ABSTRACT

Orange juice is the most favoured citrus juice by consumers for its distinctive flavour and aroma. It is a rich source of vitamins, bioactive compounds, minerals, and various sensory compounds. Atmospheric cold plasma (ACP) is a promising non-thermal technology with a wide range of potential applications in food processing and preservation. Therefore, current research comprises a total of four objectives, beginning with the standardization of process conditions for batch-type ACP-assisted processing of orange (cv. Wakro and Malta) juice using response surface methodology (RSM). Further, it was examined with different kinetics models how the ACP treatment impacted the inactivation of *Escherichia coli* (*E. coli*) and pectin methylesterase (PME) in orange (cv. Wakro) juice. Then, a storage study of ACP-treated and untreated orange (cv. Wakro) juice was conducted over time when stored at different temperatures. Finally, a lab-scale system with a continuous flow setup was developed and tested with orange (cv. Wakro) juice.

This study investigated the effect of ACP treatment on Wakro orange juice and optimized the process conditions within the range of 16–24 kV voltage, 3–5 mm juice depth, and 1–3 min treatment time. Furthermore, ACP to another cultivar of orange like Malta juice was also examined under varying conditions, including juice depth (2–4 mm), voltage (16–20 kV), and treatment time (2–10 min). The optimum conditions for Wakro juice, determined using RSM, were 19 kV voltage, 4.6 mm juice depth, and 2.6 min treatment time, resulting an ascorbic acid (AA) content of 28.08 mg/100 mL, residual activity (RA) of PME of 43.80%, bioaccessibility index (BI) of AA of 77.25%, DPPH radical scavenging activity of 53.92%, and total phenolic content (TPC) of 50.80 mg GAE/100 mL. For Malta juice, the optimum conditions were 20 kV, 3.3 mm depth, and 2 min, with AA, RA of PME, and BI values of 31.54 mg/100 mL, 61.33%, and 79.10%, respectively. The study revealed that juice depth and treatment time significantly affected AA, TPC, and BI in Wakro orange juice, while treatment time and voltage had the greatest impact on RA of PME and DPPH radical scavenging activity. The results highlighted the PME inactivation was more pronounced in Wakro juice than in Malta juice. The kinetics modeling of PME enzyme and *E. coli* inactivation of ACP-assisted processing of orange (cv. Wakro) juice at different voltages (16, 20, and 24 kV) and treatment times (0.5, 1, 1.5, 2, 2.5, and 3 min) was studied. Kinetic models showed that PME inactivation followed an n^{th}-order model, with highest fitting accuracy ($R^2 > 0.98$; RMSE < 0.012; $\Delta_i \leq 2$), while *E. coli* inactivation followed a Weibull model ($R^2 > 0.85$; RMSE < 0.2841; $\Delta_i = 0$ (for 16 and 24 kV) $\Delta_i = 8.67$ (for 20 kV)). The storage study of ACP-treated (juice depth: 4.6 mm, voltage: 19 kV, and treatment time: 2.6 min) and untreated Wakro orange juice revealed that while minor changes in physicochemical parameters like pH, total soluble solids (TSS), and titratable acidity (TA) occurred over 9 days for untreated juice and 30 days for treated juice, AA and DPPH activity decreased with storage time at 10 ºC and 25 ºC. TPC remained higher in ACP-treated juice. The ΔE was increased during the storage period. The total viable counts (TVC) and yeast and mold counts (YMC) in the juice sample significantly increased over the storage period of 9 days (for untreated) and 30 days (for ACP-treated). At 10°C, ACP-treated juice exhibited slower microbial growth (TVC and YMC) compared to the untreated juice. Continuous ACP processing of Wakro orange juice at different voltages (20–28 kV) and flow rates (50–100 mL/min) showed a slight increase in electrical conductivity (EC) and insignificant ($p > 0.05$) effect in TSS, pH, and TA. The ΔE was also slightly changed with increased residence time and voltage. The AA, DPPH radical scavenging activity, and RA of PME significantly reduced. The kinetics of AA and DPPH degradation followed the first-order model ($R^2 > 0.93$ and RMSE < 0.03), providing insights into the behaviour of these components during continuous ACP treatment.

Keywords: Orange juice, Cold plasma, Optimization, Enzyme inactivation, Microbial inactivation, Kinetics modelling, Storage study *their support and assistance. I extend my gratitude to all the research scholars, Department of Food Engineering and Technology for their assistance and insightful discussion.*

I thank all who have helped me directly or indirectly, whose names have not been mentioned.

Lastly, I express gratitude to my family members for their love, unwavering support, motivation, and contribution throughout my Ph. D. journey. I owe a debt to my parents for their blessings, unconditional love, and sacrifices.

Above all, I am grateful to 'The Almighty' who showered blessings on me that enabled me to complete my Ph. D. thesis.

Sahijul Islam

Date:

Place: Tezpur