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Research Article

EFFECT OF ULTRAVIOLET AND THERMAL TREATMENTS ON SOME PHYSICO-CHEMICAL PROPERTIES OF MOSAMBI JUICE

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Abstract- The present experiment was conducted at College of Agricultural Engineering, Kandi, Sangareddy. Effect of ultraviolet (UV-C) and thermal treatments on physiochemical properties (*viz.* pH, total soluble solids (TSS), titrable acidity, total color difference, ascorbic acid content) of mosambi juice were studied during this research work. The process conditions for ultraviolet treatment were sample thickness (1, 2, 3 mm), treatment time (15, 30, 45 min) and distance of sample from lamp source (10, 20, 30 cm). Whereas, for thermal treatment the process parameters were temperatures (60, 70, 80 °C) and treatment time (5, 10, 15 min). The result obtained from this study showed that the ultraviolet treatment (UV) doesn't have any significant effect on pH, TSS, titrable acidity of mosambi juice. However, the UV treatment conditions had significant effect on total color difference (ΔE*), ascorbic acid of mosambi juice. Further, the thermal treatment showed significant effect on pH, TSS, titrable acidity, total color difference and ascorbic acid content of mosambi juice. The maximum change in pH, TSS, titrable acidity, total color difference and loss of ascorbic acid content of mosambi juice after UV-C treatment are 0.15, 0.2, 0.018% citric acid, 0.98 and 2.7% respectively. Similarly, The maximum change in pH, TSS, titrable acidity, total color difference and loss of ascorbic acid content of mosambi juice after thermal treatment are 0.47, 0.35, 0.42 % citric acid, 5.1 and 12.1% respectively. The above results suggested that ultraviolet treatment was found superior based on changes in physical properties of mosambi juice.

Keywords- Mosambi juice, Ultraviolet treatment, Thermal treatment, pH, TSS.

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Introduction

Fruit juices are products obtained from fresh, ripe and healthy fruits. They are also good sources of vitamins and minerals and are associated with many health benefits [1]. The major component of the fruit juice is water. The other most common constituent is carbohydrates which comprise sucrose, fructose, glucose and sorbitol. Also, limited amount of protein and minerals are found in fruit juices. However, juice contains no fat or cholesterol. If it is not added, no fiber content can be observed [2]. Sweet orange (or) sweet lime [*Citrus sinensis* L. Osbeck] is commonly known as "Mosambi" in Indian subcontinent. It is native to Asia and best cultivated in India, China, Southern Japan, Vietnam, Malaysia, Indonesia and Thailand. This fruit is eaten fresh or squeezed to make juice, a rich source of vitamin- C and replenish energy.

Sweet lime is considered beneficial for an overall good health. Being a citrus fruit, it is rich in Vitamin C and folic acid content. Mosambi is considered as low calorie and low fat juice which contains carbohydrates, proteins, and minerals. Some of these minerals includes calcium, iron, phosphorus, potassium, and dietary fiber. Vitamin content includes C, A, and extremely beneficial compounds including Flavonoids, Phenolic, carotenoids and limonoids. It is cholesterol free.

Mosambi juice has numerous health benefits. However, shelf life of juice is very less as compared to freshly squeezed juice. The spoilage of juice is due to the presence of enzymes and microorganisms. It must be consumed soon after extraction because when the extracted juice is exposed to air, the oxidative and enzymatic changes deteriorate natural quality of the juice. Oxidative enzymes such as peroxidase (POD) and polyphenol oxidase (PPO) modify the typical food properties, causing reduction of nutritional value and color. The increased bitterness in juice and change in taste of the aged juice is due to bitter chemical called Limonin. In freshly squeezed juice there is almost no limonin but some precursor chemicals of limonin are present. When fresh juice is exposed to air enzymes convert those precursors into bitter Limonin. This process is known as Enzymatic Bittering and is responsible for rancid flavor development.

Presently thermal treatment is most commonly applied for inactivating enzymes fruit juices [3]. But the thermal treatment leads to destruction of heat sensitive nutrients which restricts its application for processing of Mosambi juice. Besides the loss of nutrients, it also shows a detrimental effect on color and flavor. Alternative methods are addition of microbiocidal agents, high pressure application, pulsed electric field, irradiation, and aseptic packaging. However, there are some disadvantages of these techniques in terms of cost, loss of ascorbic and some other quality attributes [4].

Considering these limitations of other techniques, UV-C radiation can be used as an alternative method for processing and preservation of juice. This process does not produce chemical residues. Besides, it is a low-cost operation and effective against many microorganisms and enzymes.

Materials and Method

The present experiment was conducted at College of Agricultural Engineering, Kandi, Sangareddy in the year 2017. Mosambi juice was selected as raw material. Detailed materials and methods is described below.

Juice Preparation

Mosambi fruits of proper maturity and ripeness was purchased from a local

market. Mosambi juice was extracted at very hygienic conditions with the help of a manually operated "screw type juice extractor" in which de-pulped fruits were pressed to extract juice. The extracted juice was centrifuged at optimum condition of 5000 rpm for 30 min to remove the suspended particles in the juice. The supernent obtained after centrifugation is collected and stored in pre-sterilized bottles for analysis.

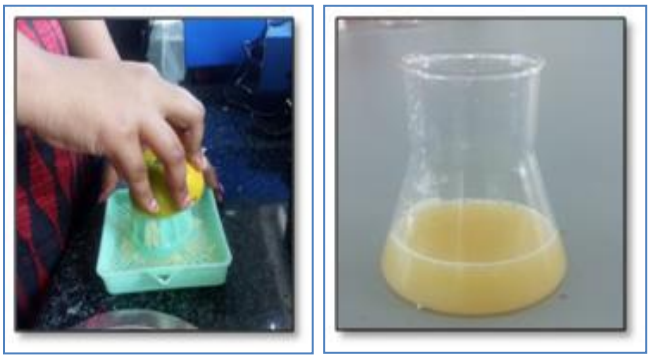


Fig-1 Extraction of juice from mosambi fruit

Chemicals and reagents

All the chemicals and reagents used in the study were analytical grade and procured from Merck, India and Sigma-Aldrich, Germany.

Ultraviolet Treatment of Mosambi Juice

Mosambi juice was poured in 100 mm standard size petri plates. Petri plates having mosambi juice was placed at the center of holder platform (holder platform was used to maintain the distance of sample from lamp source). Sample thickness was maintained by changing the volume of coconut water in petri plates. Coconut water was treated at different treatment conditions *viz*. sample thickness (1, 2, 3 mm), treatment time (15, 30, 45 min) and sample distance from lamp source (10, 20, 30 cm). Full factorial design was followed throughout the experiment. To prevent the exposure of UV light to human skin, a cover was placed in front of the system.

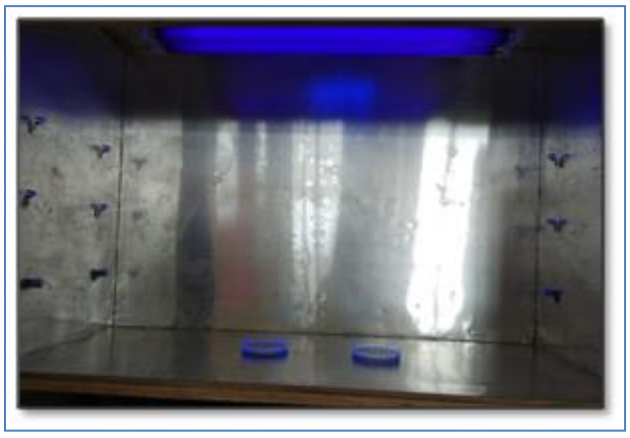


Fig-2 UV-C treatment of mosambi juice

Experimental design

Full factorial design with 2 replications was followed throughout the experiment. The independent variables *viz*. Treatment time (t), Distance from lamp source (H), Sample thickness (x) were selected with three levels of each of independent variables and their combinations had been investigated for each attribute.

Independent variables:

- Treatment time (t): 15, 30, 45 min
- Sample thickness (x): 1, 2, 3 mm
- Distance from lamp source (H): 10, 20, 30 cm

Responses: TSS, pH, Color, Titrable acidity, Ascorbic acid.

Thermal Treatment of Mosambi Juice

Thermal treatments were performed in a temperature controlled (± 0.5 ^{o}C) water bath at 60, 70, 80 ^{o}C for 5, 10, 15 min. Approximately 50 ml of sample was filled in beaker and top of the beaker was covered with aluminum foil. The beaker is placed in a water bath and the countdown time began when center of the sample reached the target temperature. All the above mentioned responses were calculated after thermal treatment of mosambi juice.

Fig-3 Thermal treatment of mosambi juice

Physicochemical Analysis of Mosambi Juice

Measurement of pH

pH of the sample was measured using a digital pH meter in triplicates. The probe of pH meter was inserted into sample and the stable reading obtained was considered as the final pH value.

Fig-4 Measurement of pH using digital pH meter.

Measurement of total soluble solids (TSS)

Total soluble solids (TSS) indicates the sweetness of juice sample.

Fig-5 Measurement of TSS using refractometer.

Total soluble solids (TSS) of sample was determined using a digital handheld refractometer (Model: PAL-1; Make: Atago, Japan), having a range of 0-53% according to the methods proposed by [5]. Before measurement of TSS of sample, the refractometer was calibrated using double distilled water. A drop of the mosambi juice was placed on the sample slot refractometer and the TSS of the sample was recorded and expressed in ˚Brix.

Measurement of titratable acidity (TA)

Titratable acidity (TA) in the sample was determined by titration method. Briefly, 10 mL of juice sample was taken and diluted to 30 mL with distilled water. 10 mL diluted sample was taken for titration, mix 2-4 drops of 1% phenolphthalein indicator and titrated against 0.1 N NaOH solution. Titer values were noted and titrable acidity was expressed as citric acid percentage since citric acid is the dominant organic acid in mosambi juice.

$$\text{T. A (\% citric acid)} = \frac{\text{Titre value} * \text{Normality of NaOH} * \text{milli Equivalent weight factor of malic acid} * 100}{\text{Vol. of sample taken}} \quad ..[1]$$

Measurement of color

Color of mosambi juice was measured based on CIE color parameters L* (0-100, Black-Lightness), a* (positive values – red, negative values- green and 0 is neutral) and b* (positive values – yellow, negative values- blue and 0 is neutral). Colorimeter was used to measure the color parameters. Results were expressed as the mean of three measurements. The overall color difference (ΔE*) can be calculated using [Eq-2].

$$\Delta E^* = \sqrt{(L_0^* - L_1^*)^2 + (a_0^* - a_1^*)^2 + (b_0^* - b_1^*)^2} \quad [2]$$

Fig-6 Measurement of color using hunter lab colorimeter.

Measurement of ascorbic acid content

Ascorbic acid content of mosambi juice was measured using 2,6-Dichlorophenol-Indophenol visual titration method. Briefly, 10 ml of mosambi juice was taken and make up to 100 ml with 3% HPO_3. After this sample is filtered through filter paper. 5 ml of diluted and filtered juice was taken in titration flask and titrated against standard dye solution to a pink end-point which should persist or at least 15 seconds. The titre value was noted and the amount of Ascorbic acid was calculated by using equation [3]

$$\text{(mg of Ascorbic acid/100ml)} = \frac{\text{Titre value X dye factor X Volume madeup X 100}}{\text{ALiquot of extract taken for estimation} \quad X \quad \text{Weight(or)volumeof sample taken for estimation}} \quad ...[3]$$

Dye factor was calculated by dye standardization method.

Experimental Design and Data Analysis

A full factorial design was used to estimate the effect of independent variables (Treatment time, sample thickness and distance sample from lamp source) on responses (Total color difference, pH, TSS, titrable acidity, ascorbic acid). Analysis of variance (ANOVA) test was conducted using Design expert version 7.0.0 software (State-Ease Inc., Minneapolis, USA) to evaluate the significance (at 95% confidence level) of the effect of independent variables and their interactions on the responses.

Optimization of Process Parameters for Ultraviolet and Thermal Treatment of Mosambi Juice.

RSM was applied to the experimental data using Design expert version 7.0.0 software (State-Ease Inc., Minneapolis, USA). The critical responses were screened out Based on the effect and importance of responses. The total color change, TSS and ascorbic acid loss were the critical quality attributes affected by ultraviolet treatment. Therefore, the ultraviolet and thermal treatment conditions were optimized for processing of mosambi juice. The optimization was targeted for the maximum retention of Ascorbic acid with minimal changes in color.

Results and Discussion

Compositions of Raw Mosambi Juice

The physicochemical *viz.*, pH, TSS, Titrable acidity, Total color difference and ascorbic acid content of mosambi juice were analyzed before treatment. The compositions of mosambi juice varied from fruit to fruit depending upon variety and maturity of fruit. Although there was important initial difference exist in physicochemical properties of mosambi juice between different verities of fruit. But for comparison these parameters kept as constant for whole experiment. The compositions of fresh mosambi juice were measured and presented in [Table-1].

Table -1 *Physicochemical characterization of raw mosambi juice.*

Parameters	Value
pH	3.56 ± 0.15
TSS (°Brix)	12.32 ± 0.16
Titrable acidity (% citric acid)	0.45 ± 0.01
Ascorbic acid	27 ± 0.25
L*	53.69 ± 0.5
a*	-4.65 ± 0.1
b*	19.32 ± 0.4

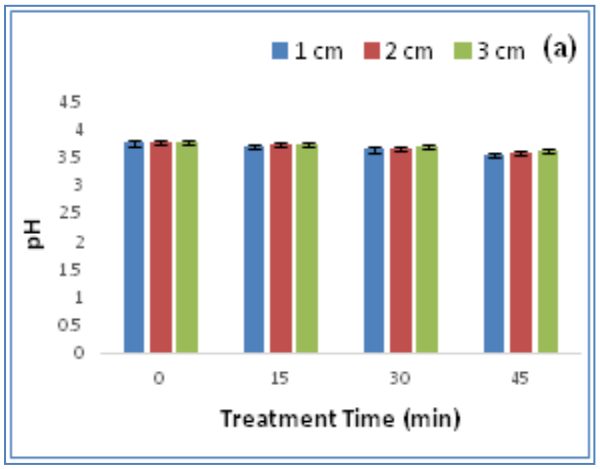
Values reported as mean ± standard deviation (N = 10).

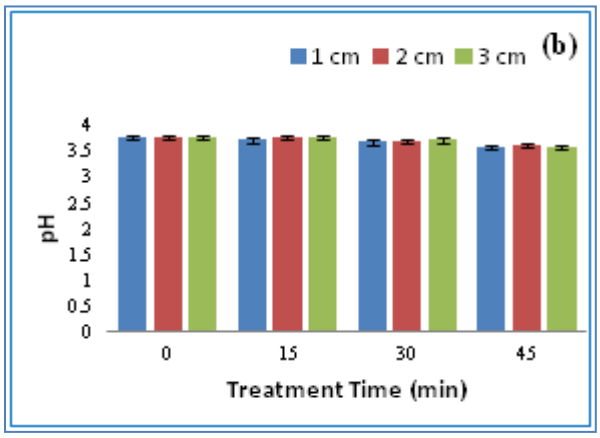
Effect of Ultraviolet and Thermal Treatments on Physicochemical Properties of Mosambi Juice

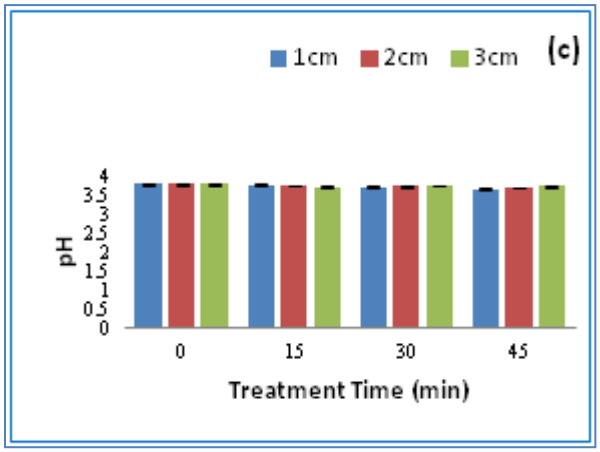
Effect on pH

The pH values of mosambi juice during Ultraviolet and thermal treatments at different conditions were presented in [Fig-7 (a-c)] and [Fig-7 (d)] respectively. Generally, The pH plays an important role in phenomenon such as enzyme activity, protein denaturation and microbial inactivation kinetics and most microorganisms show increased susceptibility and inability to recover from sub-lethal injuries at low pH values. The pH of mosambi juice at all experimental conditions were found to be in the range of 3.41 to 3.71. Further, from the ANOVA

data it was noticed that ultraviolet treatment conditions had no significant effect on pH. However, thermal processing conditions of mosambi juice had a significant effect on pH ($p<0.0001$). The ultraviolet treatments at different distances such as 10, 20 and 30 cm were shown negligible changes in pH values at different time intervals (0.0-45.0 min). Further, at different thickness levels *viz.*, 1, 2 and 3 mm at a particular distance and time interval for example 10 cm at 45 min showed slight differences in pH values. In addition, during UV treatment the pH varied from 3.55-3.7 with respect to any replication. The UV light system might not severe enough to cause the release of H^+ ions from mosambi juice and hence the pH of mosambi juice remained almost constant after ultraviolet treatment. In addition, the slight variation of pH can be considered as experimental error. Moreover, pH as a function of treatment time at various temperature intervals was shown in [Fig-7(d)]. In this, variations in pH values of mosambi juice samples at different experimental conditions were effected the pH of sample. The changes in pH after thermal treatment may be due to the stress induced due to temperature may cause release of H^+ ions in mosambi juice.







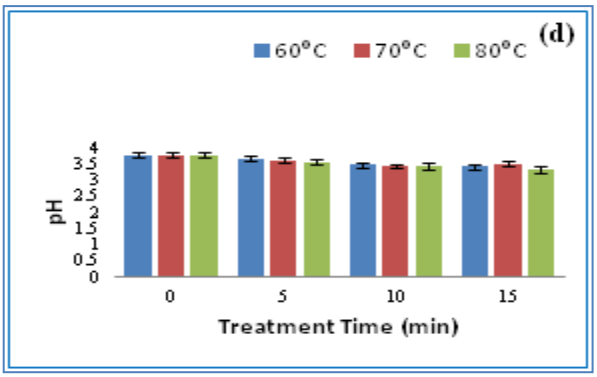
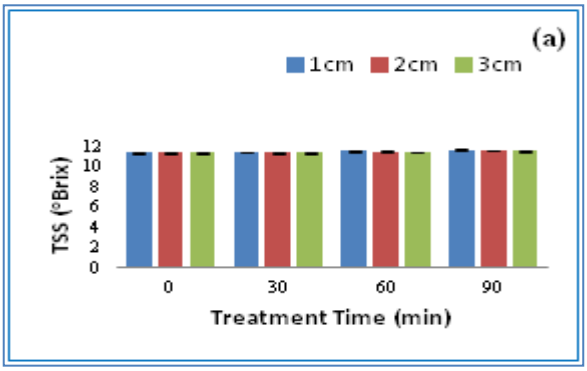
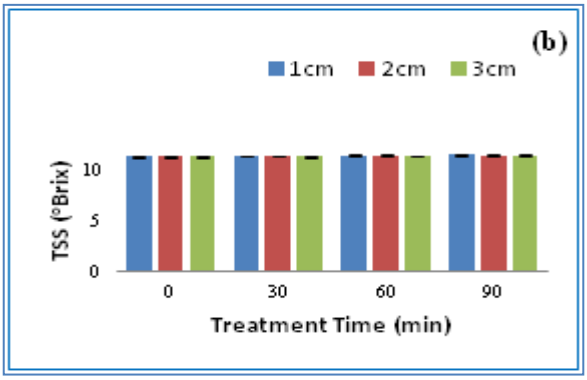


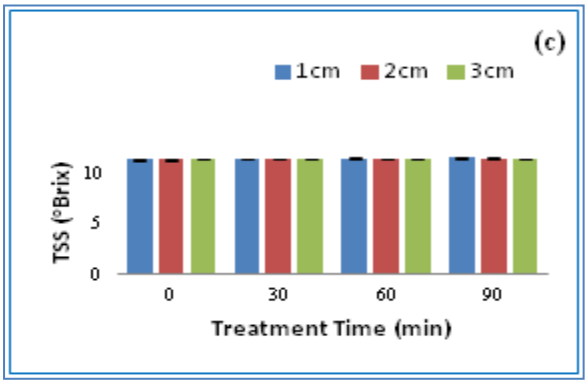
Fig-7 Effect of different treatment conditions on pH of mosambi juice (a) UV, 10 cm (b) UV, 20 cm (c) UV, 30 cm (d) Thermal treatment

Effect on TSS

The TSS values of UV light and thermal processed mosambi juice at different conditions were presented in [Fig-8(a-c)] and [Fig-8(d)] respectively. In general, Total soluble solids (TSS) indicated as the sweetness of mosambi juice and were found in the range of 12.16 to 13.2 °Brix. From ANOVA data, it was noticed that ultraviolet treatment conditions had no significant effect on TSS. However, thermal treatment conditions had significant effect ($p<0.0001$) on TSS of mosambi juice. However, the maximum deviation obtained in TSS after ultraviolet treatment is ± 0.2 with respect to any replication, whereas the maximum deviation after thermal treatment is ± 0.6 with respect to any replication. The ultraviolet treatments at different treatment time interval (0.0-0.45 min) showed slight difference in TSS. However different distances such as 10, 20 and 30 cm were shown negligible changes in TSS values. Further, at different thickness levels *viz.*, 1, 2 and 3 mm at a distance and time interval for example 10 cm at 45 min showed slight differences in TSS values. TSS represents soluble sugars. Sugars such as glucose, fructose do not absorb UV-C in the range of 200-280 nm. Hence there will be no effect of UV-C treatment on TSS of mosambi juice. The thermal treatment at different temperature and time intervals showed difference in TSS values. So far there is no review available to compare the TSS value of mosambi juice after ultraviolet treatment. The similar trend followed in the review cited in [6, 7, 8].







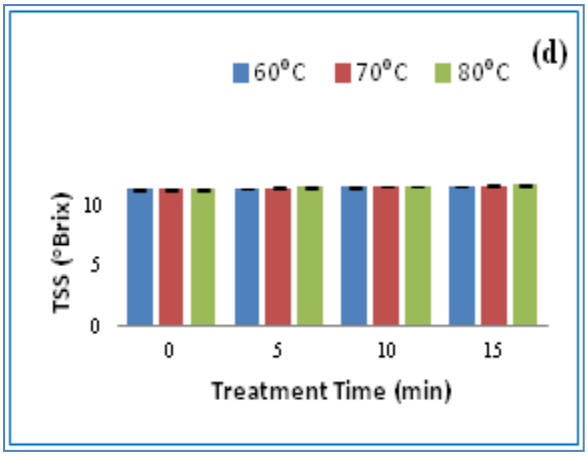
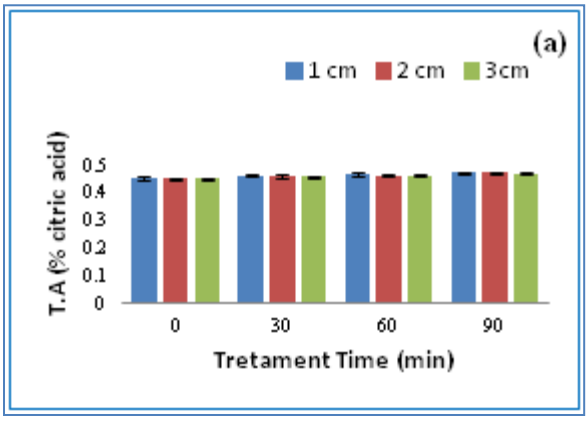
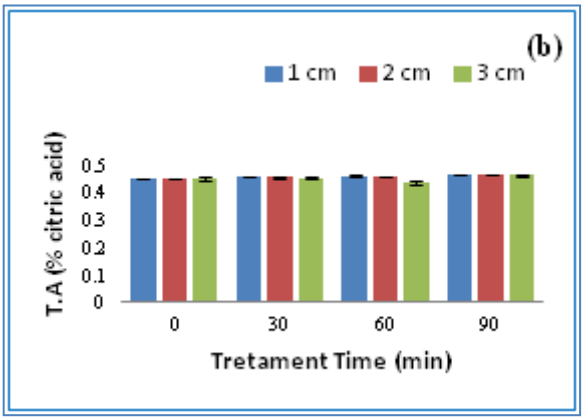


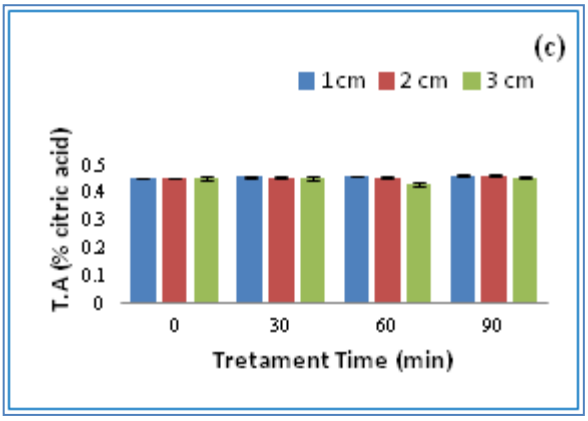
Fig-8 Effect of different treatment conditions on TSS of mosambi juice (a) UV, 10 cm (b) UV, 20 cm (c) UV, 30 cm (d) Thermal treatment

Effect on titrable acidity

The titrable acidity values of UV light and thermal processed mosambi juice at different conditions were presented in [Fig-9(a-c)] and [Fig-9(d)] respectively. In this study, the Titrable acidity of mosambi juice was expressed as citric acid percentage since citric acid is the dominant organic acid in mosambi juice. Titrable acidity of mosambi juice was found to be in the range of 0.44 to 0.46 (% citric acid). From ANOVA data, it was noticed that ultraviolet treatment conditions had no significant effect on titrable acidity. However, thermal treatment conditions had not significant ($p>0.0001$) effect on Titrable acidity of mosambi juice. However, the results show that there are slight higher values of Titrable acidity after UV and thermal treatment of mosambi juice with respect to control (Raw mosambi juice). This might be due to the UV light doesn't cause to release the H^+ during the treatment. On the other hand, there is slight increase in Titrable acidity after thermal treatment. Thermal treatment may change the metabolic pathway hence there might be some increase the amount of citric acid. The similar trend followed in the review cited in [9, 10].







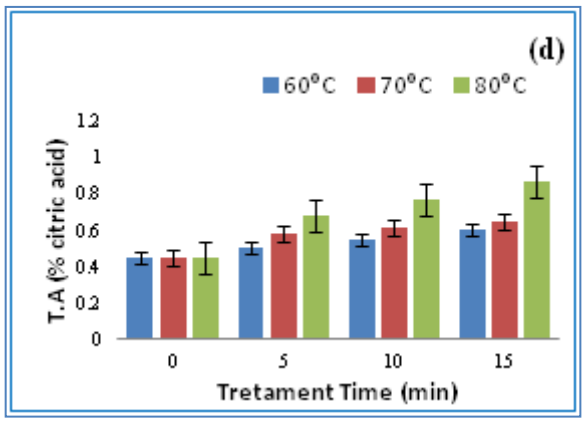
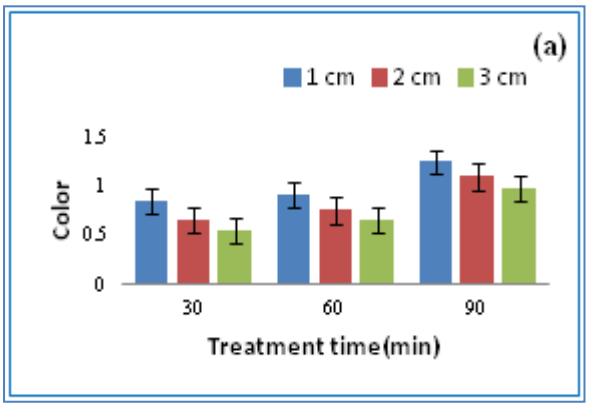


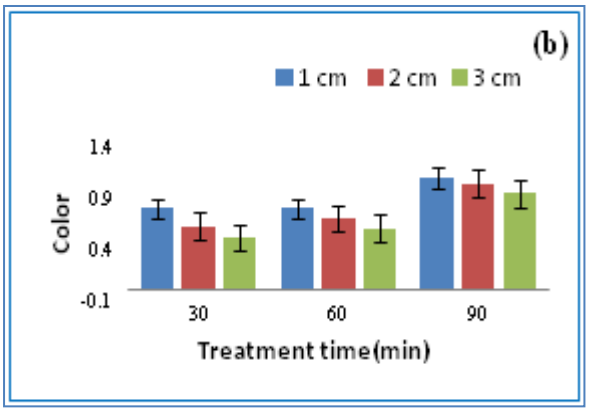
Fig-9 Effect of different treatment conditions on Titrable acidity of mosambi juice (a) UV, 10 cm (b) UV, 20 cm (c) UV, 30 cm (d) Thermal treatment.

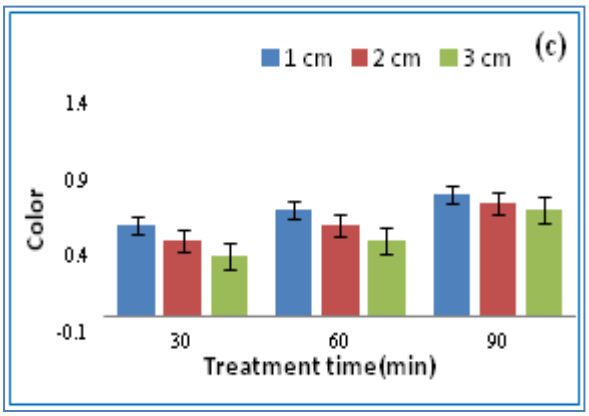
Effect on total color difference

The total color difference values of UV light and thermal processed mosambi juice at different conditions with respect to control (unprocessed mosambi juice) were presented in [Fig-10(a-c)] and [Fig-10(d)] respectively. The total color difference was calculated based on L*, a*, b* values. From ANOVA data it was showing that the ultraviolet and thermal treatment conditions had significant ($p<0.0001$) effect on total color difference in mosambi juice. The ultraviolet treatments at different distances such as 10, 20 and 30 cm were showed slight changes in total color difference at different time intervals (0.0-45.0 min). Further, at different thickness levels *viz*., 1, 2 and 3 mm at a particular distance and time interval for example 10 cm at 45 min showed negligible changes in total color difference. The changes in color after ultraviolet irradiation is due to that the UV radiation impairs some of the pigments present in the juice, either initially present or the ones formed later by the rapid action of polyphenol oxidase (melanins) as well as the Maillard reaction between sugars and amino acids (melanoidins). The maximum changes color difference was observed to be 1.25 after ultraviolet treatment. Whereas the maximum total color difference of mosambi juice after thermal treatment was

observed to be 5.1. Total color difference in ultraviolet is less as compared to and thermal treatment.







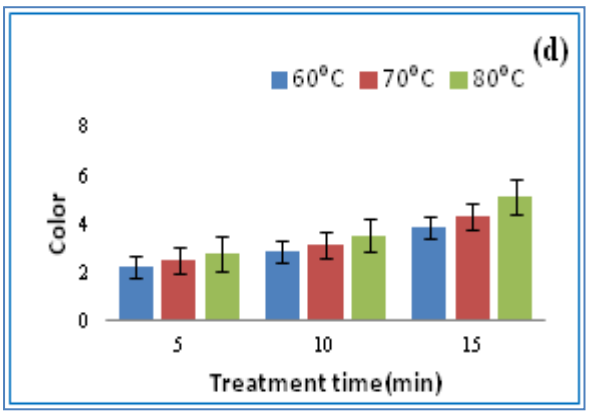
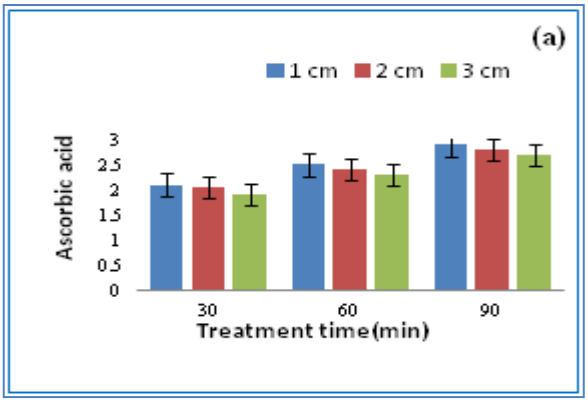
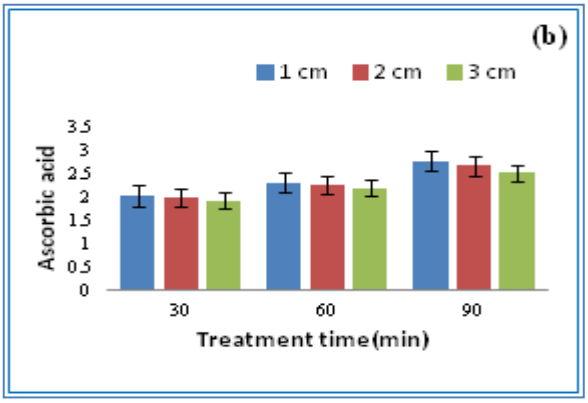


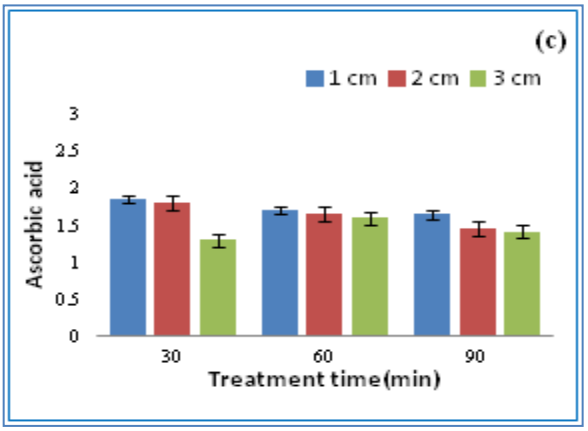
Fig-10 Effect of different treatment conditions on total color difference of mosambi juice (a) UV, 10 cm (b) UV, 20 cm (c) UV, 30 cm (d) Thermal treatment.

Effect on ascorbic acid (A.A) content

The percent loss in ascorbic acid content in mosambi juice after UV light and thermal treatments at different conditions with respect to control (unprocessed mosambi juice) were presented in [Fig-11(a-c)] and [Fig-11(d)] respectively. Ascorbic acid is a heat-sensitive bioactive compound that plays a vital role in human health and can act as an antioxidant. The AA content of mosambi juice was found to be in the range of 27 to 31 mg/100 mL. From ANOVA data it was showing that the ultraviolet and thermal treatment conditions had significant ($p<0.0001$) effect on ascorbic acid content in mosambi juice. The ultraviolet treatments at different distances such as 10, 20 and 30 cm were showed slight changes in % loss of ascorbic acid at different time intervals (0.0-45.0 min). Further, at different thickness levels *viz.*, 1, 2 and 3 mm at a particular distance and time interval for example 10 cm at 45 min showed negligible changes in % loss of ascorbic acid. The maximum loss (2.9 %) of A.A was observed at 10 cm sample distance, 1 mm thickness and 45 min treatment time. However the loss of ascorbic acid in mosambi juice after UV-C treatment was less compared to the thermal treatment where maximum loss of A.A found to be 12.1%. The result shows that the loss of A.A increases with treatment time in both.







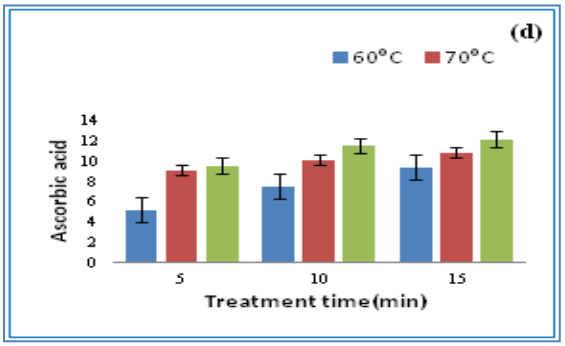


Fig-11 Effect of different treatment conditions on Ascorbic acid content of mosambi juice (a) UV, 10 cm (b) UV, 20 cm (c) UV, 30 cm (d) Thermal treatment.

Ultraviolet and thermal treatment. Heating affects the degradation of ascorbic acid in an aerobic pathway due to its heat-sensitive characteristic in the presence of oxygen. In addition to this, the depletion of ascorbic acid may be due to the formation of free hydroxyl radicals by photochemical reaction, related to oxidative processes. The similar results were reported by [11, 12]. The loss of A.A is increased with temperature in thermal treatment. So far there is no review available to compare the loss of A.A value of mosambi juice after ultraviolet treatment. The loss of A.A was found in the order of Control< Ultraviolet < Thermal treatment. This shows that UV treatment is a better processing method to retain the heat-sensitive ascorbic acid than thermal treatment due to absence of heat in UV treatment.

Optimization of Process Parameters

Optimization condition for ultraviolet and thermal treated mosambi juice were determined with the help of commercial software (Design Expert Version 7.0.0) to obtained the maximum inactivation of PPO and minimum changes in quality attributes.

Ultraviolet process parameters

The optimization of ultraviolet treatment was aimed to minimum changes in Ascorbic acid and color changes. The detailed parameters with their importance is shown in [Table-2].

Table-2 *Constraints for optimization of ultraviolet process parameters.*

Variable	Condition	Lower Limit	Upper Limit	Importance
Treatment time (min)	Minimize	15	45	4
Sample thickness (mm)	Maximize	1	3	3
Distance of sample from lamp source (cm)	Minimize	10	30	3
Responses	**Condition**	**Lower Limit**	**Upper Limit**	**Importance**
Loss of Ascorbic acid (%)	Minimize	0.35	2.7	4
Total color difference	Minimize	0.5	1.25	4

Thermal process parameters

The optimization of thermal treatment was aimed minimum changes in Ascorbic acid and total color difference of mosambi juice. The detailed parameters with their importance is shown in [Table-3] and [Table-4]. And the obtained optimum conditions were presented in [Table-5].

Table-3 *Constraints for optimization of thermal process parameters*

Variable	Condition	Lower Limit	Upper Limit	Importance
Treatment time (min)	Minimize	5	15	4
Temperature (°C)	Minimize	60	80	5
Responses	**Condition**	**Lower Limit**	**Upper Limit**	**Importance**
Loss of Ascorbic acid (%)	Minimize	5.2	12.1	5
Total color difference	Minimize	2.25	5.1	4

Table-4 *Predicted optimum values for ultraviolet variable and responses*

S.No	Time (min)	Thickness (mm)	Distance (cm)	Loss of A.A(%)	Total color difference	Desirability
1*	15	1	30	0.6	1.7	0.826
2	15	1.03	30	0.601	1.74	0.825

Table-5 Predicted optimum values for thermal variable and responses

S.No	Time (min)	Temperature (°C)	Loss of A.A (%)	Total color difference	Desirability
1*	5	60	5.81	2.17	0.97
2	5	60.5	6.1	3.51	0.754

* Selected for further studies

Summary and Conclusions

The effect of each variable and its combination was studied during this study. Statistical analysis was performed to test the significance. The major findings from the experiment are as follows.

- ✓ pH of mosambi juice was found almost same after UV-C treatment. Small changes in pH value was considered to be experimental error. Further, the thermal treatment parameters showed significant effect on pH of mosambi juice.
- ✓ TSS of mosambi juice was found almost same after UV-C treatment. Further, thermal treatment conditions has significant effect on TSS of mosambi juice.
- ✓ Titrable Acidity of mosambi juice slightly increased after UV-C treatment. On the other hand, there is significant increase in Titrable acidity after thermal treatment.
- ✓ The effect of process parameters on color of mosambi juice was evaluated. The color of mosambi juice has been significantly affected especially at higher temperature and treatment time. The maximum changes color difference was observed to be 1.25 after ultraviolet treatment. Whereas the maximum total color difference of mosambi juice after thermal treatment was observed to be 5.1.
- ✓ The effect of process parameters on ascorbic acid content of mosambi juice was evaluated. The ascorbic acid content of mosambi juice has been significantly affected especially at higher temperature and treatment time. The maximum loss of ascorbic acid content was observed to be 2.9% after ultraviolet treatment. Whereas the maximum loss of ascorbic acid content of mosambi juice after thermal treatment was observed to be 12.1%.

From above obtained results, this study was concluded that, the UV-C treatment is better processing method for mosambi juice with respective to retention of nutritional properties.

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Author's Contribution

Shivashankar S: Project Supervisor
Bhavani Namala, Harikrishna E, Swetha M, Prashanth Reddy Y, Spandana k: Conducted this study as a part of their B.Tech Project Work

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.

Conflict of Interest: None declared

Abbreviations:

TSS : Total Soluble Solids
UV : Ultraviolet
PPO : Polyphenol Oxidase

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