Mild thermal processing of pineapple juice using ohmic heating technology

GN ASHITHA and MV PRINCE

Department of Processing and Food Engineering Kelappaji College of Agricultural Engineering and Technology (KAU) Tavanur 679573 Kerala, India

Email for correspondence: ashithagn@gmail.com

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ABSTRACT

Ohmic heating is one of the novel thermal technologies widely used for the inactivation of microorganisms and enzymes. The optimization of ohmic heating process parameters such as voltage, temperature and holding time to inactivate microorganisms and the effects on the physico-chemical qualities of pineapple juice were studied. The pineapple juice was processed by ohmic heating at different voltage gradients (10-15 V/cm), temperatures (50-60°C) and holding times (1-5 min). A higher reduction of microorganisms was observed with increased voltage gradient, treatment temperature and time. The highest reduction in bacteria, yeast and mold population was obtained for treatment with voltage gradient of 15 V/cm, temperature of 60°C and holding time of 3 min. Ascorbic acid showed a significant reduction with the increase in temperature and treatment time. A voltage gradient of 14.74 V/cm, process temperature of 56.45°C and holding time of 2.98 min was found to be the optimum ohmic heating condition to achieve a predicted bacterial log reduction of 3.75 and yeast and mold log reduction of 2.85.

Keywords: Ohmic heating; voltage gradient; treatment temperature; holding time; ascorbic acid; log reduction

INTRODUCTION

Considerable attention has been focused on vegetable and fruit juices recently due to the presence of health-promoting compounds in the juices, ease of packaging and transport. In addition to this juices play a significant role in the daily nutritional supplement and offer essential nutrients. Pineapple (Ananas cosmosu) is one among the fruit crops in India and its juice is famous for its sweet taste and aroma profile. The pineapple juice is rich in antioxidant compounds, mineral compounds and enzymes like bromelain (Larson 1988, Hale et al 2005). Pineapple juice has some proven health benefits such as reduction in cardiovascular and some chronic and degenerative diseases due to the inherent antioxidant content (Dragsted 2003). The high water content makes the fruit susceptible to microbiological and physico-chemical degradation therefore pineapple is processed into a wide variety of products.

However processing may affect the organoleptic and nutritional values of processed

pineapple. The major problems encountered are loss of vitamin C during thermal treatments (Uckiah et al 2009, Achinewhu and Hart 1994, Akinyele et al 1990) and non-enzymatic browning, the Maillard reaction, during storage. Therefore novel technologies that will have a reduced impact on the nutritional content and overall food qualities need to be adopted in order to protect the nutritional and organoleptic qualities and to produce safe fruit juices with prolonged shelf-life.

Ohmic heating technology can be applied effectively in viscous liquids and mixtures containing particulate food products as a continuous in-line heating method for cooking and sterilization. The ohmic heating is a process in which electrical current is passed through the food material and heat is generated due to the electrical resistance of the material (Icier 2003). Instant heat generation inside the food and the quantity of heat generated are directly related to the voltage gradient in the field, current induced and the electrical conductivity. Conventional thermal treatments significantly damage product quality due to slow convection and conduction heat transfer. However

during ohmic process entire mass of the food material heats rapidly and volumetrically. So the product maintains higher quality than its thermally processed counterpart (Icier 2003).

The principal mechanisms of microbial inactivation in ohmic heating are due to thermal effects. Recent research indicates that ohmic heating may cause mild, non-thermal cellular damage due to the presence of the electric field (Cho et al 1999, Pereira and Vicente 2010, Sun et al 2008). The most widely accepted mechanism is that of severe electroporation (Park et al 2003). Electroporation is the formation of holes in a cell membrane due to individual ion pressure which causes changes in the permeability of the cell membrane due to the varying electric field (Weaver and Chizmadzhev 1996).

In this study the effect of process parameters of the ohmic heating of pineapple juice such as voltage gradient, temperature and holding time on the inactivation of microorganism and other physicochemical qualities (pH, TSS and ascorbic acid content) were analyzed and optimized using response surface methodology.

MATERIAL and METHODS

Fruit juice preparation

Kew variety of pineapple was purchased from the local market; was visually inspected for external bruises and blemishes and the good fruits were selected. Fruits were washed in running water and the crown and outer skin of the fruit was removed with a stainless steel knife. The pineapple was then sliced, cored, cut into small pieces and squeezed in a centrifugal extractor for extracting juice. The juice was filtered using muslin cloth to remove the coarse tissues and stored in a sterile bottle at 4°C for further experiments.

Ohmic heating

All the experiments were conducted in a laboratory batch type ohmic heating system designed and developed in the Department of Processing and Food Engineering, Kerala Agricultural University, Tavanur, Kerala. The setup composed of a feed tank, ohmic heating chamber, variable transformer, voltampere meter and temperature measuring system. A cylindrical ohmic heating chamber with inner diameter of 75 and 65 mm and made up of Teflon material served as an excellent insulator capable of withstanding

high treatment temperatures. The electrodes made of food grade and non-corrosive SS 304 material of 2 mm thickness were fastened to both sides of the cylindrical chamber with a spacing of 150 mm between them. The thermocouple was inserted in a separate port with a diameter of 0.5 mm at the geometrical center of the chamber. The ohmic heating chamber was filled with pineapple juice supplied from the feed tank. The predetermined levels of voltage gradient were set and temperature of the juice was allowed to rise to a required limit. The required temperature and the required holding time were maintained by manually adjusting power supply. The juice samples were collected and stored in sterile amber colour bottles at 4°C.

Experimental design of ohmic heating process

Experiments were framed using Box-Bekhen design of response surface methodology in the Design expert software version 6.0.8 to optimize the voltage gradient, temperature and residence time combination required for the effective mild thermal treatment and thus to study the effect of treatment on the microbial and biochemical properties of the fruit juices.

A three level factorial design was used (3³) with independent variables as voltage gradient (10, 12.50 and 15 V/cm), temperature (50, 55 and 60°C) and holding time (1, 3 and 5 min). The three factors were coded with three levels (-1, 0, +1). The analysis of variance was obtained for each response to find out the significant differences.

The response surface analysis used a generalized second-order polynomial model as per equation below:

$$\begin{aligned} \mathbf{Y} &= \mathbf{a}_0 + \, \mathbf{a}_1 \mathbf{A} + \mathbf{a}_2 \mathbf{B} + \mathbf{a}_3 \mathbf{C} + \mathbf{a}_{12} \mathbf{A} \mathbf{B} + \mathbf{a}_{13} \mathbf{A} \mathbf{C} + \mathbf{a}_{23} \mathbf{B} \mathbf{C} + \mathbf{a}_{11} \mathbf{A}^2 + \\ \mathbf{a}_{22} \mathbf{B}^2 + \, \mathbf{a}_{33} \mathbf{C}^2 \end{aligned}$$

where Y= Response variable and the independent variables denoted as A (voltage gradient), B (processing temperature) and C (holding temperature)

The adequacies of the models were determined using model analysis, lack-of-fit test and R² (coefficient of determination) analysis as outlined by Lee et al (2000). From the analyzed data response surface contour plots were generated for each response (Myers and Montgomery 2002). Optimization of the process variables was carried out with the help of desirability function. The responses were either minimized or

maximized while independent factors were kept within the experimental range. The goals were combined into an overall composite function, d(x) called the desirability function (Myers and Montgomery 2002).

Enumeration of bacteria, yeast and mold

The treated samples were observed for the growth of total plate count (TPC) and yeast and mold count (YMC) by the standard plate count method. The commonly used media, nutrient agar and chloramphenicol yeast glucose agar were used for enumeration of bacteria and yeast and mold respectively. The dilutions of 10^{-2} and 10^{-3} of the sample with 3 replications were pour-plated in respective media and then bacterial and yeast plates were incubated separately at 30° C for 48 h and 25° C for 48 h respectively.

Biochemical properties

Total soluble solids (TSS) were determined by using digital hand refractometer (Make Erma) and expressed in terms of degree Brix. The pH value of juice was measured using a digital pH meter (m/s SYSTRONICS). Ascorbic acid was determined by a visual titration method with the dye solution 2, 6-dichlorophenol indophenol (DCPIP) (Sadasivam and Manickam 1992).

RESULTS and DISCUSSION

Effect of pH on ohmic heating

The study observed a significant effect of ohmic heating process variables such as voltage gradient and holding time on the pH value of pineapple juice (P <0.05). The second order polynomial model for the pH of pineapple juice is depicted in equation below:

pH=3.99+0.38A+0.014B 6 0.19C+0.023AB 6 0.032AC 6 0.025BC 6 0.18A²+0.042B²+0.076C²

A slight reduction in pH value of the pineapple juice was observed for all ohmic heated samples. The 3D surface plot for variation in pH value (Fig 1) illustrates that higher the voltage gradient, lower is the reduction in pH value. Similar findings were reported by Makroo et al (2017) and Boldaji et al (2014) in watermelon juice and tomato paste processing respectively. The increase in holding time showed an increase in pH reduction. The pH value of the ohmic heated samples ranged between 3.38 and 4.47. The maximum percentage reduction was observed in

samples treated with 10 V/cm at 55°C for 5 min and minimum in samples treated with 15 V/cm at 55°C for 1 min. The reduction in pH might be due to the higher residence time required at lower voltage gradients and hydrolysis of pineapple juice in the presence of electric fields (Assiry et al 2010).

Effect of total soluble solids on ohmic heating

An increase in TSS of pineapple juice was observed after the ohmic heating process. A model for prediction of the ohmic heated pineapple juice is depicted beow:

TSS = 11.31 + 0.42A + 0.059B + 0.27C 6 2.5AB 6 0.048AC 6 0.025BC 6 0.31A² 6 0.094B² 6 0.078C²

The study showed a significant effect of voltage gradient and holding time on TSS of pineapple juice (p <0.05). The 3D surface plot of response surface analysis is shown in Fig 2. TSS value increased with an increase in voltage gradient and holding time. The similar results were observed during the studies conducted in guava juice, sugar cane juice and banana purees (Chakraborty and Athmaselvi 2014, Abhilasha and Pal 2018, Poojitha and Athmaselvi 2018). The TSS that increases due to the loss of water during continuous heating leads to increase in solute concentration (Purvis 1983).

Effect of ohmic heating on ascobic acid

The ascorbic acid value in pineapple juice was found to be reduced in all ohmic heated samples. The voltage gradient, temperature and holding time were found to possess a significant effect on the ascorbic acid content of pineapple juice (P < 0.05). The prediction model obtained for ascorbic acid content is stated the follwoing equation:

Ascorbic acid= 35 6 0.046A 6 0.37B 6 0.26C + 0.017AB + 0.035AC 6 0.082BC 6 0.015A² 6 0.11B² 6 0.024C²

The ascorbic acid content reported a maximum per cent reduction in treatment with voltage gradient of 12.5 V/cm, temperature of 60° C and holding time of 5 min and a minimum reduction was observed in the treatment with a voltage gradient of 10 V/cm, temperature of 55° C and holding time of 1 min (Fig 3).

Chakraborty and Athmaselvi (2014) also observed a higher reduction of ascorbic acid with an increase in the voltage gradient for pineapple juice. Temperature can be considered as one of the important

parameters due to the inherent property of ascorbic acid degradation during elevated temperatures.

Some researchers have suggested that presence of electric field does not change the ascorbic acid content (Castro et al 2004, Assiry et al 2006). Assiry et al (2003) found that the degradation of ascorbic acid was due to the oxidation of ascorbic acid molecules by the oxygen species developed in the electrolysis of water during ohmic heating. The fruit sugar content also induces degradation of vitamin C. It was also reported that solid content of banana pulp and applied voltage significantly caused breakdown of ascorbic acid (Lima and Sastry 1999).

The models for all responses such as pH, TSS and ascorbic acid were analyzed which depicted that all models were highly significant (p <0.05) with higher values of R^2 and Adj R^2 . The insignificant lack of fit represents the good fitness of the model for the responses.

Effect of bacterial, yeast and mold population

The ohmic-heated pineapple juices were evaluated for the presence of total plate count, yeast and mold population and the results are presented in Table 1. It was found that a 4.04 log 10 cfu/ml reduction of bacterial population and 3.04 log cfu/ml reduction of yeast and mold population were obtained for a voltage gradient of 15 V/cm, temperature of 60°C and holding time of 3 min. However the minimum reduction was obtained for the treatment with voltage gradient of 10 V/cm, temperature of 55°C and holding time of 1 min.

The 3D surface plot for total plate count and yeast and mold population generated by the response surface methodology (Figs 4 and 5) showed that with increase in temperature and holding time a reduction in microbial count was observed. It was found that a direct relationship between the temperatures attained during ohmic heating and the microbial survival existed and higher the temperature more the microbial lethality (Athanasiadis et al 2004, Barreto et al 2003, Zimmerman et al 2008, Kumar et al 2012).

Similarly the increase in voltage gradient also showed higher reduction in microbial count. It was found that the combined effect of processing time and applied voltage had a significant effect on the microbial count of the sample (Fig 5a and 5b). As these two parameters

increased, the microbial count of the sample was found to decrease. This may be due to the fact that as the processing time and voltage gradient increased, a higher process temperature was achieved which might have resulted in inactivation of microbial population (Piette et al 2004, Shirsat et al 2004).

Gomathy et al (2015) reported that ohmic heating of papaya pulp at a voltage gradient of 13.33 V/cm with a holding time of 2 min resulted into minimum bacterial and yeast load of 1.00 log cfu/ml after 30 days of storage period which was much below the recommended level of 50 cfu/ml (https://fssai.gov.in/cms/food-safety-and-standards-rules-2011.php).

The predicted model for total plate count and yeast and mould count are depicted in following equations:

Total plate count= 3.51 + 0.63A + 0.53B + 0.31C + 0.17AB + 0.26AC + 0.19BC 6 0.47A² 6 0.19B² 6 0.36C²

Yeast and mold count= 2.58 + 0.50A + 0.46B + 0.23C + 0.15B + 0.23C + 0.15AB + 0.15AC + 0.12BC 6 0.30A² 6 0.21B² 6 0.24C²

The ANOVA of the total plate count and yeast and mold count illustrate that all independent variables A, B and C and their interaction terms and quadratic terms were significant. The model exhibits a good fit with the responses, total plate count and yeast and mold count were found highly significant with slight variations among the mean (R^2 = 0.98 for TPC and R^2 = 0.97 for YMC). There was a low variation between Adj R^2 and R^2 proving a higher correlation between predicted and experimental values. The non-significant value (P >0.05) of lack of fit implies that model is successful in predicting the response.

Optimization and experimental validation

The ohmic heating process parameters were optimized to predict the optimum ohmic heating treatment within the specified domain which provided the desired response goal. For optimization of the process parameters during ohmic heating the responses were maximized or minimized to get the desired outcome. The response of log reduction, ascorbic acid and pH value was maximized and all independent values were kept in range. The optimum process conditions for ohmic heating and other best solutions provided higher desirability.

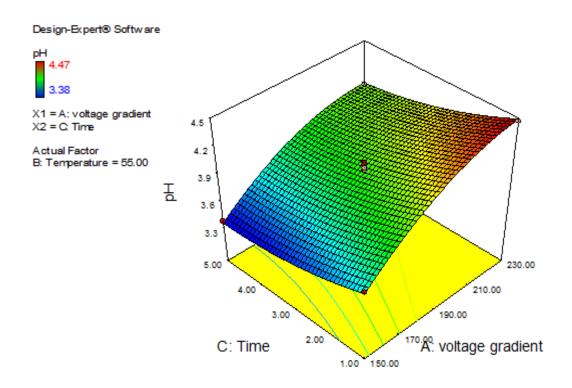


Fig 1. Effect of ohmic heating on pH of pineapple juice

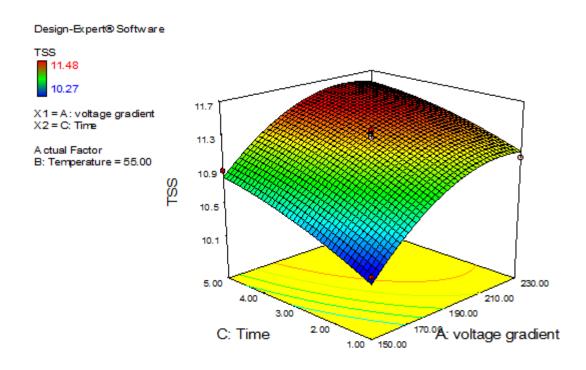
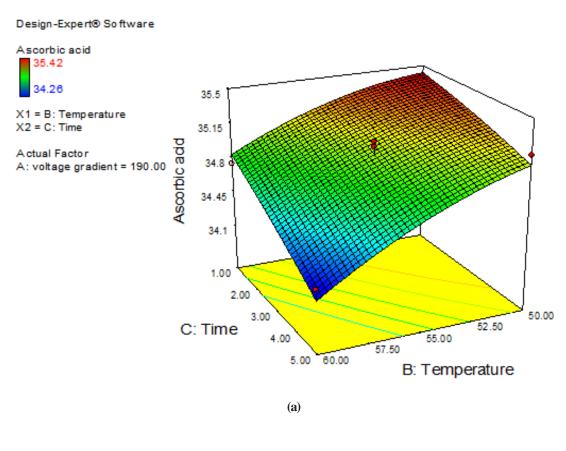


Fig 2. Effect of ohmic heating on TSS of pineapple juice



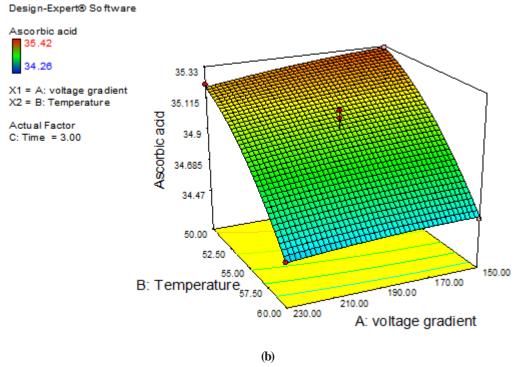


Fig 3. Effect of ohmic heating on ascorbic acid of pineapple juice

Design-Expert® Software Total plate count 4.12 1.89 4.3 X1 = A: voltage gradient X2 = B: Temperature 3.675 Total plate count Actual Factor C: Time = 3.00 3.05 2.425 1.8 60.00 230.00 57.50 210.00 190.00 ^{170.0}Å: voltage gradient B: Temperature 150.00 50.00 (a) Design-Expert® Software Total plate count 4.1 X1 = B: Temperature X2 = C: Time 3.65 Actual Factor A: voltage gradient = 190.000 0 10 3.2 2.75 2.3 5.00 60.00 57.50 55.00 2.00 C: Time B: Temperature 50.00 1.00

Fig 4. Effect of ohmic heating on total plate count of pineapple juice

(b)

Design-Expert® Software yeast and mould 1.35 3 X1 = B: Temperature X2 = C. Time 2.625 Actual Factor
A: v oltage gradient = 190.0%
u pue tse ex 2.25 1.875 1.5 5.00 60.00 4.00 57.50 3.00 55.00 C: Time B: Temperature 1.00 50.00 (a)

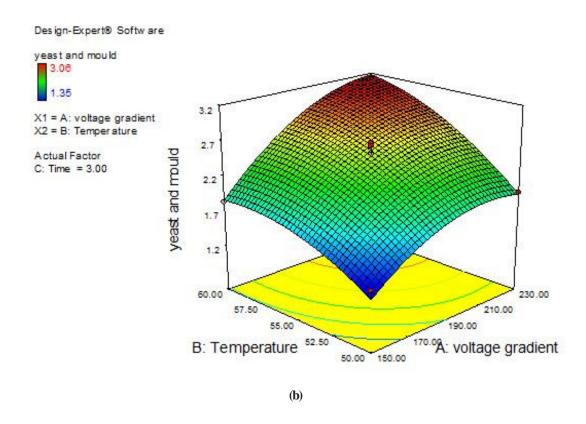


Fig 5. Effect of ohmic heating on yeast and mold count of pineapple juice

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Table I	Resnonse	SHITTACE	designs	W/1fh	inde	nendent	variables	ana	studied resp	nnse
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Treatment	Voltage gradient (V/cm)	Temperature ($^{\circ}$ C)	Holding time (min)	pН	TSS (°Brix)	Bacterial log reduction log (cfu/ml)	Yeast and mold log reduction log (cfu/ml)	Ascorbic acid (mg/100 ml)
T_1	15	50	3	4.2	11.37	2.79	1.98	28.21
T_2	12.5	55	3	3.98	11.28	3.49	2.52	28.08
T_3^2	12.5	60	5	3.89	11.39	4.12	2.94	27.26
T_4^3	12.5	55	3	3.96	11.32	3.64	2.53	27.96
T_5^{\dagger}	12.5	55	3	4.02	11.25	3.42	2.68	28.13
T_6	15	60	3	4.31	11.4	4.04	3.04	27.48
T_7°	12.5	55	3	4.04	11.36	3.58	2.54	27.87
$T_8^{'}$	12.5	50	5	3.95	11.38	2.48	1.65	28.16
T_9°	10	50	3	4.2	10.85	1.98	1.38	28.32
T ₁₀	15	55	1	4.42	11.05	2.75	2.14	28.24
T ₁₁	12.5	55	3	3.96	11.35	3.55	2.64	27.98
T ₁₂	10	55	1	3.69	10.62	1.89	1.35	28.42
T ₁₃	12.5	60	1	4.32	10.95	3.13	2.36	27.75
T ₁₄	10	60	3	3.47	10.83	2.63	1.84	27.52
T ₁₅	10	55	5	3.38	10.89	2.14	1.62	27.62
T ₁₆	15	55	5	4.03	11.42	3.98	3.02	27.58
T ₁₇	12.5	50	1	4.28	10.93	2.37	1.54	28.32

The optimum conditions for ohmic heating process were found at voltage gradient of 14.74 V/cm, treatment temperature of 56.45°C and holding time of 2.98 min with predicted responses such as bacterial log reduction of 3.76, yeast and mold reduction of 2.83 and ascorbic acid content of 27.85 mg/100 ml etc.

CONCLUSION

The present study portraits the effect of ohmic heating of pineapple juice with the objective of optimizing the process conditions. The process parameters such as voltage gradient, process temperature and holding time were studied for their influence on bacterial, yeast and mold inactivation.

A bacterial log reduction of 4.04 log cfu/ml and yeast and mold reduction of 3.06 log cfu/ml was observed during the treatment with voltage gradient of 15 V/cm, process temperature of 60°C and holding time of 3min.

The ascorbic acid showed a gradual degradation during the process. The ohmic heating reduced the bacterial, yeast and mold population in pineapple juice and the reduction in ascorbic acid content was minimum compared to other reported studies. It may be concluded that ohmic heating process at the optimized process conditions as stated could be

an alternative mild heat treatment process that retains the quality parameters of the pineapple juice.

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