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Application of UV-C irradiation on the aonla ginger and sour orange blended Ready–to–serve (RTS) beverage

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ABSTRACT

Aonla - ginger-sour orange juice blended RTS was prepared and standardized. Sensory evaluation of the juices revealed that a sour orange concentration of 4 % was accepted widely with a maximum score of taste (8.7±0.58), texture (8.7±0.58) and overall acceptability (7.7±0.58). The juice was clarified at 30°C for 72 h to increase the initial microbial load. Ultraviolet irradiation was applied to the standardized aonla - ginger-sour orange blended RTS through a developed UV reactor at 8 kJ/L and 0.58 kJ/L dosage levels. At each reactor circulation, the microbiological population was assessed. Total plate count (TPC) and yeast and mould count (YMC), which contributed to the initial load of the RTS were found to be 5.45 log CFU/ml and 6.22 log CFU/ml, respectively. TPC exhibited 5.45 log reductions (commercial sterility) after the third circulation at a UV dose of 8 kJ/L. The TPC and YMC did not significantly change at the dose level of 0.58 kJ/L, irrespective of the number of circulations.

Keywords: *Phyllanthus emblica*, *Zingiber officinale*, *Citrus aurantium*, UV reactor.

INTRODUCTION

In India, one of the food businesses with the fastest growth is the beverage sector. Due to rising health consciousness among the general public, fruit-based beverages are growing in popularity. Fruit beverages are commonly prepared by reconstitution of fruit pulp with warm sugar syrup and subsequently subjected to pasteurization and the addition of preservatives to ensure microbial stability of the product towards extended shelf life (Gupta, 8). However, the thermal and chemical application in the production line hinders the full health benefits of fruit beverages. Hence, processing techniques were explored, including non-thermal application with minimum residual effects. Ultraviolet exposure to liquid foods is a recent advancement in eliminating microorganisms toward increased shelf life of fruit beverages.

In the electromagnetic spectrum, Ultraviolet radiation is usually categorized as Ultraviolet-A (320-400 nm), Ultraviolet -B (280-320 nm), and Ultraviolet-C (200–280 nm). The highest effective germicidal action has been reported to be at a wavelength of 253.7 nm in the UV-C zone (Bintsis *et al.*, 4). The formation of thymine dimers, which stop DNA transcription and replication when UV-C radiation penetrates cells through their membranes, damages DNA and triggers cell death (Rupasinghe and Yu,14).

UV-C irradiation, a non-thermal preservation method ensures the pasteurization of juices to the tune of 5 log reduction of the most resistant microorganisms (NACMCF, 12). The USDA and the USFDA have already approved UV-C irradiation as a safe method of pasteurizing juice.

Various UV-C reactor types for pasteurizing fresh juice have been evaluated (Koutchma, 11). The use of UV-C irradiation in treating milk and fruit juices is being researched, since it is a simple production method. Researchers examined the potentially lethal effect of UV-C irradiation on spoilage bacteria in the juices of apples, oranges, grapes, cranberry and grapefruit, strawberry and mango (Caminiti *et al.*, 5; Keyser *et al.* 10; Guerrero *et al.*, 7). Sew *et al.* (15) examined the effects of heat treatment combined with UV-C irradiation on pineapple juice and revealed that combination of UV and mild heat effectively inactivated the pectin methylesterase (PME) in pineapple juice preserving good amount of bromelain and phenols. Gayan *et al*. (6) conducted an identical experiment on orange juice, and found that 5 Log_{10} cycles of reduction in *Escherichia coli.* There is no investigation on RTS manufactured fromsour orange (*Citrus aurantium* L.), aonla (*Phyllanthus emblica*), and ginger (*Zingiber officinale*). Aonla is a good source of vitamin C; 100 ml of fresh juice with an aonla fruit contains 1 g of vitamin C. Collagen, which may bind the cells of the body together, is also made possible by vitamin C. It supports healthiness, and enhances the synthesis of red blood cells. Ginger has long been used to treat stomach problems,

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nausea, indigestion, and diarrhoea. It also has anti-inflammatory and antioxidant effects. Fever, diarrhoea, digestive issues and malarial fever can all be treated effectively with sour orange. Accordingly, the primary aims of the study were to: (1) standardize aonla -ginger blended RTS with sour orange juice; (2) develop a UV reactor for treating liquid foods; and (3) demonstrate the UV system efficacy in lowering the Total Plate Count (TPC) and Yeasts and Moulds Count (YMC) in the prepared aonla -ginger-sour orange blended RTS.

MATERIALS AND METHODS

The good quality fruits of aonla cv. Krishna were collected from the farm of CFDT, Koduveli, Chennai in 2021, washed and deseeded using aonla Deseeder. The deseeded fruits were ground using a pulverizer, and the juice was extracted by filtration through a muslin cloth. The sour orange (*Citrus aurantium* L.) fruits were purchased from the local market in Chennai, and cut half and then squeezed. The aonla and sour orange juices were collected in bottles, and kept at -20 °C in a deep freezer. Before making RTS, the deep-frozen juices were thawed. The frozen juice bottles were left to thaw for 4 h at room temperature.

The large and sound pieces of ginger were purchased from the local market, peeled off chopped into small pieces. The cut pieces were combined with two folds of water and ground using a mixer grinder. The juice and ginger bits were then separated from the slurry by transferring them to a muslin towel. The sour orange and aonla juices were blended with freshly made ginger juice as needed.

The quantity of aonla – ginger juices blended with sugar syrup (Table 1) as per the suggestions by Srivastava and Kumar (17). The acidity of the RTS was adjusted with the addition of sour orange juice (volume of sour orange 20 ml (2 %), 40 ml (4 %) and 60 ml (6%) for 1 litre of water). Sensory evaluation was conducted on 9 points Hedonic Scale by varying the concentration of Bitterorange juice (Amerine *et al*. 2). Organoleptic testing of the Aonla -Ginger blended RTS for the variable concentration of sour orange juice was undertaken by an eight-person panel with some training (Aggarwal and Gill, 1). The technological flow chart for the preparation of RTS is given in Figure.1.

Table 1. Ingredients for the production of RTS except for sour orange

S. No.	Aonla juice (ml)	Ginger juice (ml)	Sugar (g)	Water (ml)
1.	100	10	160	1000

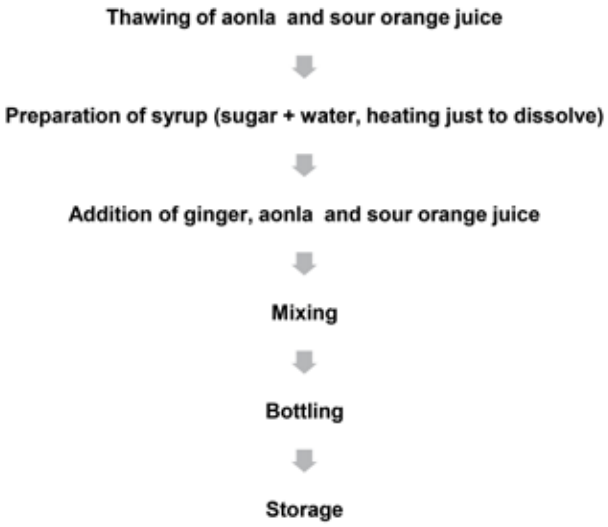

Fig. 1. Flow chart for the preparation of aonla -ginger- sour orange blended RTS

The prepared sugar syrup was filtered using a muslin cloth, and the blended Aonla, Sour orange and Ginger juices were again filtered using a muslin cloth. The juice blends were mixed with sugar syrup to prepare the final Aonla – Ginger - Sour orange RTS (Hamid *et al*. 9).

The developed UV reactor contained a tubular UV barrel, hopper and pipes connecting the UV chamber and hopper. The stainless steel outer cover of the UV barrel consisted of a UV lamp encompassed with quartz tube, inlet and outlet valves.

The hopper and UV chamber were constructed from 306 L stainless steel. The characteristics of the designed UV reactor are shown in Table 2. In addition, a diagrammatic representation of the designed UV reactor is given in Figure.2.

The prepared juice was exposed to UV irradiation through the developed UV reactor. Four litres of RTS beverage were prepared and poured inside the stainless steel hopper attached to the UV reactor. A ball valve regulated the juice flow rate in the pipeline between the hopper and the reactor. The developed

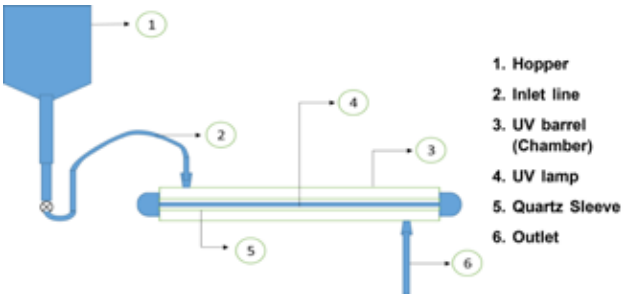

Fig. 2. The schematic view of the developed UV Reactor

Table 2. The specification of the developed UV reactor.

S. No.	Specifications	Dimension
1.	Diameter of the UV barrel	550 mm
2.	Internal diameter of the UV chamber	350 mm
3.	Diameter of a lampholder	350 mm
4.	Length of the UV reactor	290 mm
5.	The effective length of the UV reactor	250 mm
6.	Diameter of the UV lamp	15 mm
7.	Diameter of the quartz sleeve	19 mm
8.	Diameter of inlet and outlet valve	10 mm
9.	Height of the inlet and outlet valves	10 mm
10.	Area of the UV reactor	328 cm^2
11.	The power level of the UV lamp (output per unit)	16 W
12.	The volume of the UV reactor	0.25L
13.	The capacity of the hopper	4.5 L
14.	The maximum flow rate of the reactor	100 L/h
15.	Electrical adapter	220VAC input and 25W output

UV reactor was studied for minimum and maximum continuous flow. As a result, a minimum steady flow of 8 L/h without drop-by-drop flow and a maximum continuous flow of 100 L/h was observed.

The UV dosage for liquids is expressed as J L^{-1}. The intensity of the dosage can be measured using the following formula as suggested by Pala and Toklucu (13),

Intensity (I) = Total UV C output per unit (W) / Area (cm^2)

Dosage per volume = Total UV C output per unit (W) / Flow rate (L s^{-1})

Flow rate (L s^{-1}) = Volume of collection/ time to fill the volume

The following formula can be used to calculate the product retention time (T) to the reactor: Retention time in the UV reactor = T=V/F

Where,

V = Reactor volume (L); F = Discharge (L h-1)

Dosage = I × T

Where,

I = Intensity; T = Time

The dosage levels for the corresponding flow rates of 8 L/h and 100 L/h were 8kJ/L and0.58 kJ/L, respectively. The retention time for the RTS was 108 and 9 seconds for the flow rates of 8 L/h and 100 L/h, respectively. The RTS was subjected to three successive circulations at each dosage level. The samples were collected after each circulation to study the microbiological performance of the developed reactor. The RTS beverage not exposed to UV irradiation was considered a control sample.

The pour plate technique determined the total plate count from the control and Ultraviolet treated RTS samples. For 48 h, the replicate plates were incubated at 30°C. Next, the total yeast and mould count (YMC) was performed on potato dextrose agar for three days at 25°C. The outcomes were presented as colony-forming units per ml (Anonymous, 3).

The dependent and independent variables are given in Table 3. Duplicate replications of microbial counts and sensory scores were statistically analyzed using Microsoft Excel, 2013.

RESULTS AND DISCUSSION

For 1 litre of water, the quantity of sour orange juice varied from 20 ml (2%), 40 ml (4%), and 60 ml (6%). Therefore, the mean sensory scores of juices prepared from the varying concentration of sour orange juice are given in Table 4.

It was observed from Table 4 that a sour orange concentration of 4 % was accepted widely with a maximum score of taste (8.7±0.58), texture (8.7±0.58) and overall acceptability (7.7±0.58). Hence, the addition of 40 ml of sour orange juice with 1 L of water, 100 ml of aonla and 10 ml of ginger juice blend was standardized (Sharma *et al.*, 16). The standardized

Table 3. The dependent and independent variables with their levels.

S. No.	Independent variable	Levels	Dependent Variable
1.	Number of circulation	0, I, II and III	TPC and YMC (CFU/ml)
2.	Dosage	8 kJ/L and 0.58 kJ/L	

Table 4. Mean sensory score of RTS with varying concentrations of Sour orange juice.

Ingredients	The concentration of Sour orange juice	Colour (out of ?)	Flavour (out of ?)	Texture (out of ?)	Taste (out of ?)	Overall acceptability (out of ?)
10 % Aonla and 1 % of Ginger juice for 1 litre of water	2 %	7.7 ± 0.58	8.3 ± 0.58	8.2 ± 0.29	7.2 ± 1.26	7.2 ± 0.29
	4 %	8.2 ± 0.29	8.0 ± 0.50	8.7 ± 0.58	8.7 ± 0.58	7.7 ± 0.58
	6 %	8.2 ± 0.29	7.8 ± 0.29	7.7 ± 0.58	8.3 ± 0.58	7.3 ± 0.58

juice was used to evaluate the developed UV reactor in terms of microbiological performance by varying the number of circulation and UV dosages.

Juice samples analyzed for microbiological analysis were clarified at 30°C for 72 h to enhance the initial microbial load because the fresh aonla -ginger-sour orange blended RTS beverage had a minimum microbial load (Pala and Toklucu, 13). After the clarifying procedure, it was established that the initial loads of RTS with TPC and YMC were 5.45 log CFU/ml and 6.22 log CFU/ml, respectively. The YMC was found to be higher than the TPC. The lower pH (3.03) of the developed juice may be the source of the growth of YMC. Pala and Toklucu (13) noted a similar outcome for pomegranate juice.

The logarithmic reductions of TPC and YMC are given in Figure 3. UV-C dose of 8 kJ/L resulted in a 0.18 log reduction in TPC and 0.4 log reduction in YMC at first circulation. The TPC and YMC recorded a 0.24 log reduction and 0.54 log reduction at the second circulation of the RTS at 8 kJ/L, respectively. Finally, a 5.45 log reduction (commercial sterility) in TPC and a 0.75 log reduction in YMC were achieved after the third circulation at a UV dosage of 8 kJ/L. Guerrero *et al.* (7), who investigated the effects of UV-C on grape, cranberry, and grapefruit juices found the similar results in the reduction of YMC. The YMC showed resistant behaviour to UV-C radiation.

It was observed from Table 5 that a 12 % of reduction in YMC at the third pass was noticed at the dosage of 8 kJ/L, whereas the same third pass resulted in a 100 % reduction in the TPC. Table 6 depicted a gradual decline until the second circulation, and a sudden decrease of TPC at the third circulation at the dosage level of 8 kJ/L was also noticed. The reduction in the bacteria, yeast and mould population was due to the increased retention time at the reactor, when the flow (discharge) rate was 8 L/h to maintain the dosage of 8 kJ/L.

The lowest retention period was 9 seconds at the dosage level of 0.58 kJ/L with a maximum flow rate

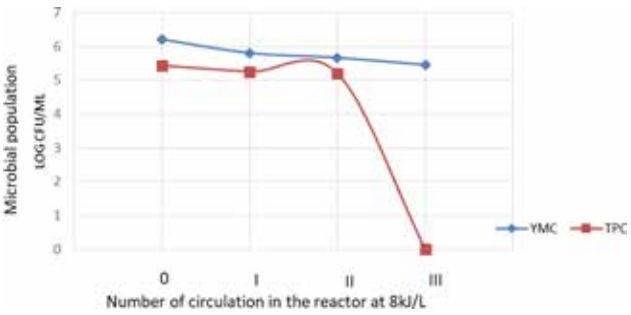

Fig. 3. Logarithmic reduction of TPC and YMC at different circulations.

Table 5. Yeast and Mould Count at the dosage level of 8 kJ/L.

Number of circulation	Mean population CFU/ml	Population in log10	Log reduction
0	165 × 10^4	6.22	-
I	65 × 10^4	5.81	0.40
II	47.5 × 10^4	5.68	0.54
III	29.5 × 10^4	5.47	0.75

Table 6. Total Plate Count at the dosage level of 8 kJ/L.

Number of circulation	Mean population CFU/ml	Population in log10	Log reduction
0	28 × 10^4	5.45	-
I	18.5 × 10^4	5.27	0.18
II	16 × 10^4	5.20	0.24
III	0	0	5.45

Table 7. Comparison between the flow rates.

Flow rate (L/h)	Dosage level (kJ/L)	Retention time (S)	Log reduction of TPC	Log reduction of YMC
8	8	108	5.45	0.75
100	0.58	9	-	-

of 100 L/h (Table 7). When the flow rate was set at 100 L/h, irrespective of the circulation, there were no decreases in the TPC or YMC. Results from Gayan *et al.* (6) and Pala and Toklucu (13) supported the findings.

The developed UV reactor established its capability to reduce TPC and YMC in the aonla -ginger-sour orange blended RTS. Higher retention time and low flow rate (8 L/h) resulted in a 5.45 log reduction of TPC, and a 0.75 log reduction of YMC. When the number of circulation increases, the significant influence of UV irradiation was observed on the decline of the microbial population. The yeast and mould were resistant to UV irradiation than the bacterial population. The higher dosage and number of circulations ensure the UV reactor's capability to be considered an alternative to a thermal pasteurizer.

AUTHORS' CONTRIBUTION

Conceptualization of research (NKSP); Designing of the experiments (NKSP); Contribution of experimental materials and Execution of field/lab experiments, data collection (NKSP, SP); Analysis of data and interpretation (NKSP, SP, KK); Preparation of the manuscript (NKSP, SP, KK).

DECLARATION

The authors declare no conflict of interest.

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