Study on Effect of Hot Water Blanching on Vital Parameters of Allahabad Safeda Guava (Psidiumguajava)

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Abstract: Effect of hot water blanching on vital parameters of Allahabad safeda guava was studied at the temperature 80^0C for 0 minutes, 4minutes, 8 minutes, 12 minutes and 16 minutes. Physico-chemical properties (moisture, total solids (T.S.), total soluble solids (T.S.S.), acidity, vitamin C (ascorbic acid), reducing sugar and total phenol content (T.P.C.) and peroxidase enzyme inactivation and changes in colour were studied. In physico-chemical properties percent acidity, ascorbic acid, reducing sugar were decreasedsignificantly (at 5% level of significance) and moisture content, T.S., T.S.S., T.P.C were increasedsignificantly (at 5% level of significance) in comparison with the raw guava sample i.e. control sample. Peroxidase inactivation followed a first order Arrhenius model, whererate of the reaction at 80^0C was 0.26 x 10^{-3} s^{-1}. Good agreement was found between estimated and experimental data (R^2 = 0.807). Colour was quantified using the L, a, b in X-rite color lab system and based on these readings, Total colour difference (TCD) was calculated.

Key words: Blanching, Color, Kinetic Modeling, Peroxidase Inactivation. Allahabad Safeda Guava

Nomenclature:

L*CIE colour space co-ordinate: degree of lightness

a*CIE colour space co-ordinate: degree of greenness/redness

b*CIE colour space co-ordinate: degree of blueness/yellowness

C Residual of peroxidase, at time t

K rate of reaction (s^-1)

t time (min)

T absolute temperature (K) *Subscripts*

nNormalised value

HWB Hot water blanching

1. INTRODUCTION

Guava(*Psidiumguajava*L.)is believed to introduce in India since early 17th century. In India it is fifth most widely grown fruit, In India it occupies an area of 2.03 lac hectares with annual production of 22.7 lacs MT. Allahabad area in U.P is reputed for the production of high quality of guava in India .**[1]**"Allahabad Safeda" is possibly the world's most cultivated cultivar and is highly popular in India. It is a medium to large size cultivar, characterized by its thin, smooth skin; dense, white flesh; and only a few seeds, which makes it suitable for canning. It has pleasant flavour, high TSS and vitamin C content. **[2]**Guavas are useful sources of nicotinic acid, phosphorous and soluble fiber. While having low fat and calories, guavas are cholesterol and sodium free. In order to preserve and commercialize this product, the heat treatment of blanching before further processing such as canning, freezing and dehydration is a necessary step in order to inactivate enzymes responsible for quality changes that occur during distribution and storage. Blanching has some additional advantages like destroying microorganisms and elimination of off-flavor. However, the degree of thermal treatment during blanching process can have adverse effect on sensorial (excessive loss of texture and unwanted changes of colour) and nutritional quality attributes. Many researchers studied these alterations in different fruits. They have observed the dramatic effect of blanching on the degradation of fruit and vegetables nutrient content (namely vitamin C and protein) and antioxidant properties.Colour is a primary consumer perceived characteristic of a product and plays an important role on food acceptance. Furthermore, colour of a processed product is often expected to be as similar as possible to the raw one. Therefore, maintaining the natural colourin processed fruits and vegetables products has been a major challenge in food processing. Changes in fruits and vegetablescolour can be associated with its previous heat treatment history and is also an indicator of heattreatment severity. The retention of total colour can be used as a quality indicator to evaluate the extent of deterioration due to thermal processing.

Peroxidases are the most heat stable enzymes in fruits and vegetables and their inactivation is used to indicate the adequacy of blanching. The presence of residual peroxidase in processed products may cause quality, changes, such as texture, colour, flavor and nutritional losses, however its role on quality losses during storage period of fruits and vegetables is not clear yet. For these reasons, it is desirable to keep blanching treatment conditions at a level strictly sufficient to cause inactivation of the deleterious enzymes and minimize quality losses.

Knowledge on degradation kinetics of enzyme inactivation and quality changes including the reaction order and the reaction constant is essential to predict quality losses during thermal processes. Several researches have been studied themodeling of thermal degradationkinetics of colour in different range of temperature. The majority of the published works on inactivation kinetics are well described byzero (Eq. (2)), first-order models (Eq. (3)) or thefractional conversion (also known as reversible first ordermodel).**[3]** There is currently no published data forthe changes in vital parameters, colour changes and peroxidase inactivation kinetics due to thermal processing inAllahabadsafedaguava.Therefore, the aim of this study was todevelop mathematical model for peroxidase inactivation kinetics

of 'Allahabad safeda guava' during hotwater blanching. This information will help to optimize hotwater blanching process for 'Allahabad safeda guava'.

2. MATERIALS AND METHODS

Materials: Raw 'Allahabad safeda' guavas *(Psidiumguajava*L.)at commercial maturity were purchased from a localmarket in Allahabad U.P. The fruits were washed andhand peeling (**HP**) was done. Then,seeds were removed and it was cut into cubes (2×2×2cm).Analysis of initial characteristics of the raw guava(T_0) was done. **[3]**

Hot Water Blanching (HWB) Process: Guavas(*Psidiumguajava*L.) cubes was blanched in a digital water bath maintained at desired temperatures (80°C±0.5°C).changes in different parameterswere studied at temperatures ranging from 80°C, with different times of exposure ($T_1$for 4,$T_2$for 8and $T_3$for 12 and T_4 for 16 minutes). After preset times, the samples were removed from the water bath and placed immediately in cooled water (2-5°C) in order to stop thermal inactivation instantaneously. Thetemperature of the water bath and cooled water was verified with a digital thermometer and. Each experiment was done in 5 replications. An unblanched sample was taken as a control (T_0). **[3]**

Moisture (%): Moisture (%) was estimated using standard method as given in 'handbook of analysis and quality control for fruit and vegetable products". [4]

Total Soluble Solids (TSS): TSS was determined using a hand refractometer. The TSS is expressed in °Brix

Acidity (%): Acidity (%) was estimated using standard method as given in 'handbook of analysis and quality control for fruit and vegetable products". **[4]**

Vitamin C (ascorbic acid): Ascorbic acid (mg/100gm) as % citric acid was estimated using "2, 6-Dichlorophenol-Indophenol Visual Titration Methodasgiven in 'handbook of analysis and quality control for fruit and vegetable products".**[4]**

Reducing sugar: Reducing sugar (%) was estimated using "Lane and EynonMethod"asgiven in 'handbook of analysis and quality control for fruit and vegetable products". **[4]**

Total Phenol Content: Total phenol content (TPC) was determined using the Folin-Ciocalteu's reagent **[5]**.TPC is expressed in Gallic acid equivalent GAE (mg GAE/100gm)

Enzyme Extraction Procedure: In order to determine the presence of peroxidase in guava and ratiobetween sample weight (g) and the buffer solution volume (mL), preliminary experiments were carried out. Blanched samples were mixed with cold potassium phosphate buffer in the proportion of 3:25 w/v. Each sample was homogenized in tissue homogenizer for 1 min at 13,500 rpm under chilled condition. Thehomogenate was filtered using filter paper. (Whatman No.1). The filtrate was centrifuged in acoolingcentrifuge at 6000×g and 4°C for 20 min with polypropylene tubes. The supernatants were kept on ice until chemical analysis **[6].**

Determination of Peroxidase Activity: Peroxidase activity inactivation and was measured according to the method reported by **[6]**.Peroxidase substrate solution was prepared daily by mixing 0.1 mL guaiacol, 0.1 mL hydrogen peroxide (30%)and 99.8 mL potassium phosphate buffer (0.1mol/L, pH 6.5). Peroxidase assays was conducted by pipetting0.12 mL of enzyme extract and 3.48 mL of substrate solution in the 10 mm path-length quartz cuvette. Peroxidase activities was measured from the increase in absorbance at 470 nm using an UV/vis spectrophotometer .The reaction wasmonitored for 20 min at 5sec intervals at 25°C. Enzyme activity was calculated from the slope of theinitial linear portion of a plot of absorbance vs. time. All experiments were run in 5 replications. Residual enzymeactivity (REA) in heat-treated samples is expressed as afraction of initial activity (P_0)

Residual enzyme activity (REA) = $P/P_0 \times 100$

Where; P and $P_0$are Absorbance/minute after heat treatment for time t and native enzyme, respectively.

Colour Measurement: Colour of fresh and heat-treated guava cubes was measured using a color lab terms of L-value (lightness), a-value (redness and greenness) and b-value (yellowness and blueness) as an average of three measurements at three different locations. From these values, total colour difference (TCD) was calculated according to the following equations:

$$TCD = \sqrt{(L - L_{0)+}(a - a_0) + (b - b_0)} \qquad (1)$$

Where; L_0, a_0 and b_0 are the readings at time zero and L, a and b the individual readings at each processing time.

To minimize the variability between different raw samples, the individual L, a and b values will be normalized, dividing the parameters by the corresponding initial values

Kinetic Modeling: The zero- (Eq. (2)) and first-order (Eq. (3)) equations will be used to describe the enzyme inactivation and colour changes in guava:

$$C = C_0 \pm kt \qquad (2)$$

$$C = C_0' \exp(-kt) \qquad (3)$$

Where; C is the measured value for residual peroxidase activity, C_0 the initialC, t is the heating time and k is the reaction rate constant. **[7]**

Statistical analysis: Data obtained was analyzed by using "two-way Anova" statistical technique. Rate constant of "Allahabad safeda" guava peroxidase enzyme was estimated by exponential and linear regression analysis.

3. RESULTS(TEXT, TABULAR):

Physico-chemical analysis of raw and blanched Allahabad safeda Guava

In the present study, four treatments namely, T_0, T_1, T_2, $T_3$and T_4 were formulated in which T_0 was the control i.e. raw guava wherein T_1, T_2, T3and T_4treatments, were blanched at **80 ^{0}C±5 for 4min**, **8min 12min and 16 min**respectively. These treatments were subjected to physico-chemical evaluation. The results are presented in Table 1.

Moisture content: It was observed that there was significant difference (at 5% level of significance) among moisture score in T_0, T_1, T_2, T_3 and T_4 Moisture content of T_1& T_2 decreased in comparison with T_0. This might be due the structural change in the product during blanching, which causes the pore openings of the food material to become enlarged thus increasing the rate at

which water is evaporated. This is similar to the observations of **[8]** who reported that pretreatment reduced the effect of skin thickness, which is a normal resistance to water loss at the surface of the product. But moisture content of T_3 and T_4 increased in comparison with T_2. This might be because blanching increases moisture content because of absorption of water by damaged cells and adhesion of water to the surface of the product **[9]**

Vitamin C (ascorbic acid): It was observed that there was significant difference (at 5% level of significance) among vitamin C (ascorbic acid) content of T_1, T_2, & T_3 decreased in comparison with T_0. This is because vitamin C (ascorbic acid) is a heat labile vitamin and blanching causes vitamin loss by thermal degradation, diffusion and leaching. **[10]**

Total soluble solids (TSS): It was observed that there was significant difference (at 5% level of significance) among Total soluble solids (TSS) contents in T_1, T_2, & T_3increased in comparison with T_0. Increase in blanching time caused an increase in TSS;this might bedue to water loss on

Continuous heating, the solute concentration increases that leads to increase in TSS (Purvis, 1983)**[11]**.The degradation of cellulose, hemicellulose and pectin releases soluble components which affects the TSS (Echeveirra*et al*., 1988)**[12]**.T_4 decreased in comparison with T_3,this might be because of long exposure to heat**.[13]**

Reducing sugar: There was significant difference observed (at 5% level of significance) among reducing sugar content of T1, T2 & T3 decreased in comparison with T0. This might be because during heat treatment reducing sugar decreases due to caramalization**.[14]**

It was also observed that there was significant difference (at 5% level of significance) among reducing sugar contents in T4 increased in comparison with T1, T2 &T3. This might be because of increase in blanching time caused an increase in the furan levels, as in earlier study it is observed increase in heating time increase in furan levels of reducing sugars. Furthermore, less furan was generated in non-reducing sugar system (sucrose) than in reducing sugar system (glucose and fructose). **[15]**

TPC: It was observed that there was significant difference (at 5% level of significance) among TPCcontents in T_1, T_2,T_3& T_4 increased in comparison with T_0. This increase in TPC could be explained by the fact that these compounds are easier to extract as a result of structural alterations that takes place during processing. According to literature the application of thermal treatments can also be associated with the dissociation of conjugated forms into free phenolic compounds. **[16].**

Table 1.Mean values of physico-chemical properties of samples of Allahabad safeda guava

Parameters	Unit	T_0	T_1	T_2	T_3	T_4
Acidity	%	0.56	0.28	0.28	0.27	0.26
Ascorbic acid	mg/100gm	16.65	15.00	10.00	6.66	10.3
Moisture content	%	88.00	86.00	82.40	84.00	84.5
Reducing sugar	%	12.87	11.69	10.74	10.25	11.36
Totalsolublesolids	^{0} Brix	6.00	6.20	6.60	6.85	6.00
Total solids	%	12.00	14.00	17.60	16.00	15.5
Totalphenolcontent	(mgGAE/100gm)	138.00	165.33	170.83	182.34	201.75

Evaluation of adequacy of blanching by modeling the kinetics of Peroxidase Inactivation of 'Allahabad Safeda'during blanching

Peroxidase Inactivation-in the hot water blanching study, it was observed that the enzyme inactivation was significantly affected ($P< 0.05$) by the time gradient of the blanching process (Table 2). The residual Peroxidase activities in Allahabad safeda guava against processing time are presented in Fig. 1. Inactivation first-order kinetic model was tested for its acceptability to the thermal inactivation data. The monophasic first-order kinetic model yield good R^2values (0.888) at the test temperature.Similarly, theperoxidase inactivation in seedless guava [3] and in different vegetables, such ascarrots, potatoes, tomato, green beans, green asparagusand pumpkin has been reported to follow a first-ordermodel to describe the enzyme inactivation **[17, 18, 19,20].**

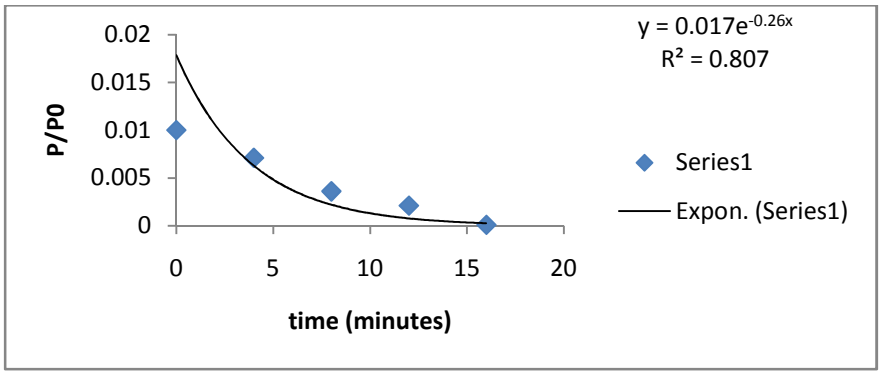

Fig. 1: Allahabad Safeda guava peroxidase inactivation during blanching process (experimental data at 80 ^{0}C±5 for 4min, 8min 12min and 16 min respectively.). The lines represent model fits to experimental data

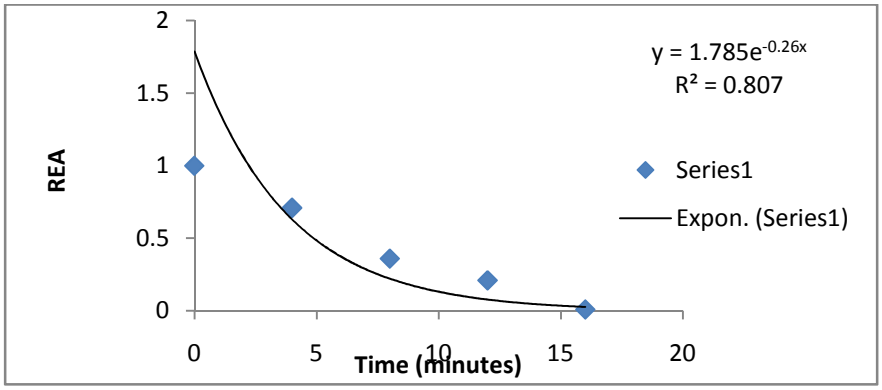

Fig. 2: Allahabad Safeda guava residual enzyme activity during blanching process (experimental data at 80 ^{0}C±5 for 4min, 8min 12min and 16 min respectively.). The lines represent model fits to experimental data

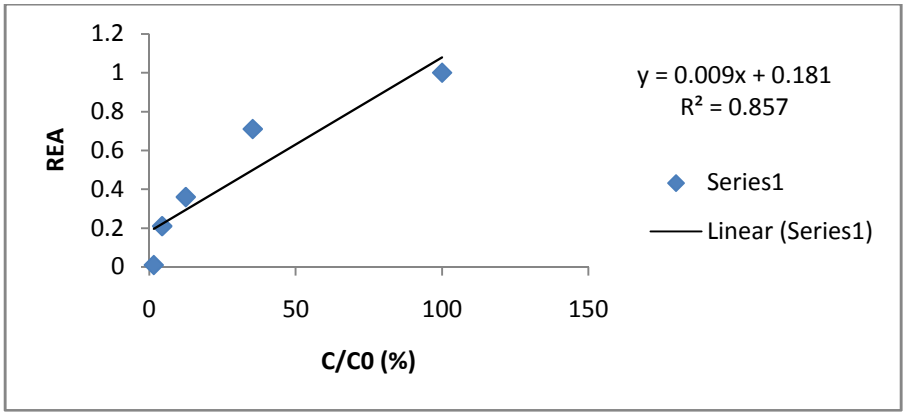

Fig. 3: Plot of residual for C/C0 experimental data against the predicted values of the model

Colour Changes: The L, a, b parameters were significantly affected ($P < 0.05$) by the time and temperature of the blanching process. The normalized lightness L (n) values were higher than that of the control. Initial increase and decrease with treatment time can be observed in table 2 since L is a measure of the colour in the light-dark axis [(ΔL = difference in lightness/darkness value) (+ = lighter) (– = darker)], this increased value indicates that the samples were turning lighter. Same results in other fruits were reported by **[21]**

According to Matsuura (1994) **[22]**, initial increase in luminosity could be caused by the destruction of the carotenoid structure giving a paler color. Along the time, other compounds, resulting mainly from the non-enzymatic browning reactions, like Maillard´s and oxidation of ascorbic acid or precipitation of the pigments**,** contributed to reduction of the luminosity, giving a darker appearance to the juice. **[21]**

Table 2 shows changes in the normalized **b** (n) values. These values increased after first treatment and decreased after thathowever The normalized lightness a(n) values were higher than that of the control.Since**b** is a measure of the colour in the yellow-blue axis [(Δb = difference on yellow/blue axis)(+ = yellower) (– = bluer)], this increased value indicates that the samples were turning yellower.

Table 2 shows change in the normalized a (n) values. These values increased with first treatment and after that decreased with treatment time, however The normalized lightness a(n) values were higher than that of the control.since a value is a measure of the colour in the red-green axis [(Δa = difference on red/green axis)(+ = redder),(– = greener)], this increased value indicates that the samples were turning redder (red to dark). According to Delicio S. et al. (1986) **[21]**, beta-carotene partially lost its red color after heat treatment, probably because it changed to the *cis* form **[23].** With time, the formation of other compounds, as already discussed, increased the juice dark color, contributing to the red intensity.

[19]follows the same trends of decreased in L and a value with increasing blanching time for carrot

One of the best parameters for describing the colour variation is the total colour difference (TCD) since it is a combination of parameters L, a, b. shows the variation of this parameter with the treatment time at the test temperature. Fig. shows changes in the TCD (n) values. These values decrease with treatment time. Differences in visual colour can be classified based on total colour difference (TCD).**[24**] reported that TCD values were corresponded to the noticeable differences in the visual perception of products. In the present study TCD was observed to be very distinct for the treatment conditions investigated. It should be noted that changes in colour values may be regarded as a negative sensory impact of processing.

Looking at these results, it is possible to conclude that the guava blanched treatments had the smaller variation compared to the control, for the X-rite color dimensions.

TABLE: 3 Effect of Blanching (time, temp.) on colour characteristics of Allahabad safeda guava.

SAMPLE	TREATMENT	TEMP(0C)	TIME(min)	L*	a*	b*	TCD
T0	NO	0	0	40.5	-2.87	6.05	0
T1	HWB	85	4	45.05	-3.14	7.02	1.84
T2	HWB	85	8	43.97	-2.79	6.48	1.77
T3	HWB	85	12	43.12	-2.63	6.15	1.73

4. CONCLUSION

Blanching in hot water was carried out in order to inactivate peroxidase enzymeand to improve the quality and to increase shelf life of Allahabad safeda guava. Effect of hot waterblanching on different physical and nutritional quality like acidity ascorbic acid moisture content reducing sugar total soluble solids total solids total phenol content were evaluated. Blanching of Allahabad safeda guava leads to degradation of various nutrients. Loss was maximum at 800C for 16 min and minimum at 800C for 4 min ofblanching. At the 80 ^{0}C T1 (4 min) the heat-labile fraction was rapidly inactivated and the kinetic behavior would correspond to the heat- resistant fraction that followedmonophasic first-order inactivation kinetics. Blanching results ininitial increase and decrease with treatment time and increase in L*, a* and b* value observed. Loss of acidity ascorbic acid moisture content reducing sugar total soluble solids total solids total phenol content might be related to the migration or leaching of component into the water. Propercombination of time and temperature of blanching is very important to retain thenutrients and quality of Allahabad safeda guava. At 80 ^{0}C temperature short time blanching havebetter retention of the entire nutritional component along with colour properties. Therefore low temperature treatment with short duration was most suitablemethod of blanching.

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