Impact Of Ultra Sonication On Safety And Quality Characteristics Of Orange Pulp

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Abstract
Fruit pulp has a huge amount of phenolic and other bioactive compounds as well as carotenoids, vitamins, and anthocyanins. They are usually processed by conventional pasteurization that allows obtaining juice pulp with a low microbial count. However, the use of thermal treatments results in adverse changes in composition and loss of organoleptic attributes such as loss of aroma, color, flavor, and texture of juices. With an increasing demand for high-quality, safe, nutritious, and minimally processed pulp with fresh-like characteristics, modern fruit processing industries are looking for alternative processing. Hurdle technologies are sought to maintain the nutritional profile of the fruit pulp. The purpose of this research is to combine ultrasound and additives as a hurdle technology to preserve the quality of pulp as consumer demands for safe and nutritious products. In this research, we study the combined effects of ultrasound and their additives on the overall quality of orange pulp was investigated. The effectiveness of ultrasound treatment was enhanced with a combination of additives which enhanced the overall quality of mixed orange pulp. The physicochemical and microbiological analysis were performed. Bioactive compounds were also measured. Finally, the data obtained by analyzing quality parameters was brought to statistical analysis. A statistically significant increase was noticed in total phenolic contents and total flavonoid contents whereas a decrease in microorganisms (TPC, Y&M count) were found in all the samples. The total value of antioxidant capacity as well as DPPH radical scavenging activity both increased significantly high. Physicochemical characteristics such as pH, total soluble solids, and titratable acidity were retained, and the results were non-significant. There were also some differences in the color values. However, maximum improvement for Ascorbic acid contents was observed in the T3 treatment. The results of this study revealed that the combined treatment T3 (ultrasound and additives) produced the best results and has the ability to improve the overall quality of orange pulp and can also be employed for industrial processing.

Keywords: Ultrasonication, Orange Pulp, Fruit Juices

Introduction

Orange contains vitamins, carotenoids, fiber, proteins, and iron, in addition to its good taste and nutrition. It helps in immunity, blood circulation, cancer treatment, eyesight, brain development and skin diseases. It is also an anti-inflammatory agent and an antioxidant. (Siddique *et al.,* 2017). A Mandarin (citrus fruit family) variety known as Kinow is commonly grown in Punjab, Pakistan. In Pakistan, the festival of Kinow begins in December and lasts until April. If kept at 5°C + 2°C and 85–90% relative humidity, Kinow has a shelf life of 60–90 days, depending on when it was harvested. Fruit pulp possesses high importance because of the high nutrition and availability of bioactive compounds, including vitamins, phenolic compounds, carotenoids, and anthocyanins (Aadil *et al.*,2020). Due to sensory characteristics and high antioxidant capacity, an increase in consumer demand is observed for fruit juices as they are being consumed worldwide for attraction in diets and healthy lifestyles (Mesquita *et al.*,2020).

Nowadays, consumers are more conscious of their diet. As a result, demand for food products with fresh-like characteristics, minimally processed, natural taste, nutritional value, and microbiological safety is increasing. Pasteurization usually processes fruit juices to reduce microbial loads and extend shelf life. Fruit pulp that has been processed using traditional thermal treatments is healthy and has a longer shelf life but negatively impacts the sensory,

physicochemical, and organoleptic properties of food products. These nutritional losses occur due to long treatment times and the low efficiency of heat transfer (Jin *et al.,* 2020)**.** Thermal processing of fruit pulp can also result in the denaturation of bioactive compounds and heat-sensitive nutrients (Nadeem *et al.,* 2018). Fruit and vegetable juices are subjected to thermal pasteurization to destroy enzymes and bacteria (Lagnika *et al.,* 2017). The most common thermal heat process is preserving fruit and vegetable juices. This process can improve the stability and shelf-life of different juices. Still, this treatment impacts the quality of juices in terms of nutritional and physicochemical parameters such as polyphenols, carotenoids, pH, vitamins, and minerals (C and E), aroma and taste evaluations (Rawson *et al.,* 201l).

The modern food industry is focusing on using new alternative non-thermal food processing technologies. Novel techniques like US, HPP, PEF, ultraviolet, irradiation, etc., are being explored on a large scale to extend juice quality and minimize nutritional losses. Researchers are looking forward to retaining the original properties and improving the nutritional profile of the products (Yildiz, 2019). Ultrasonic processing, which is being used as an alternative to thermal treatments for fruit juices, is one of the recent developments in the food industry. It improves food quality by disrupting microbial cells with high pressure, temperature, and shear produced by cavitation, resulting in lower microbial loads (Ahmad *et al.,* 2019). As a non-thermal viable method, ultra-sonication has been used in numerous petitions related to fruit juice production (Santhirascgaram *et al.,* 2015). In the case of fruit juices such as strawberry and orange, sonication treatment did not affect their quality (Adekunte *et al.,* 2010).

Thermo-sonication (1S) is a novel and practical method and this method can increase the deactivation rates of enzymes and microbes, the shelf life of produce extended, and the nutritional content of fruits and vegetable juice reduced. Bioactive components, sensory properties, enzymatic and microbiological characteristics, and physicochemical parameters are all affected. According to scientific evidence, this method appears to preserve the juice quality associated with heat treatment (60 degrees Celsius for 30 minutes) by using thermo-sonication. Heat and ultrasound can be used as preservatives for fruits and vegetables, and their enzymes and bacteria can be deactivated. Fruit and vegetable juices can be kept fresh for a more extended period by using this method (Anaya-Esparza *et al.,* 2017). Objectives of this research were as follows;

Objectives

- To enhance the shelf-life by maintaining orange pulp's physicochemical and nutritional profile.
- To retain heat-sensitive compounds and reduce the microbial load using the non-destructive non-thermal technique.

Methodology

Preparation of fruit juice

Pretreatment of raw materials

After sorting, the oranges were washed to remove the soil, and the damaged oranges were separated. Oranges were peeled, trimmed, washed, and sliced manually in a juice extractor.

Washing and Peeling

Before juice extraction washing and peeling of oranges are done.

Treatment plan

Individual and combined effects of ultrasound and additives on orange pulp were assessed.

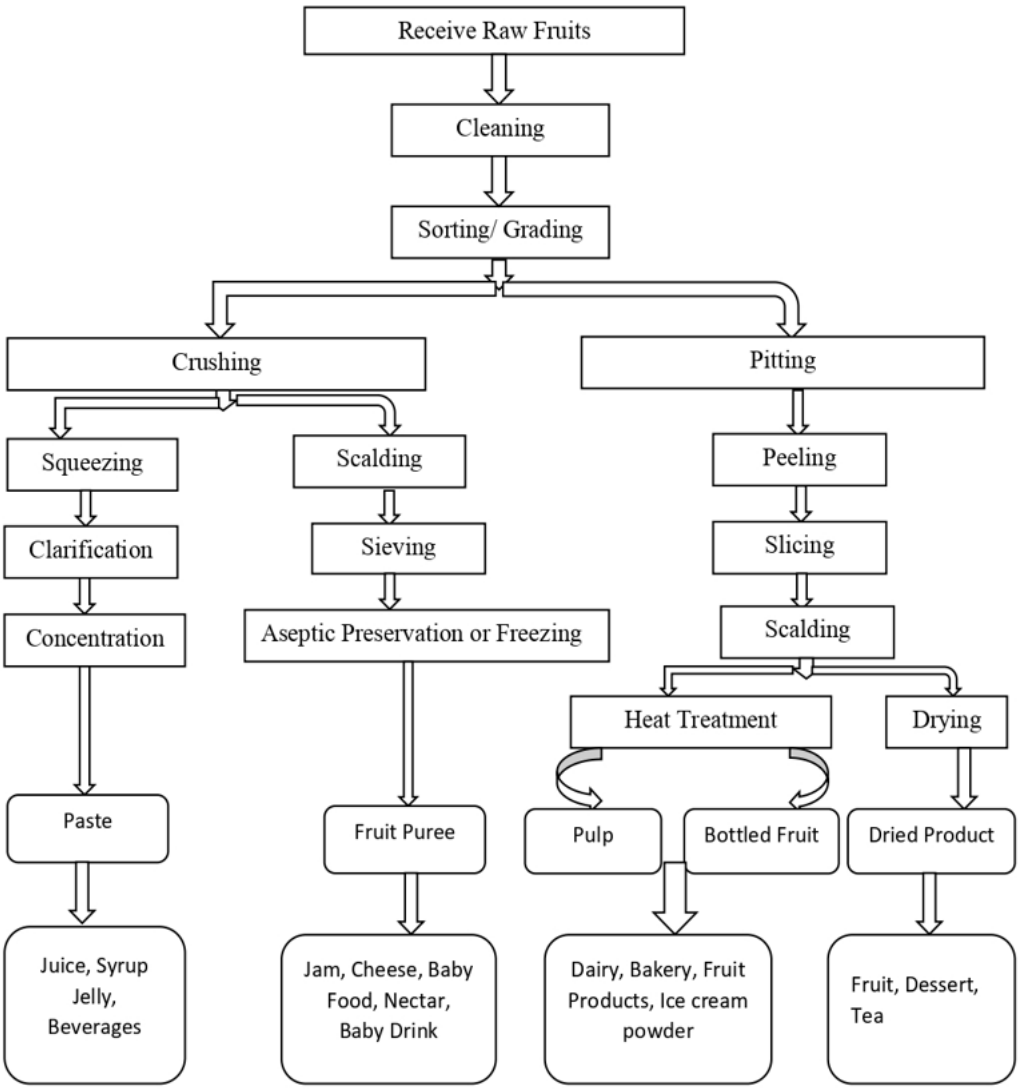
Table: Treatment plan for effects of ultrasound and additives

Treatment No.	Treatment applied
T_0	Control
T_1	Additives (10g Citric acid+ 2g sodium benzoate+ 2.5g Potassium meta bisulphite / per liter)
T_2	US (20 kHz, 1 min)
T_3	US (20 kHz, 1 min) + Additives (10g Citric acid+ 2g sodium benzoate+ 2.5g Potassium meta bisulphite / per liter)

US = Ultrasound treatment

Extraction and Filtration

Juice is then extracted with help of simple juice extractor. After extraction to remove foreign particles, gummy, and foamy substances from the juice it is filtered with the help of sieve and eight layered muslin clothe. After. That we used Whatman filter paper No. 1 and stored in aseptic way and treated to thermosonication method. Orange Juice were treated with ultrasonic homogenizer. Juice samples were treated at 20kHz frequency with probe diameter 10mm at constant power 99W. for T2 and T3 application of sonication for 1 min.



Physicochemical analysis

The mixture's physical qualities and chemical composition were evaluated using physicochemical analysis.

pH value

In both the treated and untreated samples, we used a digital pH meter to determine the pH. Juice pH is measured by the amount of negative log hydrogen ions present in the combination. – Buffer solutions with varied concentrations (4, 7, and 10%) were used to calibrate the pH meter in order to ensure accurate readings Using the process outlined above, the juice was injected into the probe, and a reading was taken after a few minutes (Yildiz, 2019).

Titratable acidity (TA)

The standard AOAC technique was used to determine the edge (%) according to the specified approach by Chang *et al*. (2017). To determine the titratable edge, we used the AOAC technique for orange juice. The benefit of the juice was measured by adding 80 mL of pure water to a 20 mL sample in a 250 mL beaker. After that, 2-3 drops of phenolphthalein indicator were added, and the mixture was vigorously mixed before being titrated against a standardized with 0.1 N NaOH. As the titration progressed, a pale pink hue emerged on the test stripe.

Calculation

Titratable acidity was estimated by using the following equation:

$$\text{Titratable acidity (\%)} = \frac{\text{Equivalent weight of acid x titer value x 100}}{\text{Volume of juice blend taken x 1000}}$$

Total soluble solids (TSS)

Total soluble solids (TSS) were measured at room temperature using a hand refractometer (25 °C). To acquire the zero reading, the equipment was calibrated with distilled water before use. TSS were recorded and converted to °Brix when a drop of the juice was put on the refractometer's pure prism. After taking measurements at room temperature, the refractometer's prism was properly cleaned with distilled water before being used again (Yildiz, 2019).

Microbiological analysis

Microbiological studies were done to assess the safety. Total plate count (TPC) and yeast/mold counts were assessed. The microbiological examination of the juice was carried out according to the FDA's Bacteriological Analytical Manual's standard technique (FDA, 2001). In the case of nutritional agar medium, the pour plate technique was used to calculate the total plate count (TPC). In order to determine the total number of yeast and mould cells, we employed the PDA medium pour plate technique. The outcome was quantified in log colony-forming units per milliliter of juice (log CFU/mL) (Aadil *et al*., 2017). The microbial level of orange pulp was analyzed through Total plate count (Moussa and El-Gendy, 2019) and Yeast & mold count (Caminiti *et al.*, 2012). Microbial analysis was done at 0 days, 7 days, and 14 days.

Total plate count

Total plate count is the method used to measure the total amount of microorganisms present in the sample. It measures the total viable microbes present in the sample. Nutrient agar is used as a growth medium for total plate count. Growth media was prepared according to the standard protocol dissolving 14 grams of agar in 500 milliliters of distilled water with continuous mixing and heating on a hot plate for a while until the clear color appears and all the agar was properly dissolved in the distilled water. After that, the prepared media is tightly covered with aluminum foil. Similarly, peptone water was prepared according to the standard method after mixing peptone salt in the required amount of distilled water and then putting that graduated flask on a hot plate for some time until the color changed and peptone salt properly dissolved in the water.

All the required test tubes according to the different treatments and dilutions were first washed and dried then each test tube was filled with 9ml of peptone water and covered with aluminum foil. After that, all the test tubes filled with peptone water and nutrient media were kept in the autoclave for a period time of 15 minutes until the autoclave attains a temperature of 121 degrees Celsius. After the autoclave has been done and the temperature of the autoclave reaches below 80 degrees Celsius then the lid of the autoclave was opened so that the entrapped steam in the autoclave moves out. After the autoclave was done all the required material was placed in laminar airflow and growth media was poured onto the surface of the required Petri dishes. After that different dilutions were performed according to the required test tubes that were already marked according to treatment and dilution number. 1 ml from the sample was poured into the required test tube and a series of dilutions was performed after taking 1ml of solution from that test tube and pouring it into the next test tube.1 ml of the solution was taken from the test tube and poured into the Petri dish with culture media on it. After that, all the Petri plates were wrapped with the help of Pipette tape and kept in the incubator for 24 hours. The next day colonies were counted on the colony counter.

Calculation

Total plate count = Number of colonies × dilution factor

Yeast & mold count

Total yeast & mold count is used to detect and quantify the total amount of fungal growth on food materials. Potato dextrose agar was used as a growth medium for total yeast and mold count. Growth media was prepared according to the standard protocol dissolving 19.5 g of agar in 500 ml of distilled water with continuous mixing and heating on a hot plate for a while until the clear color appears and all the agar was properly dissolved in the distilled water. After that, the prepared media is tightly covered with aluminum foil. Similarly, peptone water was prepared according to the standard method after mixing peptone salt in the required amount of distilled water and then putting that graduated flask on a hot plate for some time until the color changed and the peptone salt properly dissolved in water.

All the required test tubes according to the different treatments and dilutions were first washed and dried then each test tube was filled with 9 ml of peptone water and covered with aluminum foil. After that, all the test tubes filled with peptone water and potato dextrose media were kept in the autoclave for 15 minutes until the autoclave attains a temperature of 121 degrees Celsius. After the autoclave has been done and the temperature of the autoclave reaches below 80 degrees Celsius then the lid of the autoclave was opened so that the entrapped steam in the autoclave moves out. After the autoclave was done all the required material was placed in laminar airflow and growth media was poured onto the surface of the required Petri dishes. After that different dilutions were performed according to the required test tubes that were already marked according to treatment and dilution number. 1 ml of the sample was poured into the test tube and a series of dilutions was performed after taking 1 ml of solution from that test tube and pouring it into the next test tube.1 ml of solution was taken from the test tube and poured into the Petri dish with culture media on it. After that, all the Petri plates were wrapped with the help of pipette tape and kept in the incubator for 24 hours. The next day colonies were counted on the colony counter.

Calculation

Total yeast and mold count = Number of colonies × dilution factor

Estimation of ascorbic acid

Ascorbic acid concentration of the control and treatment orange juice was determined using the AOAC Titrimetric Method. The ascorbic acid level of the treated orange juice was evaluated using redox titration, which comprises oxidation and reduction processes. Using 0.5 g/100 mL oxalic acid in a volumetric flask at 4 degrees Celsius, we diluted 15 milliliters

of known juice volume to 100 milliliters. A known volume of the aliquot (5 mL) filtrate was titrated with the 2,6 dichlorophenol indophenol indicator until the endpoint was attained. An ascorbic acid standard solution was titrated to an ordinary dye solution until a pink hue was formed, and this was used to compute the coloring factor. Ascorbic acid concentration in sample juice was determined using a concentration ratio of mg/100 mL (milligrams/milliliter) (Dias *et al*., 2015).

Principle

The indicator dye is reduced to a colorless solution by ascorbic acid. 2,6-dichlorophenol indophenol is rendered colorless by ascorbic acid. Ascorbic acid has a unique reaction at pH 1 - 3.5, which is only possible at this pH range. In acidic solutions, the indicator dye turns red, whereas in alkaline solutions, it turns blue. After filtering with (Whatman® No# 1 Filter paper), fruit juice was extracted from all samples. The quantity of ascorbic acid in the food sample is determined by the volume of titration used.

Preparation and standardization of dye

Soda benzoate, sodium dichlorophenol indophenol and the distilled water were measured out, dissolved in the final volume of 250ml, and mixed well. To produce another Oxalic acid solution, 1liter of distilled water was mixed with 4 grams of oxalic acid salt. Using 2ml of that ascorbic acid solution, an indicator dye solution was titrated against it until the appearance of a pink color lasting 15-20 seconds was attained.

Titration

A 30 ml sample from each treatment was individually mixed with 70ml of 0.4 oxalic acid solution. All these contents were transferred to a 100ml volumetric flask. 15ml of diluted beverage sample was collected after filtration using filter paper in another flask and titrated against indicator dye until the appearance of the pink color and that color persisted for about 15 seconds. Three consecutive values were taken for each sample.

Calculation

The following formula was used to calculate ascorbic acid:

$$\text{Vitamin C (mg/100mL)} = \frac{\text{Volume of sample used x Titre x Dye factor x 100}}{\text{sample reading x Aliquot of extract taken}}$$

Determination of Total phenolic and total flavonoid contents

TPC was measured according to the method described by Nadeem *et al.* (2018) with some slight modifications. Using the colorimetric Folin–Ciocalteu reagent, the TPC of an orange juice blend was determined spectrophotometrically. A 10 % Folin–Ciocalteu reagent solution (1 mL) was mixed with a known concentration of orange juice blend (0.5 mL). The mixture was thoroughly mixed, and a 6-minute stay time was given before adding 2 mL of a 20 % sodium carbonate solution to the mix. The combination was given a 60-minute stay time at 30 °C. Using a spectrophotometer, the TPC was measured at 760 nm wavelength. The results were expressed as microgram Gallic acid equivalents per gram (µg GAE/g) of each orange juice sample using a calibration curve made with a standard solution of Gallic acid.

TFC in orange juice was calculated following the method described by Aadil *et al.* (2013) with some slight modifications. The spectrophotometric method was used to determine TFC. Orange juice (0.25 mL) was mixed with a 5 percent sodium nitrate solution (75 µL) in deionized water (1.25 mL). After 6 minutes, 150 µl of a 10 % aluminum chloride solution was added, followed by 0.5 mL of 1 M sodium hydroxide after 5 minutes. The final volume of the mixture was adjusted to 2.5 mL with distilled water and thoroughly mixed. A spectrophotometer was used to measure the absorbance at 415 nm wavelength. TFC results were expressed as microgram catechin equivalents per gram of orange juice (µg CE/g).

Determination of DPPH-free radical scavenging activity

The orange juice DPPH free radical scavenging activity was evaluated using the procedure reported by Aadil *et al*. (2020). A 2 mL sample of juice was treated with 2 mL of DPPH solution and allowed to sit at room temperature (25 1 °C) for 30 minutes. The absorbance at 517 nm was measured using a spectrophotometer. In the same manner, the control (ethanol) was made. Reduced absorbance was caused by proton-donating activity. The decrease in juice mix absorbance was used to compute the percentage inhibition of DPPH radical scavenging activity:

Calculation

DPPH radical scavenging activity (%) = $100 \times (A_0 - A_1/A_0)$.

where A_1 is the absorbance of the orange pulp, while A_0 is the control absorbance.

Determination of total antioxidant capacity

The approach described in Aadil *et al*. (2020) was slightly modified in order to test antioxidant capacity in the samples. An ammonium molybdate, sulfuric acid, and sodium phosphate solution were added to the orange juice, each at a volume of 4 milli liters (0.4 mL). A water bath of 95 degrees Celsius was used to incubate this combination for 90 minutes. 4 ml of the reagent solution and 0.4 mL of methanol made up the blank solution. After cooling to room temperature (25°C), the spectrophotometer was used to measure the mixture's absorbance, which is related to proton donating activity. Standard

solution was ascorbic acid (100–400 g/mL), and total anti-oxidative activity was evaluated in ascorbic acid equivalents (g AAE/g). The calibration curve was constructed using this solution.

Sensory analysis

An evaluation of orange pulp consumer acceptability was carried out utilizing the 9-point Hedonic scale (Chauhan *et al*., 2015). An expert panel of from the department's faculty and graduate students, assessed the functional beverage samples for color, flavor, taste, and overall acceptance. Instructed to fill out a nine-point hedonic scale sensory sheet, the panelists were asked to record their impressions (9 points for extremely like and 1 point for extremely dislike).

Statistical Analysis

To evaluate the significance level, the data of every studied parameter was submitted to statistical analysis by following the approach of Montgomery (2017). The results of this investigation were expressed as the mean standard deviation (SD) (standard deviation). Factorial followed by two-way ANOVA (analysis of variance) at a significance level of ($p < 0.05$), and significant differences between mean values were determined using Statistix 9.0 software.

Results

Table. Mean values for pH of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	4.41	4.26	4.17	4.28
T_1 (Food additives)	4.44	4.38	4.31	4.37
T_2 (Ultrasound)	4.43	4.36	4.28	4.35
T_3 (US + Food Additives)	4.45	4.41	4.38	4.41
Mean	4.43	4.35	4.28	

Table. Mean values for TA of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	8.25	8.55	8.88	8.56
T_1 (Food additives)	8.27	8.42	8.54	8.41
T_2 (Ultrasound)	8.28	8.44	8.56	8.42
T_3 (US+ Food Additives)	8.29	8.37	8.48	8.38
Mean	8.27	8.44	8.61	

Table. Mean values for TSS of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	10.63	10.48	10.40	10.50
T_1 (Food additives)	10.62	10.54	10.45	10.53
T_2 (Ultrasound)	10.65	10.52	10.43	10.53
T_3 (US+ Food Additives)	10.66	10.62	10.54	10.60
Mean	10.45	10.54	10.64	

Table. Mean values for Ascorbic Acid of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	51.220	41.430	32.650	46.880
T_1 (Food additives)	51.230	45.920	38.660	45.270
T_2 (Ultrasound)	51.240	43.750	36.480	43.823
T_3 (US+ Food Additives)	51.250	48.670	40.720	41.767
Mean	51.235	44.943	37.128	

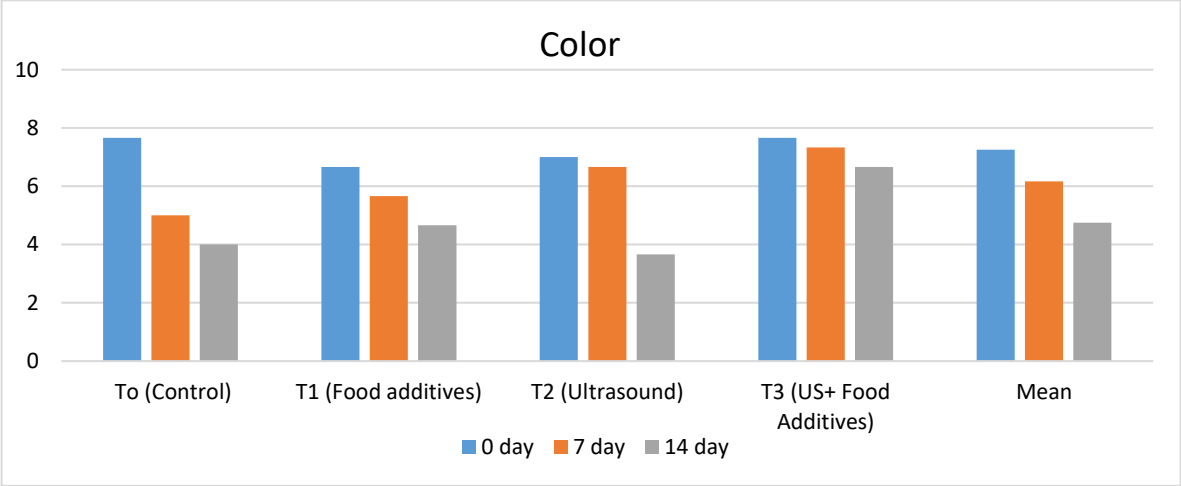
Table. Mean values for Total Plate Count of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	1.95	1.99	2.06	2.00
T_1 (Food additives)	1.98	2.05	2.13	2.05
T_2 (Ultrasound)	1.96	2.10	2.16	2.07
T_3 (US+ Food Additives)	1.95	2.02	2.10	2.02
Mean	1.96	2.04	2.11	

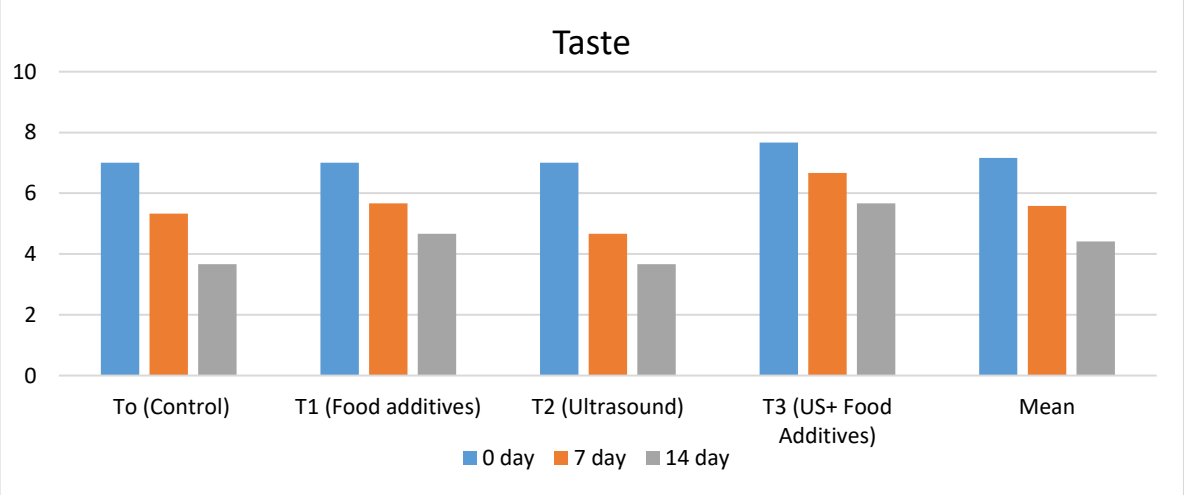
Table. Mean values for Yeast-Mould Count of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
$T_{o\ (Control)}$	4.26	4.98	5.88	5.04
$T_{1\ (Food\ additives)}$	4.28	4.62	5.64	4.84
$T_{2\ (Ultrasound)}$	4.26	4.77	5.67	4.90
$T_{3\ (US+\ Food\ Additives)}$	4.27	4.56	5.43	4.75
Mean	4.27	4.73	5.65	

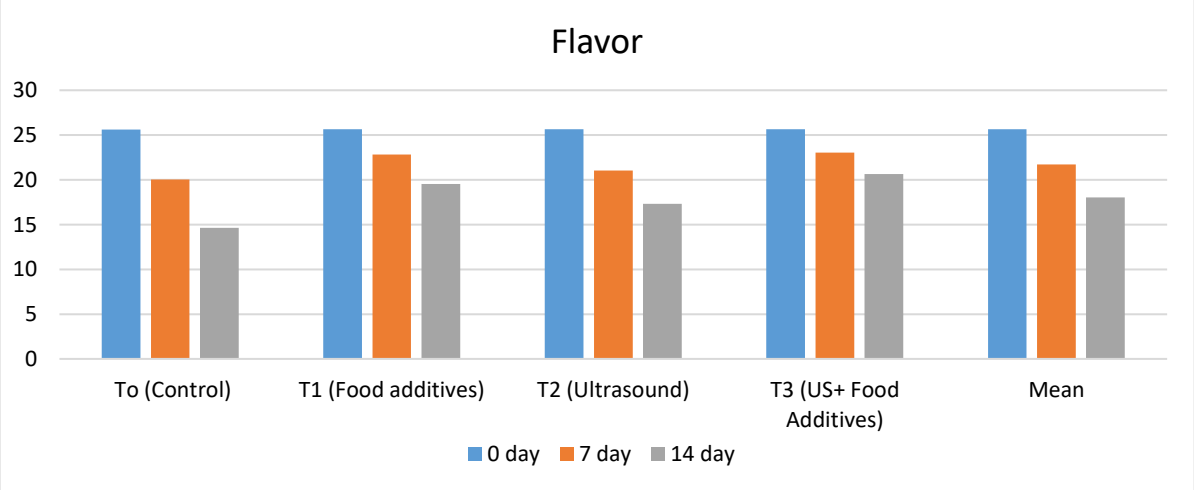
Mean values for Sensory Evaluation (Color) of orange pulp



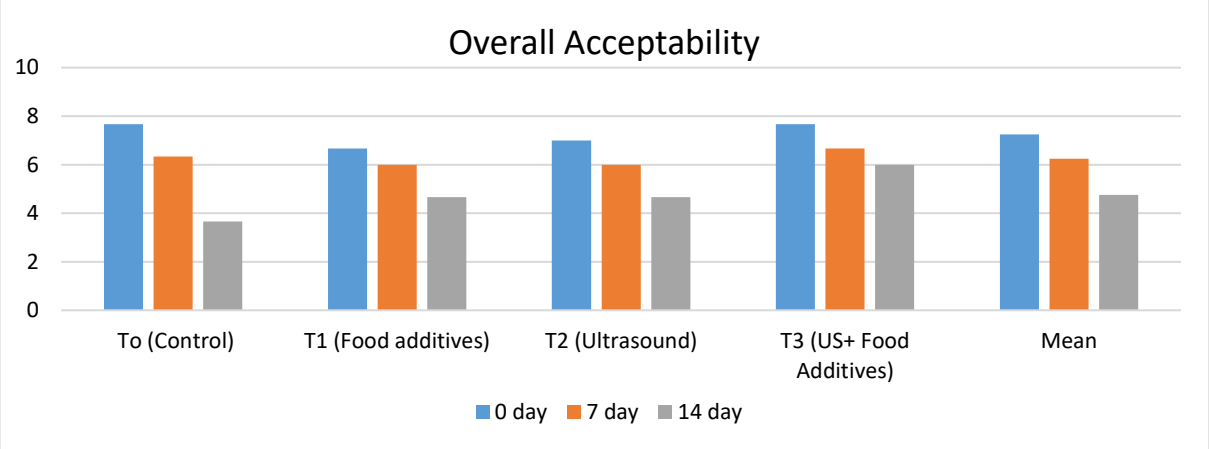
Mean values for Sensory Evaluation (Taste) of orange pulp



Mean values for Sensory Evaluation (Flavor) of orange pulp



Mean values for Sensory Evaluation (Overall Acceptability) of orange pulp



Mean values for Phenolic Content of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	230.24	212.44	191.03	211.24
T_1 (Food additives)	230.25	221.46	210.95	220.89
T_2 (Ultrasound)	230.26	218.37	206.78	218.47
T_3 (US+ Food Additives)	230.27	223.84	212.72	222.28
Mean	230.26	219.03	205.37	

Mean values for Total Flavonoid Content of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	25.620	20.020	14.650	20.097
T_1 (Food additives)	25.630	22.820	19.530	21.340
T_2 (Ultrasound)	25.640	21.050	17.330	22.660
T_3 (US+ Food Additives)	25.650	23.030	20.640	23.107
Mean	25.635	21.730	18.037	

Mean values for DPPH of orange pulp

Treatments	Storage Interval			Mean
	0	7	14	
T_0 (Control)	2.92	2.63	2.41	2.6533
T_1 (Food additives)	2.93	2.73	2.54	2.7333
T_2 (Ultrasound)	2.94	2.68	2.48	2.7
T_3 (US+ Food Additives)	2.95	2.75	2.62	2.7756
Mean	2.9350	2.6992	2.5125	

Discussion

It is clear from the data that the pH of Orange is affected by applying different treatments. The pH of Orange pulp varied significantly with respect to treatment. During 7 days of storage study the lower pH (4.26) was recorded in T_0 (industrial product) followed by T1(Additives) having pH of 4.38, whereas the higher pH (4.41) was found in T_3 (US+ Additives) followed by 4.36 in T_2(Ultrasound). Similarly, Bhat and Singh (2014) investigated the production and storage of Orange pulp determine the beverage's shelf life, which was pasteurized at various temperatures and times. It was found that the prepared drink's pH varied from 4.10 to 3.83 and 3.93 to 3.87 for the control and experimental drinks, respectively Moreover, (Maheswarlu *et al.*, 2010) revealed a decreasing trend in pH of pomegranate juice after 1 month of storage. The acidity of orange pulp varied significantly with respect to the storage intervals (Table). The lower acidity (8.25) was observed in treatment T_0 on 0^{th} day of storage whereas the higher acidity 8.88 was recorded on 14^{th} day of storage. It is cleared from results that titratable acidity of orange pulp was increased with the passage of storage intervals. Similarly, (Skryplonek *et al.*, 2019) demonstrated that the titratable acidity of beverages containing Lactobacillus acidophilus enhanced during storage, whereas the acidity of samples containing Bifidobacterium animalis was more constant.

It is cleared from the results that orange pulp has significant effect on the TSS. The TSS of orange pulp varied significantly with respect to treatment. During 7 days of storage study the lower TSS (10.48) was recorded in T_0 (control) followed by T_1 (food additive), T_2 (US) and T_3 (US + food additives) with TSS of 10.54, 10.52 and 10.62 respectively. (Bal *et al.*, 2014) conducted a research after preparing nectar from orange pulp and physicochemical attributes were determined after

every 2-month interval for 8 months. The results showed an increasing trend in TSS during the whole storage interval. It is clear from the data that different combinations of ultra-sound and food additives of orange pulp have significant effect on the vitamin C content of orange pulp. The vitamin C content of orange pulp varied significantly with respect to treatment. During 14 month of storage study the lower ascorbic acid content (51.220 mg/100g) was observed in T_o (industrial sample) followed by T_1 (food additive), T_2 (ultra-sound) and T_3 (ultra-sound and food additive) with vitamin C content of 51.230 mg/100g, 51.240 and 51.250 mg/100g respectively. Pandey and Ojha (2020) formulated a drink that mango pulp and discovered that as the proportion of pulp was lowered, the amount of Vitamin C and the reducing sugar was discovered to declined. Similarly, the proportion of ascorbic acid in juice decreased with the passage of storage. The concentration of ascorbic acid was decreased up to 65% during the 16th week of storage.

The total plate count of orange pulp varied significantly with respect to treatment. During 14 days of storage study lower total plate count (1.95 log 10 cfu/ml) was recorded in T_o (control) and T_3 (US + food additives) followed by T_2 (ultrasound) with total plate count of 1.96 log10 cfu/ml respectively. whereas the higher total plate count (1.98 log10 cfu/ml) was found in T_1 (food additives). Alane *et al.* (2017) Developed a beverage from mango and discovered that fresh sample contained 30 colony forming units/milliliter of TPC. After 30 days of storage, the sample was tested and found an increase in microbial load. The results of yeast and mold count of orange pulp for various treatment as well as storage intervals are showed in Table. It is clear from the results that different treatments of ultra-sonication and additives on orange pulp significantly impact on yeast and mold count of orange pulp. Log 10 was applied to all the colonies counted on colony counter. Yeast and mold count of orange pulp varied significantly with respect to treatment. During 14 days of storage study, lower yeast and mold count (4.26 log 10 cfu/ml) was recorded in T_0 (control) and T_2 (ultra-sonication treatment), followed by T_3 (4.27 log 10 cfu/ml) which was treated with ultra-sonication and along with the additives. The highest yeast mold count was recorded in T_1 (food additive treatment) at about 4.28 log 10 cfu/ml.

The phenolics in T_o (thermal) decreased from 230.24±0.01 to 191.03±0.01 on day 14. On day 0; T_1 (food additives), T_2 (US), T_3 (US + food additives), had 230.25±0.01, 230.26±0.01 and 230.27±0.01 of phenolics and had 210.95±0.01, 206.78±0.01 and, 212.72±0.01 at day 14. The decrease in the phenolics of orange pulp may be due to the sensitivity of phenolics to heat and storage conditions along with enzymatic activity (Yilmaz *et al.*, 2017). The TFC peak value was recorded with T3 during treatment (28.68), while, the value for T2 was 27.72. Similarly, minimum value of 26.63 was found at To. The maximum value was observed at 0 storage day (30.59), while minimum value (24.13) was observed at the relngeratron temperature at the time of storage, indicating that TFC contents decreased during storage time. The result revealed that sonication and microwave treatments had positive effect on the flavonoids contents. . The DPPH inhibition of the thermal (control) orange pulp was found to be 2.65. When ultrasound treatment was applied for T_1 (5 min) and T_2 (10 min), this value was significantly increased ($p < 0.05$) to 2.73 and 2.75, respectively. The cavitation process during ultrasound treatment caused a significant increase in DPPH values ($p < 0.05$). This increase in DPPH values can be attributed to the cavitation process during ultrasound treatment. Also, the application of Ohmic heating significantly increased ($p < 0.05$) the DPPH values as compared to the control value T_0. When Ohmic heating was applied for T_3 (120 V), the value of DPPH increased to 2.77.

The minimum value for color (3.66) was observed in T_2 (US) at 14th day of storage while maximum value for color was recorded during zero day of storage in T_3 (US + Food additives) which was 7.66 followed by T_1 (Food additive) with 7.00. Ribeiro *et al.*, 2009 Followed treatment with a combination of technologies (thermosonication and pulse electric field), the shelf life and sensory qualities of orange juice were assessed. Using a hedonic scale (1–9 points), 37 panelists evaluated the sensory qualities (i.e., color, odor, sweetness, acidity, flavor, and overall acceptability) of different orange juices treated with TS/PEF or HTST pasteurization. The lower score for taste (4.41) was observed on 14th day of storage whereas the higher taste value (7.16) was recorded on zero day of storage. It is evident from results that taste property of orange pulp showed a declined trend with the passage of storage intervals. Orange pulp's taste was unaffected by treatment and storage interval interactions. According to the findings, the flavor of orange pulp is not significantly affected by the different treatments. Hedonic scale was applied to all the treatments. Flavor of orange pulp varied significantly with respect to treatment. During 14 days of storage study lower points for flavor (5.11) were recorded in T_2 (US) followed by T_0 (control) and T_1 (Food additive) with flavor points on hedonic scale 5.33 and 5.77 respectively, whereas the higher points for flavor (6.66) was found in T_3 (US + Food additives). Overall acceptability of orange pulp varied significantly with respect to treatment. During 14 days of storage study lower points for overall acceptability (4.43) were recorded in T_0 (thermal) followed by T_1 (food additives) and T_2 (US) with overall acceptability points on the hedonic scale 3.6, 4.6, and 4.7 respectively, whereas the higher points for overall acceptability (6) was found in T_3 (US + food additives).

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