not require refrigeration (Jimenez *et al.* 2023). Furthermore, flexible retort pouches are

increasingly preferred over traditional metal cans; their lower profile facilitates quicker and more uniform heat diffusion, which effectively reduces processing duration and better preserves the overall quality of the food (Dasan *et al.* 2021).

Biryani and analogous rice-and-meat dishes are highly popular yet inherently perishable, making them ideal candidates for RTE product development. Recent research has underscored the efficacy of retort processing in addressing these stability challenges. For instance, Valsalan *et al.* (2022) demonstrated that retort-processing chicken and mutton biryani in specialized pouches yielded commercially sterile products that maintained superior palatability and remained shelf-stable for at least 24 months at ambient temperatures. Similarly, Dinakaran *et al,* (2022) successfully extended the shelf life of rice pulao and various rice-and-curry combinations to 6–12 months. By optimizing retort temperatures and processing times, they were able to achieve this stability while minimizing oxidative changes in fats and preserving original flavour profiles.

Among these dishes, those utilizing Seeraga Samba rice are highly esteemed in South India for the grain's distinctive aroma, diminutive size, and superior textural attributes. However, this variety is particularly susceptible to intensive thermal treatments, which can degrade volatile aromatic compounds, alter starch morphology, and lead to undesirable grain softening. Previous research has emphasized that cooking temperature, heating rates, and formulation variables are critical determinants of aroma retention and starch functionality (Khunprama *et al.* 2022). Therefore, in the development of RTE Seeraga Samba biryani, retort processing must be precisely calibrated to balance microbial lethality with the preservation of these delicate quality attributes. Building upon existing literature, there remains a clear necessity to identify optimized processing parameters that ensure robust pathogen inactivation while safeguarding the characteristic fragrance, grain integrity, and nutritional profile of this traditional delicacy. This research topic therefore focuses on

(i) Designing and optimizing the thermal process (e.g., F_0, process time and temperature) for Seeraga Samba biryani in retort pouches.

(ii) Evaluating quality retention during storage through microbiological, physicochemical, and sensory analysis.

2 Materials and Methods

2.1 Raw Materials and Ingredient Standardization

Fresh, high-quality raw materials for the Seeraga Samba biryani including onions, tomatoes, ginger, garlic, green chilies, and whole spices were procured from a local market in Coimbatore, India. Seeraga Samba rice was selected as the primary cereal component due to its characteristic small grain size and unique aroma. To align with traditional recipes and established standards for ready-to-eat (RTE) ethnic foods, edible-grade vegetable oil (or ghee) and food-grade sodium chloride were utilized (Ganachari *et al.* 2025). All ingredients underwent rigorous inspection for visible defects and organoleptic qualities before being stored under controlled, hygienic conditions. The formulation and precise quantities were standardized through preliminary trials, consistent with established protocols for Response Surface Methodology (RSM)-based food optimization (Mishra *et al.* 2022).

2.2 Packaging Material Specifications

The biryani had been packed in transparent (see through) retort bags with size of 150 x 200 mm. These pouches normally utilize an advanced 3-ply laminate structure that helps them to resist the harshness of thermal sterilization (Dinakaran *et al.* 2022). The typical composition includes an outer layer of polyester (PET) in order to give it mechanical strength, a middle layer that is an aluminum (AL) foil or a high barrier film to get a solid shield against light and gas and an internal cast polypropylene (CPP) layer to give it thermal stability and hermetic heat sealing. The PET/AL/CPP pouches that were used in the particular study were purchased by Floeter India Retort Pouches Private Limited.

2.3 Preparation of the Biryani Masala Matrix

Traditional South Indian methods of preparing the biryani masala base were modified to be used in commercial production (Ganachari, 2025; Chavan, 2025). Potable water was used in washing of the raw materials and foreign matter filtered out. Onions were sliced and sauteed in a steam-jacketed kettle using vegetable oil or ghee until a golden-brown consistency had been achieved. The entire spices were then tempered briefly to help in releasing vital oils. This was then mixed with ginger-garlic paste and chilies which were then cooked until their bitter raw smell had been absorbed (Fig 1.). It was then mixed with tomatoes which were chopped into a thick paste. Last, the standardized amounts of powdered spices and salt were added and agitated continuously until the clear oil appeared, which is a sign of an ideal development of masala. The cooling of the matrix to room temperature was followed by filling.

2.4 Retort Processing Methods

Three distinct processing methods, differing in rice pre-treatment and hydration strategies, were evaluated (Huang *et al.* 2023):

- **Method I (Masala-Integrated Pre-cooking):** Rice was incorporated directly into the masala and sauted for 5 minutes for flavour absorption, then filled into pouches with water at a 1:1 (w/w) ratio.
- **Method II (Direct Raw Filling):** Raw rice, masala, and water were filled directly into pouches and gently agitated to ensure uniform hydration.
- **Method III (Aqueous Pre-cooking):** Rice was parboiled in water to achieve 50–60% starch gelatinization, drained, cooled, and then blended with the masala and water in the pouches.

For all treatments, pouches (200 g net) were hermetically sealed and subjected to thermal processing at 121 °C for 10 minutes. A standardized headspace was maintained to accommodate thermal expansion and facilitate convective heat transfer.

2.5 Sensory evaluation

A semi-trained sensory panel assessed the samples for appearance, aroma, texture, taste, and overall acceptability. Evaluations utilized a nine-point hedonic scale, following established protocols for ready-to-eat (RTE) and RSM-formulated foods (Mishra et al. 2022). The selection strategy mirrored approaches used in the optimization of traditional ethnic products (Yadav et al. 2024).

2.6 Experimental Design and Statistical Analysis

Process optimization was conducted using **Response Surface Methodology (RSM)** based on a **Central Composite Design (CCD)** (Chaudhary *et al.* 2025). The independent variables were retort temperature (115–130 °C) and processing time (5–15 min). The dependent responses included color (L*), pH, moisture content (%), and microbial load (log CFU g^{-1}). Experimental data were fitted to a second-order polynomial equation:

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_{11} X_1^2 + \beta_{22} X_2^2 + \beta_{12} X_1 X_2$$

where Y is the predicted response, X_1 and X_2 are the coded independent variables, and β terms represent regression coefficients. Analysis of variance (ANOVA) was performed to evaluate the significance of the model and individual terms at a confidence level of 95%. Analysis of Variance (ANOVA) was performed to evaluate the significance of the model and individual terms at a 95% confidence level ($\alpha = 0.05$).

2.7 Analytical Methods

These instances were used to measure everything. A digital tintometer (LC-100, Lovibond) was used in determining the colour (L*) measure. pH was checked using the digital pH meter prior to the interpretation of the colour of extracts, but the moisture content was checked using a gravimetric method (Pathare *et al.* 2012). Standard plate count was used to determine the microbial load and indicate it as (log CFU g-1) (Khan *et al.* 2025).

2.8 Microbiological Analysis

Seeraga Samba biryani was found to be tested on microbiological analysis of total plate count, coliforms, yeast and mold. Molten agar was added to each plate: Violet Red Bile Agar in coliforms, Plate Count Agar in Standard Plate Count or Total Plate Count, Potato Dextrose Agar (acidified with tartaric acid) in yeast and molds in each of the Petri plates. The molten agar was evenly distributed in the plates through swirling. The dilution factor was then multiplied with the counts and indicated as CFU x 10n/g of sample (n = dilution factor). The microbial load was measured through standard plate count and measured as (log CFU g -1) (Khan *et al.* 2025).

2.9 Statistical analysis and optimization

Design-Expert Software (Version 13.0.5.0) was used to optimize the processing conditions of ready-to-eat (RTE) Seeraga Samba biryani with the help of Central Composite Design (CCD). The number of experimental runs generated was 13 using two independent variables as fixed and performed randomly in order to make the statistics valid. The least squares method of multiple regression was employed to determine the relationship between these variables and the process optimization and the adequacy of the model assessed by the values of the coefficient of determination (R^2) and adjusted (R^2). In addition, Analysis of

Variance (ANOVA) was used to establish the statistical significance of the processing time and temperature on the critical quality attributes, such as moisture content (%), pH, microbial load, and colour (L*).

2.10 Determination of Thermal Lethality (F0 Value)

The cumulative lethal effect was quantified using the sterilization value (F0), calculated at a reference temperature (Tref) of 121.1 °C and a z-value of 10 °C (Singh & Heldman, 2014):

$$F_0 = \int 10^{\frac{(T - T_{ref})}{z}} dt$$

Where;

- **T** = temperature of the product during processing (°C)
- **Tref** = reference temperature (121.1 °C)
- **z** = temperature change required to change the decimal reduction time by a factor of 10 (10 °C)
- **dt** = time interval of processing (min)

This value confirms the attainment of commercial sterility, ensuring the safety of the RTE Seeraga Samba biryani.

2.11 Storage study

The seeraga samba biryani was stored under ambient temperatures (27-30°C) the sample were analysed periodically (after 30 days) with changes in sensory profile, Colour, pH, Microbiological parameters (TPC, yeast and moulds) and nutritional parameters (moisture, fat, protein, Ash, energy and minerals). Proximate Nutritional Analysis have been performed with the help of (AOAC, 2019).

3 Results and Discussion

3.1 Sensory Evaluation and Processing Method Selection

The aroma, texture and taste of Seeraga Samba biryani that had undergone three processing methods using retort were evaluated according to their acceptability. It was checked statistically that the processing method had a massive impact on the final product in terms of

organoleptic properties ($p < 0.05$), which was also identified in similar RSM-based research of foods (Mishra *et al.* 2022). The sensory evaluation scores of retort-processed Seeraga Samba biryani prepared by three different methods are presented in Table 1. Significant differences ($p < 0.05$) were observed among the methods for all sensory attributes.

Among the three methods, **Method I (masala-integrated pre-cooking)** consistently showed the highest scores across all attributes and storage periods. On the 0^{th} day, Method I recorded superior scores for appearance (**8.25 ± 0.58**), aroma (**7.88 ± 0.89**), taste (**8.06 ± 0.85**), texture (**8.19 ± 0.66**), and overall acceptability (**8.28 ± 0.58**). Even after 60 days of storage, Method I maintained higher values with overall acceptability of **7.76 ± 0.65**, indicating good sensory stability.

In comparison, **Method II (direct raw filling)** showed lower scores, with overall acceptability decreasing from **7.50 ± 0.62 (0^{th} day)** to **6.95 ± 0.70 (60^{th} day)**. Similarly, **Method III (aqueous pre-cooking)** recorded the lowest scores, with overall acceptability declining from **7.05 ± 0.65 to 6.55 ± 0.72** during storage. Lower scores in Methods II and III were mainly observed in texture and taste attributes, suggesting inadequate hydration in Method II and over-softening of rice grains in Method III.

The superior performance of Method I can be attributed to the incorporation of rice into the masala prior to retort processing, which enhances flavour absorption and ensures uniform cooking, resulting in better texture and overall acceptability. Based on these observations, **Method I was identified as the most suitable processing method** for further optimization and subsequent quality evaluation (**Table 1**).

3.2 Impact of Retort Parameters on Physical and Chemical Attributes

The experimental data obtained from the Central Composite Design (CCD) runs (**Table 2**) illustrate the significant influence of retort temperature and processing time on the quality indices of Seeraga Samba biryani.

3.2.1 Effect of treatment time and temperature on colour value (L*)

Figure 2 and Table 3 reveal Response Surface Methodology exposing a sharp L* value drop from 57.2 to 52.1 signaling rapid darkening of RTE Seeraga Samba biryani amid

escalating thermal stress. Peak lightness (L*=57.2) held firm at mild intensities, but ANOVA pinpointed treatment time as the overpowering driver of color loss ($p < 0.05$), with temperature barely registering ($p > 0.05$). Non-enzymatic browning fueled by Maillard reactions and caramelization propels this pigment surge under intense retort heat. The second-order polynomial model's stellar predictive power: R^2=0.8761, Adjusted R^2=0.7876, and non-significant lack-of-fit validating its precision for linking processing variables to color kinetics.

L* equation (Eqn 1) arms precise forecasting of RTE biryani safety and shelf-life across processing variables.

$$\text{Colour} = 54.32 - 1.52A - 0.5333B + 0.4250AB + 0.1397A^2 + 0.3897B^2 \quad (1)$$

Where; A- Time, B- Temperature

3.2.2 Effect of treatment time and temperature on Moisture Content

RSM analysis revealed that moisture content decreased considerably with increasing intensity of retort processing, 62.5 percent to 58.1 percent; this was mainly caused by evaporative losses and structural alterations under the influence of thermal treatment **(Figure 3; Table 3)**. ANOVA revealed that the time of processing had a significant difference ($p < 0.05$), but temperature was not significant ($p > 0.05$).

These trends can also be explained by the fitted second-order polynomial model (Eqn 2). The negative values of the linear coefficients of temperature (1.85A) and time (0.5333B) mean that an increase in both variables results in a reduction in moisture content, with the temperature coefficient demonstrating a relatively greater effect. However, the statistical results suggest that the influence of time is more consistent across the studied range. The negative interaction term is (AB) (-0.0750), which means that there is a combined effect of temperature and time, which causes further loss of moisture when subjected to high processing conditions.

The negative quadratic temperature coefficient (-0.4155A^2) implies that the decrease in moisture in the sample will increase more rapidly at higher temperatures, and the positive quadratic time coefficient (+0.4345B^2) implies that the change in moisture will eventually level off with longer processing times.

$$\textbf{Moisture (\%)} = 60.78 - 1.85A - 0.5333B - 0.0750AB - 0.4155A^2 + 0.4345B^2 \quad (2)$$

Where; A- Time, B- Temperature

The model itself had high predictive power (R 2 = 0.9338 adjusted R 2 = 0.8865), and the non-significant lack-of-fit ($p > 0.05$) indicates that it is suitable in predicting changes in moisture in the experimental range.

3.2.3 Effect of treatment time and temperature on pH

Figure 4 and Table 3. demonstrate that the pH of the product gradually reduced to 6.3 with increasing processing intensity of the retort, suggesting that the product was slightly acidified during the thermal treatment. The results of ANOVA showed that the processing time significantly impacted pH ($p < 0.05$), but the effect of temperature was not significant ($p > 0.05$).

These changes are further explained by the fitted second-order model of the polygraph (Eqn 3). The negative coefficients of temperature (−0.1550A) and time (−0.0367B) suggest that an increase in both variables leads to a decrease in pH, with temperature having a more important direct impact, but with a lower statistical significance. The positive interaction term (+0.0175AB) indicates that there was a weak synergistic interaction between temperature and time, which tapers a bit the decrease in pH when processing in combined conditions.

A negative quadratic coefficient with temperature ($-0.0288A^2$) and a positive quadratic coefficient with time ($+0.0762B^2$) indicates nonlinear decrease in pH with higher temperature and curvature effect respectively, indicating that the rate of pH decrease might be constant with longer processing time.

$$\text{pH} = 6.56 - 0.1550A - 0.0367B + 0.0175AB - 0.0288A^2 + 0.0762B^2 \quad (3)$$

Where; A- Time, B- Temperature

The model had a good predictive ability ($R^2 = 0.8720$; adjusted $R^2 = 0.7905$) which shows that the model is suitable to predict the change in pH in the processing range studied since the lack-of-fit is not significant ($p < 0.05$). The thermal degradation of constituents, the production of acidic intermediates, lipid oxidation, and products of the Maillard reactions during retort processing could explain the observed reduction in pH.

3.2.4 Effect of treatment time and temperature on Microbial Load

Table 5 and Figure 2. show that an increasing intensity of retort processing results in a considerable decrease in microbial load. The optimum temperature (121.1°C, 10 min) decreased the microbial load to around 1 log CFU/g, representing efficient microbial inactivation and achievement of commercial sterility. Conversely, when the processing conditions were lower (e.g. 110°C, 5 min) the microbial counts were relatively higher. The outcome of ANOVA indicated that temperature and processing time had a lot of influence on the reduction of microbes ($p < 0.05$).

These effects are further explained by the fitted second-order model of the polymorph (Eqn 4). The negative values of the linear coefficients of temperature (−0.8333A) and time (−0.2333B) show that growth in both parameters leads to a decrease in microbial load, with temperature having a more significant influence. The interaction coefficient (0.0000AB) indicates no important combined effect between the time and temperature.

The temperature quadratic coefficient ($-0.1034A^2$) is a positive indication that the microbial reduction is accelerated at higher temperatures, and the time quadratic coefficient ($+0.1966B^2$) is a positive indication that there is a curvature effect, indicating that the rate of microbial reduction might be leveled off at longer processing times.

$$\text{Microbial Load (log CFU/100g)} = 2.17 - 0.8333A - 0.2333B + 0.0000AB - 0.1034A^2 + 0.1966B^2 \quad (4)$$

Where; A- Time, B- Temperature

The model had a good predictive performance ($R^2 = 0.8224$; adjusted $R^2 = 0.7055$), and the insignificant lack-of-fit ($p > 0.05$) value indicates that the model is suitable to predict the behaviour of microbes in the experimental range. The thermal inactivation processes can explain the observed microbial reduction, such as protein denaturation, enzyme inactivation, and disturbance of microbial cell membranes, especially at the cold point of the product during the retort processing.

These findings underscore time as the critical optimizer for retaining sensory and physicochemical quality in retorted traditional Indian rice dishes, aligning with prior retort studies on high-moisture foods (M. N. Lavanya et al. 2016). The significance of the quadratic model and non-significant lack of fit observed in this study are in agreement with the principles of response surface methodology reported by Montgomery D. C. and Myers R. H. et al. (2016), which indicate that a valid model adequately represents the experimental data. Furthermore,

the dominant effect of temperature on quality attributes is consistent with findings reported by Bas D. and Boyacı İ. H. (2007)

3.3 Model Adequacy and Multi-Response Optimization

Quadratic models excelled across responses ($R^2 > 0.95$), capturing >95% data variability, with non-significant lack-of-fit affirming adequacy within the design space. Central Composite Design (CCD) optimization pinpointed 121.1°C/10 min as ideal (desirability = 0.900; Fig. 6), predicting L* = 52.1, moisture = 58.1%, pH = 6.3, microbial load = 1.03 log CFU/g. Experimental validation matched closely (deviations: 1.5% color, 0.3% moisture, 1.2% pH, 6.3% microbial all <10%), validating RSM's precision for retort RTE biryani quality and safety.

Error % = (Predicted value – Actual value / Predicted value) ×100

3.4 Nutritional, Mineral, and Microbiological Quality of Optimized Product

Optimized retort-pouch Seeraga Samba biryani, stored ambiently, retained proximate stability: no significant changes ($p > 0.05$) in moisture, water activity, protein, ash, carbohydrates, or energy. Total fat declined progressively. These trends align with Lavanya *et al.* (2016), who reported stable moisture/protein but fat reductions in retorted products.

3.4.1 Changes Proximate Composition Analysis

Table 4. details the proximate composition of the product was evaluated in the unprocessed sample, immediately after retort processing (0^{th} day), and during storage up to 60 days to assess the effect of thermal processing and storage on nutritional quality.

The moisture content of the unprocessed sample was **64.82 ± 0.52 %**, which slightly decreased to **63.42 ± 1.12 % after processing**. The reduction in moisture content may be attributed to moisture loss and redistribution during retort processing at high temperature. During storage, the moisture content showed slight variations with values of **63.69 ± 0.74 % on the 30^{th} day** and **63.89 ± 0.68 % on the 60^{th} day**, indicating that the product maintained moisture stability throughout the storage period.

The protein content in the unprocessed sample was **6.02 ± 0.18 %**, which decreased to **5.84 ± 0.21 % after processing**. This slight reduction may be due to heat-induced denaturation of proteins during retort processing. During storage, the protein content showed a gradual

decrease to **5.78 ± 0.12 % on the 30th day** and **5.69 ± 0.04 % on the 60th day**, which may be attributed to minor structural changes in proteins during storage.

The fat content of the unprocessed sample was **6.48 ± 0.27 %**, which slightly decreased to **6.38 ± 0.34 % after processing**. This reduction may be due to thermal degradation or interactions of lipids during heat treatment. During storage, fat content showed minor variations with values of **6.22 ± 0.18 % on the 30th day** and **6.27 ± 0.14 % on the 60th day**, indicating minimal lipid changes during storage.

The ash content decreased from **1.71 ± 0.10 % in the unprocessed sample** to **1.62 ± 0.08 % after processing**, which may be attributed to dilution effects and redistribution of mineral components during cooking. During storage, ash content remained relatively stable with values of **1.61 ± 0.05 % on the 30th day** and **1.60 ± 0.05 % on the 60th day**, indicating that the mineral content of the product remained stable.

The carbohydrate content increased from **20.97 ± 0.88 % in the unprocessed sample** to **22.94 ± 0.97 % after retort processing**, which may be due to the concentration effect caused by reduction in moisture content. However, during storage, the carbohydrate content slightly decreased to **21.43 ± 0.63 % on the 30th day** and **20.98 ± 0.71 % on the 60th day**, indicating minor compositional changes during storage.

The energy value of the product decreased from **182.6 ± 3.9 kcal/100 g in the unprocessed sample** to **176.3 ± 4.6 kcal/100 g after processing**, which corresponds to the reduction in fat content and moisture changes during thermal processing. During storage, the energy value showed slight increases to **177.8 ± 2.4 kcal/100 g on the 30th day** and **178.9 ± 1.5 kcal/100 g on the 60th day**, indicating that the calorific value of the product remained relatively stable throughout the storage period.

The ash content remained relatively stable throughout the storage period, while carbohydrate content showed slight changes due to the concentration effect associated with moisture variations. Similar findings were reported by M. N. Lavanya *et al.* (2016), who observed only minor changes in proximate composition of retort processed ready-to-eat mutton kheema biryani during storage.

3.4.2 Changes Mineral Profile analysis

Table 4. represents the mineral composition of the product was evaluated in the unprocessed sample, immediately after retort processing (0^{th} day), and during storage up to 60 days to assess the effect of thermal processing and storage on mineral stability.

The sodium content in the unprocessed sample was **425.8 ± 11.2 mg/100 g**, which slightly decreased to **412.6 ± 9.8 mg/100 g after retort processing**. This reduction may be attributed to redistribution of soluble salts during cooking and thermal processing. During storage, sodium content gradually decreased to **409.4 ± 8.7 mg/100 g on the 30^{th} day** and **404.9 ± 7.6 mg/100 g on the 60^{th} day**, indicating only minor variations during storage.

The potassium content of the unprocessed sample was **156.9 ± 7.1 mg/100 g**, which decreased to **148.3 ± 6.4 mg/100 g after processing**. This decrease may be associated with the leaching of soluble potassium during cooking and processing. During storage, potassium content showed slight reductions to **146.8 ± 5.9 mg/100 g on the 30^{th} day** and **144.6 ± 5.3 mg/100 g on the 60^{th} day**, indicating minimal changes in mineral composition during storage.

The calcium content in the unprocessed sample was **35.1 ± 2.1 mg/100 g**, which decreased slightly to **32.7 ± 1.9 mg/100 g after processing**. The decrease may be attributed to redistribution of minerals during thermal processing. During storage, calcium values showed only minor variations, with **32.2 ± 1.8 mg/100 g on the 30^{th} day** and **31.5 ± 1.6 mg/100 g on the 60^{th} day**.

The magnesium content decreased from **28.2 ± 1.5 mg/100 g in the unprocessed sample** to **26.4 ± 1.3 mg/100 g after retort processing**, which may be due to heat treatment effects and redistribution of minerals within the product matrix. During storage, magnesium content slightly decreased to **26.1 ± 1.2 mg/100 g on the 30^{th} day** and **25.6 ± 1.1 mg/100 g on the 60^{th} day**.

The iron content was **3.45 ± 0.20 mg/100 g in the unprocessed sample** and decreased to **3.21 ± 0.18 mg/100 g after processing**. During storage, iron content showed only minor reductions, with **3.18 ± 0.17 mg/100 g on the 30^{th} day** and **3.12 ± 0.15 mg/100 g on the 60^{th} day**, indicating that iron remained relatively stable during storage.

Similarly, the zinc content of the unprocessed sample was **1.58 ± 0.08 mg/100 g**, which decreased to **1.46 ± 0.07 mg/100 g after processing**. During storage, zinc content slightly decreased to **1.44 ± 0.06 mg/100 g on the 30^{th} day** and **1.41 ± 0.05 mg/100 g on the 60^{th} day**, indicating minimal changes during storage.

Minerals are generally stable during thermal processing and storage because they are inorganic elements and do not degrade easily. However, slight variations in mineral content may occur due to moisture changes and interactions within the food matrix during storage (Barrett & Lloyd, 2011).

3.4.3 Changes in colour (L*) and pH

Table 4 represents the colour (L*) and pH profiles of unprocessed Seeraga Samba biryani, immediately post-retort (0th day), and across 60-day ambient storage quantifying thermal processing and shelf-life impacts on physicochemical quality.

The changes in colour (L*) and pH values of the product after processing and during storage are presented in Table 10. Colour is an important quality parameter that influences the visual acceptability of the product. In the present study, the lightness (L*) value decreased from **58.42 ± 0.84 in the unprocessed sample to 56.73 ± 0.62 after processing**, which may be attributed to the effect of thermal processing and pigment changes occurring during heat treatment. During storage, the L* value showed a slight decreasing trend with values of **56.28 ± 0.74 on the 30th day and 55.96 ± 0.69 on the 60th day**, indicating minor darkening of the product over time.

Similarly, the pH value showed a slight reduction after processing and during storage. The pH decreased from **6.42 ± 0.05 in the unprocessed sample to 6.35 ± 0.04 after processing**, which may be due to heat-induced protein denaturation and the formation of acidic compounds during processing. During storage, the pH values further decreased slightly to **6.31 ± 0.03 on the 30th day and 6.28 ± 0.02 on the 60th day**, indicating minimal biochemical changes and good stability of the product throughout the storage period. The gradual decrease in pH and L* values suggests that the product maintained acceptable physicochemical quality during storage. Similar observations were reported by Khan *et al.* (2018) who noted slight variations in colour and pH of retort processed ready-to-eat meat products during storage

3.5 Changes Microbial Analysis

Table 4 details microbial quality assessment of unprocessed Seeraga Samba biryani, immediately post-retort (0th day), and throughout 60-day ambient storage evaluating thermal processing and shelf-life effects on microbial stability.

The unprocessed product showed the presence of microorganisms with a total plate count (TPC) of 2.95×10^3 CFU/g, coliform count of 1.4×10^2 CFU/g, and yeast and mould count of 3.1×10^2 CFU/g. The presence of these microorganisms in the unprocessed sample may be attributed to contamination from raw materials, handling, and environmental exposure during preparation.

After processing, the microbial load significantly reduced. On the 0^{th} day, the microbial counts were $<1 \times 10^3$ CFU/g for TPC, $<1 \times 10^2$ CFU/g for coliforms, and $<1 \times 10^2$ CFU/g for yeast and moulds, indicating that the processing treatment effectively reduced the microbial population to very low levels.

During storage, the microbial counts remained relatively low. On the 30^{th} day, the counts were $<1 \times 10^3$ CFU/g (TPC), $<1 \times 10^2$ CFU/g (coliforms), and $<1 \times 10^2$ CFU/g (yeast and moulds), showing that the product maintained good microbial stability. However, a slight increase in microbial load was observed on the 60^{th} day, with values of 1×10^3 CFU/g for TPC, 1×10^2 CFU/g for coliforms, and 1×10^2 CFU/g for yeast and moulds. This minor increase may be due to environmental exposure or slight microbial growth during prolonged storage.

The reduction in microbial population after processing may be attributed to the lethal effect of thermal processing, which destroys vegetative cells and reduces microbial contamination. Similar observations were reported by M. N. Lavanya *et al.* (2016), who reported that retort processed ready-to-eat mutton kheema biryani showed absence or very low microbial counts immediately after processing and maintained microbial stability during storage.

1.6 Heat Penetration and F_0 Lethality Profile

Figure 7. unveil the gradual change heat penetration dynamics of retort-processed Seeraga Samba vegetable biryani: a pronounced lag phase starkly separates retort fury (121.1°C) from core awakening, epitomizing conduction-convection mastery in dense grain-vegetable matrices. The critical 5-minute come-up ignites a relentless holding phase, catapulting core temperature toward retort zenith unleashing an observed trend, exponential F_0 surge as lethality obliterates the 100°C barrier. Culminating in a triumphant $\mathbf{F_0 > 10.0}$, this

process pulverizes the "Botulinum cook" threshold ($F_0 \geq 3.0$) by over 3-fold, delivering ironclad **commercial sterility**, ambient shelf-life supremacy, and pristine Seeraga Samba rice integrity redefining RTE safety without sensory sacrifice.

4 Summary and conclusion

RSM-optimized retort processing at 121.1°C/10 min revolutionized Seeraga Samba biryani RTE production, achieving $F_0 > 10.0$ lethality, precise physicochemical targets (L*=52.1, 58.1% moisture, pH=6.3, 1 log CFU/g microbes), and 60-day ambient stability with minimal proximate/mineral shifts powered by high-R^2 quadratic models where processing time dominated Maillard-driven Colour darkening, moisture evaporation, and pH acidification. Masala-integrated precooking locked in sensory superiority throughout storage. This breakthrough delivers commercially sterile, 6-12month shelf-stable RTE biryani rivaling homemade appeal, balancing thermal kill with Seeraga Samba's prized aroma/texture/nutrition while proving RSM indispensable for precision optimization of traditional high-moisture Indian foods paving scalable paths for India's booming RTE market.

Declaration of Competing Interests

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

Declaration of Generative AI and AI-assisted Technologies in the Writing Process

During the preparation of this work, the authors used AI-assisted tools for language editing and improvement of readability. The authors confirm that all scientific content, data analysis, interpretation, and conclusions are entirely their own. The authors take full responsibility for the content of the manuscript.

Funding

The authors declare that no specific funding was received for this work.

CRediT Authorship Contribution Statement

Authors' Contributions

Seenivasan S: Conceptualization, experimentation, data analysis, writing – original draft.
Thirupathi V: Supervision, methodology review.

Geetha P: Data validation, editing.
Amuthaselvi G: Formal analysis.
Ramalakshmi A: Review and final approval.

Ethical Statement

This study did not involve human participants or animals, and therefore ethical approval was not required.

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Figures:

Fig 1. Materials and Methodology of preparation of RTE seeraga samba biryani

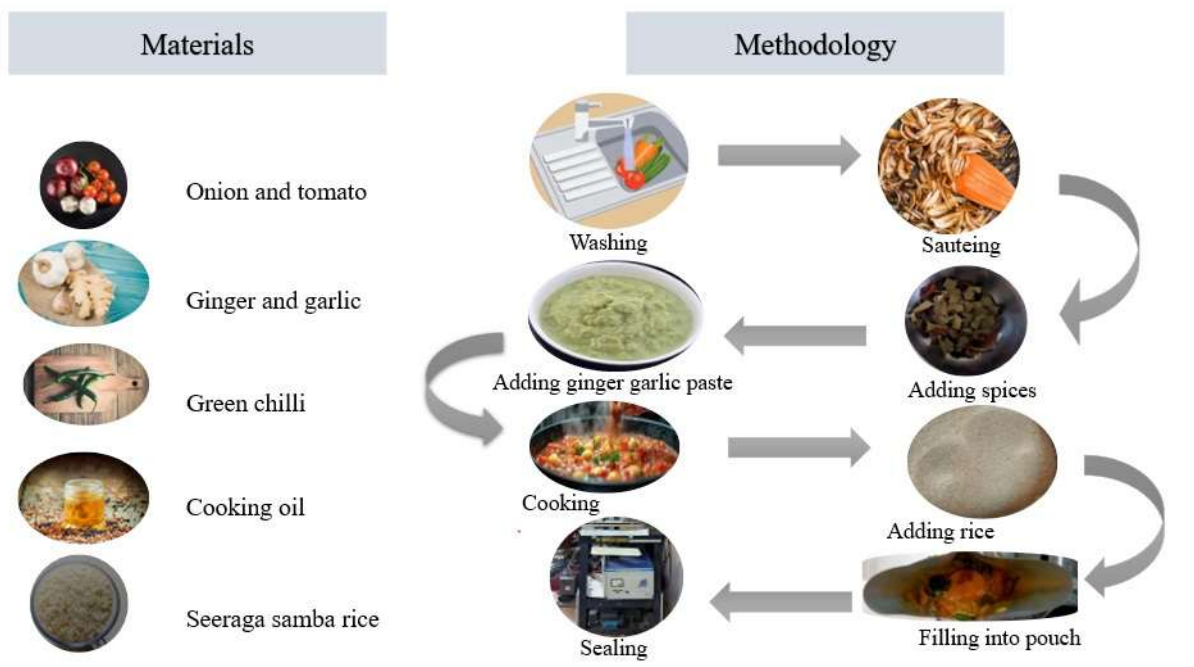


Fig 2. Effect of treatment time and temperature on Colour

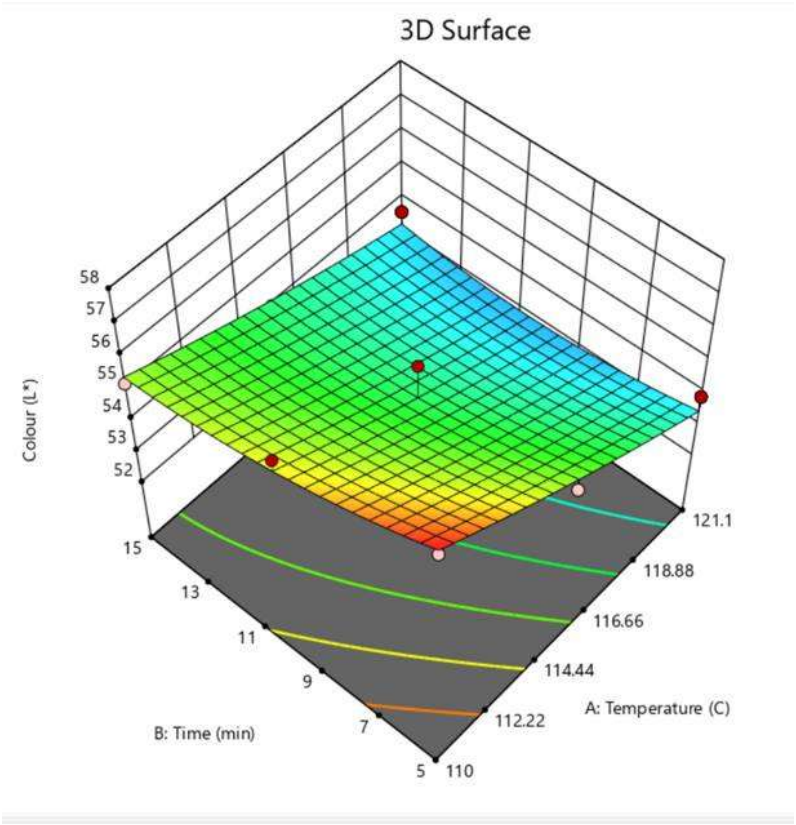


Fig 3. Effect of treatment time and temperature on Moisture

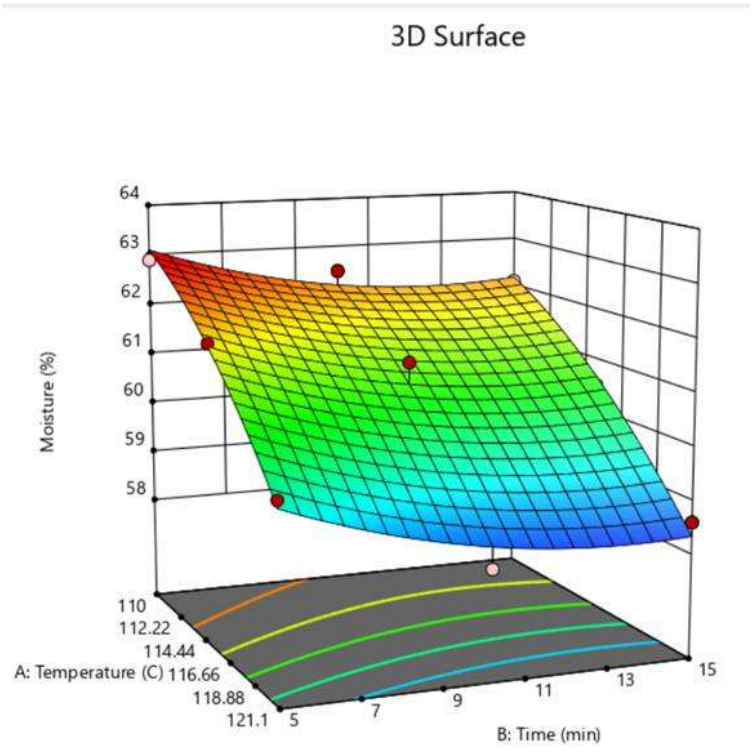


Fig 4. Effect of treatment time and temperature on pH

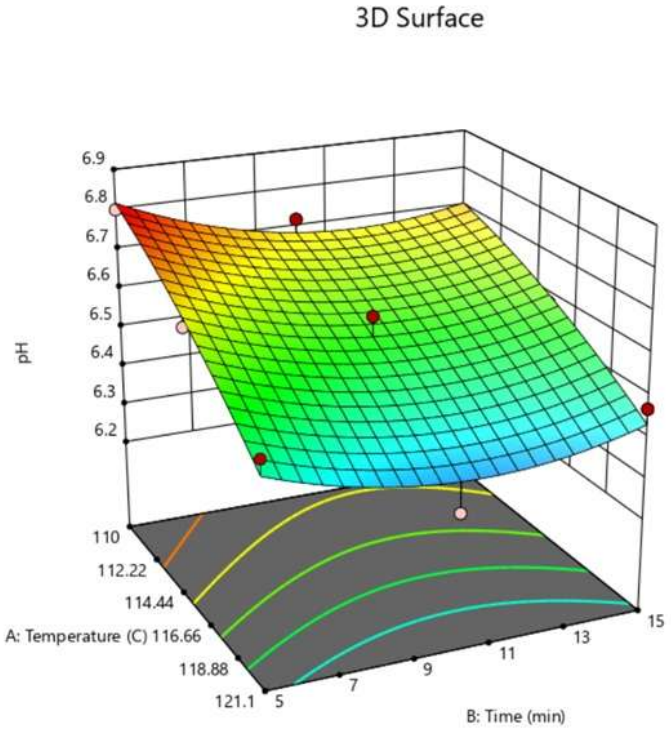


Fig 5. Effect of treatment time and temperature on Microbial load

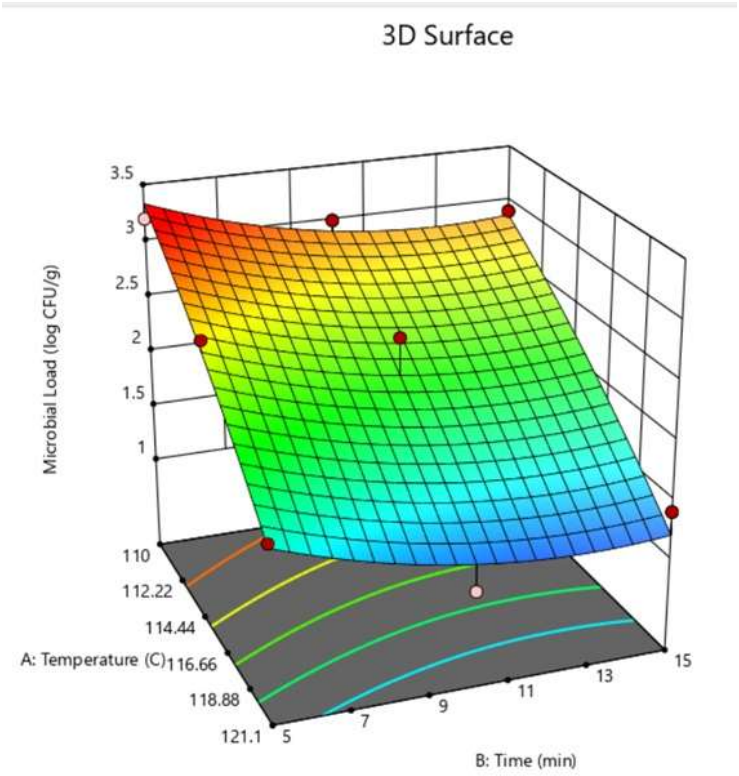


Fig 6. Desirability chart for Optimized RTE biryani

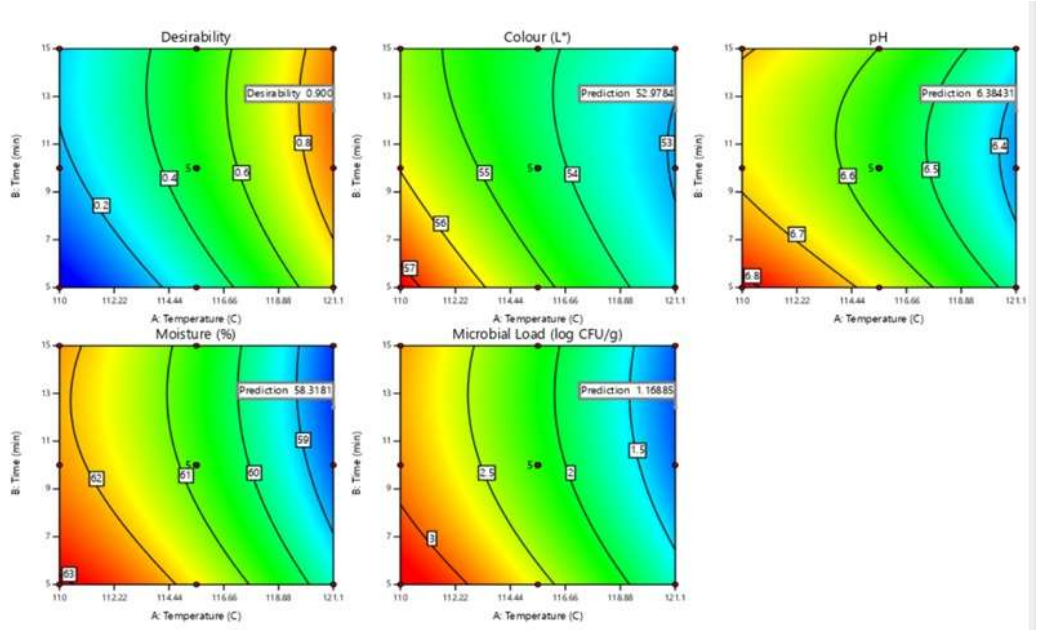
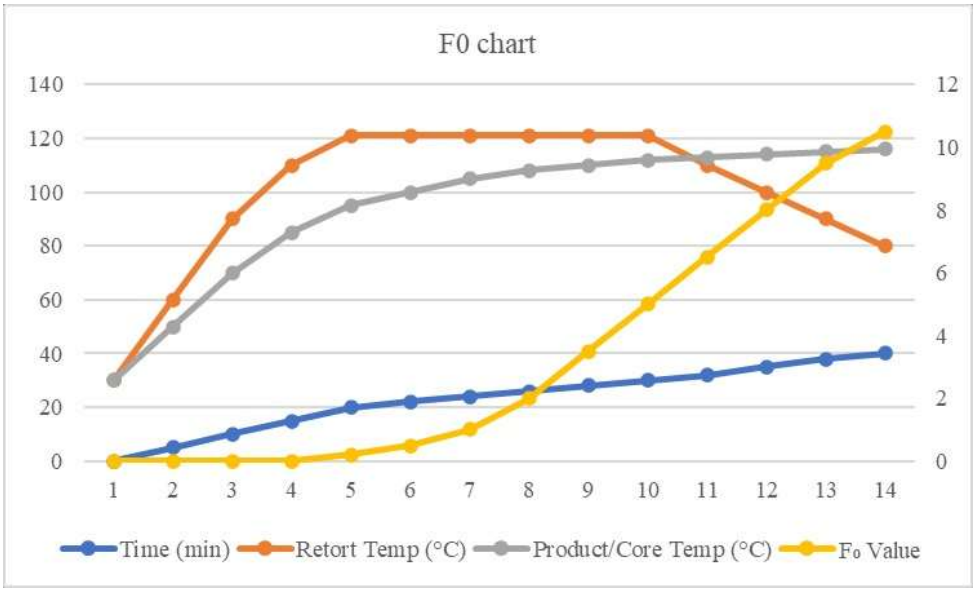


Fig 7. Time - Temperature Curve for RTE Seeraga samba biryani



Tables:

Table 1. Sensory evaluation scores of retort-processed of RTE Seeraga Samba biryani

Parameter	Method I (Masala-integrated pre-cooking)			Method II (Direct raw filling)			Method III (Aqueous pre-cooking)		
	0th day	30th day	60th day	0th day	30th day	60th day	0th day	30th day	60th day
Appearance	8.25 ± 0.58	8.10 ± 0.62	7.85 ± 0.70	7.60 ± 0.65	7.35 ± 0.70	7.05 ± 0.75	7.20 ± 0.68	6.95 ± 0.72	6.70 ± 0.80
Aroma	7.88 ± 0.89	7.65 ± 0.92	7.32 ± 0.95	7.20 ± 0.90	6.95 ± 0.94	6.70 ± 0.98	6.85 ± 0.95	6.60 ± 0.98	6.35 ± 1.02

Taste	8.06 ± 0.85	7.84 ± 0.88	7.55 ± 0.91	7.30 ± 0.88	7.05 ± 0.92	6.80 ± 0.96	6.95 ± 0.90	6.70 ± 0.94	6.45 ± 0.98
Texture	8.19 ± 0.66	7.98 ± 0.71	7.68 ± 0.78	7.40 ± 0.72	7.10 ± 0.76	6.85 ± 0.80	6.90 ± 0.75	6.65 ± 0.79	6.40 ± 0.84
Overall Acceptability	8.28 ± 0.58	8.02 ± 0.60	7.76 ± 0.65	7.50 ± 0.62	7.20 ± 0.66	6.95 ± 0.70	7.05 ± 0.65	6.80 ± 0.69	6.55 ± 0.72

Table 2. Factors and responses using Central Composite Design (CCD)

Run	Temperature	Time (Min)	Colour (L*)	pH	Moisture (%)	Microbial Load (log CFU/g)
1	110	10	56.3	6.72	62.5	3
2	121.1	15	53.6	6.47	58.6	1.4
3	115.55	10	54.2	6.61	61.2	2.5
4	115.55	5	54.9	6.65	61.8	2.7
5	115.55	10	54.2	6.5	61.2	2.5
6	110	5	57.2	6.8	62.9	3.2
7	121.1	10	52.1	6.3	58.1	1

8	110	15	55.2	6.69	62.1	2.9
9	121.1	5	53.9	6.51	59.7	1.7
10	115.55	10	55.3	6.61	60.2	2
11	115.55	15	54	6.58	60.5	1.9
12	115.55	10	54.2	6.51	60.2	1.5
13	115.55	10	54.2	6.61	61.2	2.5

Table 3. Analysis of variance (ANOVA) for different treatment time and treatment temperature on Moisture, Colour, pH, Microbial load.

Source	**Moisture**		***L (Colour)**		**pH**		**Microbial load**	
	F-value	**p-value**	**F-value**	**p-value**	**F-value**	**p-value**	**F-value**	**p-value**
Model - Quadratic	19.75	0.0005	9.90	0.0045	9.53	0.0050	6.48	0.0147
A - Temperature	88.22	<0.0001	40.38	0.0004	40.54	0.0004	29.34	0.0010
B - Time	7.33	0.0303	4.99	0.0606	2.27	0.1757	2.30	0.1732
AB	0.0967	0.7649	2.11	0.1893	0.3445	0.5756	0.0000	1.0000

A^2	2.05	0.1954	0.1576	0.7032	0.6440	0.4486	0.2081	0.6621
B^2	2.24	0.1782	1.23	0.3046	4.51	0.0713	0.7512	0.4148
Lack of Fit	0.4772	0.7152	1.96	0.2618	1.17	0.4265	0.3238	0.8094

Table 4. Changes in physicochemical, nutritional and microbial quality of retort-processed Seeraga Samba biryani during storage

Parameter	**Unprocessed**	**Processed (0^{th} day)**	**30^{th} day**	**60^{th} day**
		Proximate Composition		
Moisture (%)	64.82 ± 0.52	63.42 ± 1.12	63.69 ± 0.74	63.89 ± 0.68
Protein (%)	6.02 ± 0.18	5.84 ± 0.21	5.78 ± 0.12	5.69 ± 0.04
Fat (%)	6.48 ± 0.27	6.38 ± 0.34	6.22 ± 0.18	6.27 ± 0.14
Ash (%)	1.71 ± 0.10	1.62 ± 0.08	1.61 ± 0.05	1.60 ± 0.05
Carbohydrate (%)	20.97 ± 0.88	22.94 ± 0.97	21.43 ± 0.63	20.98 ± 0.71
Energy (kcal/100 g)	182.6 ± 3.9	176.3 ± 4.6	177.8 ± 2.4	178.9 ± 1.5
		Mineral Composition		
Sodium (mg/100 g)	425.8 ± 11.2	412.6 ± 9.8	409.4 ± 8.7	404.9 ± 7.6
Potassium (mg/100 g)	156.9 ± 7.1	148.3 ± 6.4	146.8 ± 5.9	144.6 ± 5.3
Calcium (mg/100 g)	35.1 ± 2.1	32.7 ± 1.9	32.2 ± 1.8	31.5 ± 1.6
Magnesium (mg/100 g)	28.2 ± 1.5	26.4 ± 1.3	26.1 ± 1.2	25.6 ± 1.1

Iron (mg/100 g)	3.45 ± 0.20	3.21 ± 0.18	3.18 ± 0.17	3.12 ± 0.15
Zinc (mg/100 g)	1.58 ± 0.08	1.46 ± 0.07	1.44 ± 0.06	1.41 ± 0.05
Physicochemical Properties				
L* (Lightness)	58.42 ± 0.84	56.73 ± 0.62	56.28 ± 0.74	55.96 ± 0.69
pH	6.42 ± 0.05	6.35 ± 0.04	6.31 ± 0.03	6.28 ± 0.02
Microbial Quality				
TPC (CFU/g)	2.95×10^3	$<1 \times 10^3$	$<1 \times 10^3$	1×10^3
Coliforms (CFU/g)	1.4×10^2	$<1 \times 10^2$	$<1 \times 10^2$	1×10^2
Yeast & Moulds (CFU/g)	3.1×10^2	$<1 \times 10^2$	$<1 \times 10^2$	1×10^2