IMPROVEMENT OF KOMBUCHA FERMENTATION USING BOX BEHNKEN EXPERIMENTAL DESIGN

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INTRODUCTION

High beverage consumption worldwide has opened the opportunity for the improvement of different traditional drinks as part of the functional food concept. In recent years, the development of kombucha fermentation is part of the scientific and industrial focus in the field of functional drink. Kombucha optimization has become very important because of the large importance of the definition of chemical composition from health-improving, economic, and industrial points of view.

MATERIALS AND METHODS:

Fermentation was performed using the local tea fungus culture with at least five yeast strains (*Saccharomycodes ludwigii, S. cerevisiae, S. bisporus, Torulopsis sp., and Zygosaccharomyces sp*.) and two bacterial strains of *Acetobacter genera*. Three operating parameters of kombucha fermentation (specific surface area of the vessel (0.06, 0.15 and 0.3 cm-1), inoculum (2.5, 5 or 10% (v/v)), and initial tea concentration (0.15, 0.3, or 0.45% (w/v)) were independent factors in the selected Box-Behnken experimental design, while the output variables were the pH values and the titratable acidity of kombucha.

OBJECTIVE: In this study, the influence of the specific surface area of the vessel, inoculum size, initial tea concentration, and fermentation time on the efficiency of kombucha fermentation was examined. The focus of this study is the optimization and standardization of kombucha fermentation conditions using Box- Behnken experimental design

RESULTS AND DISCUSSION

Observing fermentation processes through Box-Behnken experimental design, a higher concentration of total acids was recorded in the cultivation medium with a larger inoculum size, and the time required to obtain the beverage of optimal acidity was shorter. Both three-dimensional plots show the interaction of two tested operating parameters on the pH value and TA (Figure 1). It can be observed that minimal initial tea concentration (0.15%) provided sufficient nitrogen compounds and mineral elements that are necessary for kombucha fermentation under stationary fermentation conditions. Based on unattainable adequate values of titratable acidity and pH value that despite the high C and N sources contents, the fermentation process with high tea concentration can be slower and therefore economically less acceptable. The accuracy of the ANN model could be visually assessed by the dispersion of points from the diagonal line in the graphics presented in Figure 2.

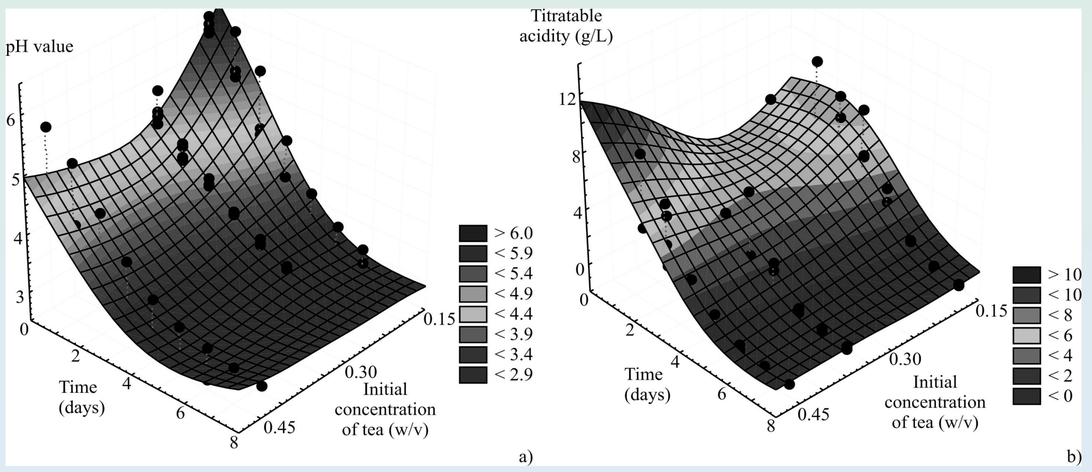

Figure 1. The influence of time (days) and initial concentration of tea on the pH value and titratable acidity (g/L) (3d surfaces - predicted values, black dots - experimental values)

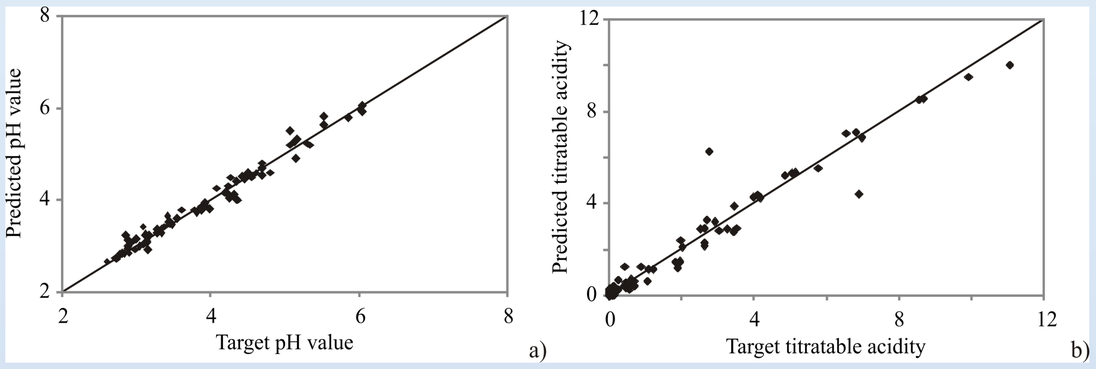

Figure 2. Comparison between experimental and calculated pH values and titratable acidity (g/L)

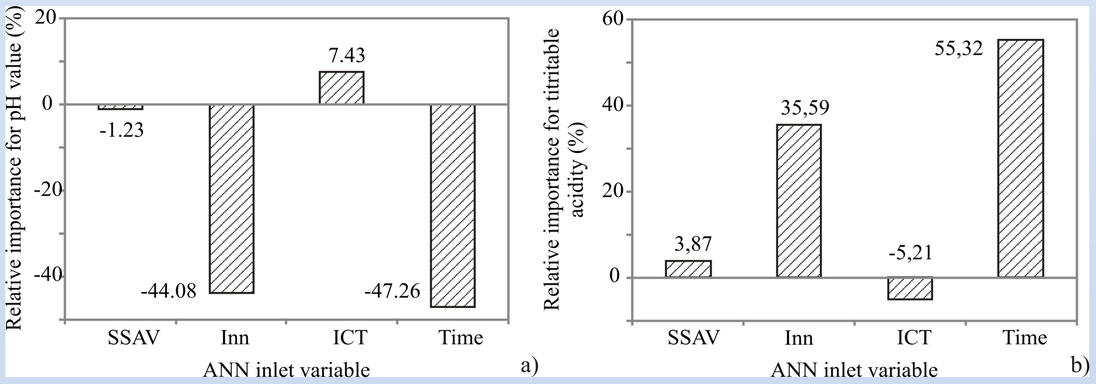

Figure 3. The relative importance of operating parameters on pH value and titratable acidity determined using Yoon interpretation method

The obtained ANN for the prediction of the pH value and titratable acidity (Figure 3) showed good prediction capabilities (the r^2 values during training cycle for output variables were 0.990 and 0.994, respectively). Predictive ANN modelling demonstrated to be effective and reliable in establishing optimum kombucha fermentation process using selected operating parameters.

Box Benhken's experimental design was applied and predictive ANN modelling was developed to establish the optimum kombucha fermentation process to achieve the effective fermentation of the kombucha beverage.

Acknowledgments: The financial support of the Ministry of Education, Science, and Technological Development of the Republic of Serbia (contract no. 451-03-9/2021-14/200134) is gratefully acknowledged.