Quality Attributes of Ultrasound-Treated Prebiotic Fibre-Enriched Strawberry Juice

Mehr Un Nisa^{a,b}, Valente B. Alvarez^{b*}, and Muhammad K. I. Khan^{a,c}

^a National Institute of Food Science and Technology, University of Agriculture, Faisalabad-Pakistan

^b Department of Food Science and Technology, The Ohio State University, Columbus, Ohio-USA

^c Department of Food Engineering, University of Agriculture, Faisalabad-Pakistan

^{*}Corresponding author

alvarez.23@osu.edu

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Abstract

Strawberries are amongst the most desirable fruits with a rich flavour, appealing taste, high fibre content and many other health benefits. Diets rich in dietary fibre provide many health benefits. In the present work, strawberry juice was prepared in two batches, one with preservative sodium benzoate (treated) and another without (untreated). Prebiotic fibre i.e., apple pomace was added to both batches in concentrations of 5%, 8%, and 11%. Dietary fibre in both batches was analysed by enzymatic-gravimetric method. The additional analyses included pH, acidity, total soluble solids (TSS), colour, total phenolic content (TPC), antioxidant, ascorbic acid, anthocyanin, microbial and sensory parameters. Dietary fibre was increased significantly in all the treatments as well as TS, while pH and acidity were not affected. Ascorbic acid, anthocyanin, antioxidant, total phenolic content, and sensory analysis of treatment 2 (T_2 , 8% treated) showed the best results. The microbial load on the other hand increased more in the untreated batch. The T_2 treatment of both the batches was given ultrasound treatment. The sonication temperature (20 °C), frequency (20 kHz), and power (650 W) were kept the same, and the time was varied (0, 15, 30, 45, and 60 minutes). Dietary fibre showed a slight increase as fibre became more soluble by cavitation in sonication while pH acidity and TSS were not significantly affected. Anthocyanin increased, but only at lower sonication times. Antioxidants, total phenols, and colour and sensory parameters were significantly improved with sonication time. Similarly, the microbial load was reduced significantly.

Keywords: Strawberry juice; Prebiotic fibre; Apple pomace; Ultrasound; Nutrition; Stability

1 Introduction

Strawberry (*Fragaria X ananassa*) is highly nutritious and is one of the most consumed fruits of the Rosaceae family. It is the most popular berry with 20 species and 600 varieties with variations in size, colour, flavour, and texture (Hossain et al., 2016). It is preferred because of its aroma and juice texture. Botanist Carl Linnaeus named the genus Fragaria after the Latin word "Fragrans", meaning sweet-scented, and described it as a highly aromatic and striking fruit genus (Edger et al., 2019). Worldwide, strawberries are produced in 76 countries with China being the biggest producer followed by the USA, Mexico, Turkey, and Spain (Simpson, 2018). Strawberries contain antioxidants, minerals, vitamins, and other nutrients. The antioxidant potential of strawberries is high compared to other fruits because of their high vitamin C content and polyphenolic compounds (Mandave et al., 2017). Consuming strawberries

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can help prevent inflammation, cardiovascular diseases, obesity, cancers, and neurological diseases. Phenolics in strawberries can detoxify free radicals by blocking their production, modulate the gene expression involved in cell survival, metabolism, and proliferation and protect and repair overall DNA damage (Lakshmi et al., 2021).

Prebiotic fibre is defined as food ingredients that feed, grow, or activate beneficial microorganisms. It must possess three characteristics i.e., resists digestion and absorption in the upper gastrointestinal tract (small intestines), is fermented by the intestinal microflora and selectively stimulates the growth and/or activity of intestinal bacteria potentially associated with health and well-being. Prebiotics are different from dietary fibre in the specific stimulation of the microbiota (Davani-Davari et al., 2019). Previously known prebiotic fibre sources were carbohydrate compounds, mostly oligosaccharides but later inulin, oligofructose, lactulose, and resistant starch were also defined as prebiotic fibre. Similarly, there are other carbohydrate-containing compounds i.e., galacto-oligosaccharides (GOS), polydextrose, transgalacto-oligosaccharides (TOS), fructooligosaccharides, wheat dextrin, acacia gum, banana, and whole grain wheat that have the prebiotic effect (Carlson et al., 2018). Apple fibre obtained from apple pomace is identified as prebiotic fibre. The prebiotic fibre enhances the sensory quality and antioxidant capacity as well (Cassani et al., 2016).

Sonication treatments are of two types i.e., high intensity and low intensity. Low intensity is a non-destructive technique that is mostly used for the identification of unseen components and is used in food industries as a processing aid or for characterizing food components. High intensity is a material-altering technique that enhances or alters certain food processes (Huang et al., 2020). The frequency range of ultrasound enables its use in food analysis, food processing, and quality control. It enhances emulsification, promotes the extraction of food bioactive components, modifies the functional properties of food products, and acts as a preservation technique by inactivating enzymes (Bhargava et al., 2021). Ultrasound processing is widely

used for beverage processing to enhance flavour, stability, quality and shelf-life extension. It is a non-thermal processing technique, that maintains the stability of vitamins and minerals present in fruits, thus preserving the nutritional value as well (Alqahtani et al., 2014).

Adding dietary fibre in beverages is an alternative approach to address the fibre gap. However, adding fibre presents challenges in terms of juice organoleptic, sensory and overall quality. Therefore, the present work is aimed at improving the nutritional value of strawberry juice by adding apple fibre (prebiotic fibre) from apple pomace to increase fibre content and to evaluate the effect of ultrasound on the quality and shelf life of fibre-enriched juice.

1.1 Rationale and significance

The growing awareness among global consumers about the health benefits associated with a diet rich in fibre has prompted significant attention from food scientists. Despite not contributing to calorie intake, dietary fibre exerts various physiological effects on the health of the human body. This has led to a notable interest in fibre-based, low-calorie food products, which have become integral components of people's daily diets. Incorporating dietary fibre into beverages serves multiple purposes, both from a technological and a nutraceutical perspective. It can function as a thickening, gelling, dispersing, and carrying agent, while also offering potential benefits such as improving glucose tolerance, reducing blood cholesterol levels, and lowering the risk of colon and other types of cancers. Beverages serve as excellent vehicles for nutrient supplementation, efficiently delivering essential nutrients to different parts of the body when consumed with meals (Ahmad & Ahmed, 2019).

In terms of nutrition, increasing the fibre content of foods is a good strategy to add more fibre to the daily diet. The Institute of Medicine recommends that healthy adults consume 25 g of daily fibre for women and 38 g of daily fibre for men. Fibre recommendations for children and the elderly are 14 g of fibre for every 1000 cal (kcal) consumed. But the current dietary fibre intake is less than 50% of the recommended level

(Snauwaert et al., 2023).

To mitigate this issue, fibre is being added to food products such as breakfast cereals, bread, yogurt, beverages, and snacks. In this sense, enrichment of the beverages with dietary fibre is gaining importance. However, the addition of fibre has some limitations and challenges due to its effect on sensory properties and stability of the product (McGill et al., 2016). These challenges are related to phase, separation, sedimentation, sensory aspects, undesirable texture, and, importantly, the visual appearance of the final product. Therefore, there is a lot of interest in adding more fibre to food products as well as utilizing a processing technique that could maintain the overall quality and shelf life of fruit beverages.

2 Materials and methods

2.1 Apple pomace preparation

The apples were procured from the local market and were selected based on their physical appearance i.e., colour, shape, size, and absence of damage. Apple pomace was obtained by pressing 2 kg of whole apples with a manual juice extractor and separating them from the juice. The pomace was placed in zip lock bags and stored in a refrigerator at 4 o C for further use in the preparation of fibre-enriched strawberry juice.

2.2 Preparation of fibre-enriched strawberry juice

The strawberries were placed in a strainer and washed with potable water. Strawberries (1 kg) were blended in a blender (Black Decker power crush multifunctional blender) with water (1 litre) until pureed, then transferred to a pan. More water (1 litre) was added to the puree. Then sugar (280 g) and citric acid (2.5g) were added and heated until it reached the pasteurization temperature (72 o C). The juice was cooled to 30 o C and sodium benzoate (1g) was added as a preservative. Another batch of juice was prepared following the same procedure but with no preservative. Both batches of strawberry juice were formulated with apple pomace at 0 % (T₀), $5 \% (T_2), 8 \% (T_3), \text{ and } 11 \% (T_4).$ The juices

were analysed for physicochemical, sensory, and microbial properties. The best treatment of juice was selected for ultrasound treatment based on its good nutritional and sensory profile.

2.3 Ultrasound treatment

The best-selected juice was sonicated using the JY-96-IIN Ultrasonic Homogenizer (Ningbo Scientz Biotechnology Co., Ltd., China). The sonication temperature (20 °C), frequency (20 kHz), and power (650 W) were kept the same and only the time was varied (0, 15, 30, 45, and 60 minutes) to treat juice samples.

2.4 Physicochemical analysis

The fibre-enriched strawberry juices and sonicated juices were analysed for various physic-The fibre was deochemical parameters. termined by enzymatic-gravimetric method by Megazyme assay kit K-TDFR. The pH was measured with an Inolab-720 pH meter (WTW, Germany). A hand refractometer RFM-340 (UK) was used for recording total soluble solids (TSS) value. Titratable acidity was determined by the standard titration method (AOAC International, 2006). Ascorbic acid was determined by titration with 2,6-dicholoroendophenol (Ordóñez-Santos et al., 2017). Total phenolic content (TPC) was measured by spectrophotometer U-2020 (IRMECO, Germany) according to the Folin-ciocalteu method (Ordóñez-Santos et al., 2017). Antioxidants were measured by the DPPH method (Pérez-Grijalva et al., 2018). Anthocyanin was also determined by spectrophotometer Model-U2020 (IRMECO-Germany) (Kapoor et al., 2015). The colour was determined using a colorimeter COLORTEST-II (Neuhaus Neotec, USA). Readings for L^{*} (Lightness), a^* (+ redness, - greenness) and b^* (+ vellowness, - blueness) were recorded.

2.5 Microbial analysis

Total plate count (TPC) was determined by the spread plate method and the results were expressed in colony forming units (cfu) (Amelia

et al., 2018).

2.6 Sensory evaluation

Sensory evaluation of all samples was done by randomly selected participants using the 9-point Hedonic scale ranging from 1 (extremely dislike) to 9 (extremely dislike). The juices were stored in plastic bottles and assigned random numbers for the participants. Sensory parameters including colour, flavour, taste, and overall acceptability of samples were measured (Stone et al., 2020).

2.7 Statistical Analysis

All experiments were conducted in triplicate and results were subjected to statistical analysis using two-way ANOVA and the difference among means was determined using a significance level of $\rho < 0.05$ (Montgomery, 2017).

3 Results and Discussion

In the first phase, apple pomace as prebiotic fibre was added to the strawberry juice in different concentrations (0, 5, 8 and 11 %) for fibre enrichment and was analysed for physiochemical, microbial, and sensory parameters. The samples T_0 , T_1 , T_2 , and T_3 were divided into two main groups i.e., treated (T), and untreated (UT) based on the addition of preservative. From the first phase the juices with the highest score of analysis were taken for the second phase of the study i.e., ultrasound application. The treatments were prepared with varying sonication times (0, 15, 30, 45, and 60 minutes) to analyse their effect on physiochemical, bioactive, sensory, and microbial parameters and the overall quality of fibre-enriched strawberry juice.

3.1 Phase-I: Effect of apple pomace addition

Dietary fibre

The results showed a highly significant effect of apple pomace addition to strawberry 62 Nisa et al.

juice. Increasing the concentration of apple pomace increased the dietary fibre content from $2.11\pm0.01\%$ to $12.66\pm0.01\%$ (p < 0.05) in prebiotic strawberry juice (Table 1). Strawberry juice has a dietary fibre content of 2g per serving (Giampieri et al., 2012) while in apples it is 8.19 g per serving (Macagnan et al., 2015). These results similar to the findings of (Tanska et al., 2016), who formulated food products with apple pomace and found increased dietary fibre content. Both the treated and untreated samples showed similar increase in dietary fibre content irrespective of preservative addition.

pH, Acidity, and TSS

TSS values increased from 15.98 ± 0.01 to 17.08 ± 0.01 (p < 0.05) in treated and untreated samples as shown in Table 1. However, the effect on acidity and pH was non-significant. Issar et al. (2017) reported comparable results in yogurt enriched with apple pomace.

Ascorbic acid

The ascorbic acid content increased with pomace addition with maximum increase in T2. The treated juice proved more stable and maintained higher ascorbic acid content during storage while that of untreated juice decreased, as shown in Figure 1a (p < 0.05). It is reported that ascorbic acid (vitamin C) is heat labile and sensitive to chemical and enzymatic oxidation when processed and stored. The external factors such as temperature of storage, light and oxygen can also accelerate oxidation of ascorbic acid (Cassani et al., 2018). The untreated and treated samples both showed the same pattern of reduction in ascorbic acid content. However, preservative tends to reduce the speed of loss. The results of the present work resembled the findings of Zou and Jiang (2016).

Total phenolic content

The results of TPC showed that adding apple pomace increased the phenolic content from 117.93 ± 0.01 to 140.99 ± 0.01 mg GAE/100mL (p < 0.05) for T₀ to T₂, respectively then it dropped

Table 1: Physicochemical, sensory, and microbial properties of prebiotic fibre-enriched strawberry juice. Treatments T_0 , T_1 , T_2 and T_3 : treated with 0, 5, 8 and 11 % apple pomace respectively. T – preservative added, UN – no preservative added. Means within rows with different superscripts (a – d) differed significantly (p < 0.05).

Parameters		Treatments							
		TO		T1 (5%)		T2(8%)		T3(11%)	
		т	UN	т	UN	т	UN	т	UN
Dietary fibre (%)		$2.11 {\pm} 0.01^d$	$2.11 {\pm} 0.01^d$	$12.42{\pm}0.01^{c}$	$12.42{\pm}0.01^{c}$	12.56 ± 0.01^{b}	12.56 ± 0.01^{b}	$12.62{\pm}0.01^{a}$	$12.62{\pm}0.01^{a}$
pH		3.47 ± 0.01^{d}	3.47 ± 0.01^{d}	3.71 ± 0.01^{c}	3.51 ± 0.01^{c}	3.71 ± 0.01^{b}	3.55 ± 0.01^{b}	3.71 ± 0.01^{a}	3.59 ± 0.01^{a}
Acidity (%)		0.27 ± 0.02^{a}	0.27 ± 0.02^{a}	0.26 ± 0.02^{b}	0.26 ± 0.02^{b}	0.22 ± 0.02^{ab}	0.22 ± 0.02^{ab}	0.21 ± 0.02^{ab}	0.21 ± 0.02^{ab}
TSS (^o Brix)		17.99 ± 0.01^{d}	15.98 ± 0.01^{d}	18.17 ± 0.01^{c}	17.6 ± 0.01^{c}	18.23 ± 0.0^{a}	18.5 ± 0.01^{a}	18.08 ± 0.01^{b}	17.08 ± 0.01^{b}
Total phenolic conten	nt (mg $GAE/100mL$)	120.02 ± 0.03^d	117.93 ± 0.03^{d}	137.12 ± 0.05^{c}	$122.04{\pm}0.05^{c}$	$150.88 {\pm} 0.02^{a}$	$140.99 {\pm} 0.02^{a}$	140.05 ± 0.04^{b}	130.99 ± 0.04^{b}
Antioxidant activity	(% DPPH assay)	$67.96 {\pm} 0.05^c$	$64.96{\pm}0.05^c$	74.95 ± 0.05^{b}	68.03 ± 0.05^{b}	$75.06 {\pm} 0.04^{a}$	$68.88 {\pm} 0.04^a$	73 ± 0.05^{a}	68 ± 0.05^{a}
Colour	L*	$68.04{\pm}0.03^d$	$67.59 {\pm} 0.03^d$	$77.50{\pm}0.02^{c}$	$77.14{\pm}0.02^{c}$	$84.24{\pm}0.02^{b}$	84.00 ± 0.02^{b}	$98.89{\pm}0.03^a$	$92.35 {\pm} 0.03^{a}$
	a*	$76.68 {\pm} 0.01^{a}$	87.03 ± 0.01^{a}	46.48 ± 0.02^{b}	56.53 ± 0.02^{b}	$42.54{\pm}0.02^{c}$	52.02 ± 0.02^{c}	38.47 ± 0.04^{d}	$43.92{\pm}0.04^{d}$
	b*	50.36 ± 0.02^{a}	56.08 ± 0.02^{a}	28.53 ± 0.03^{d}	31.04 ± 0.03^{d}	33.90 ± 0.01^{c}	34.01 ± 0.01^{c}	49.50 ± 0.03^{a}	54.02 ± 0.03^{a}
Sensory	Colour	8.56 ± 0.03^{a}	8.54 ± 0.03^{a}	7.67 ± 0.02^{b}	7.82 ± 0.02^{b}	7.52 ± 0.02^{c}	7.20 ± 0.02^{c}	7.50 ± 0.04^{d}	7.14 ± 0.04^{d}
	Flavour	7.05 ± 0.04^{c}	7.03 ± 0.04^{c}	7.81 ± 0.02^{b}	7.51 ± 0.02^{b}	8.52 ± 0.02^{a}	8.52 ± 0.02^{a}	7.74 ± 0.03^{b}	7.55 ± 0.03^{b}
	Texture	7.64 ± 0.03^{c}	7.54 ± 0.03^{c}	8 ± 0.02^{b}	7.94 ± 0.02^{b}	8.64 ± 0.03^{a}	8.54 ± 0.03^{a}	7.54 ± 0.03^{d}	7.04 ± 0.03^{d}
	Overall acceptability	7.64 ± 0.03^{d}	7.55 ± 0.03^{d}	$7.84{\pm}0.02^c$	$7.77{\pm}0.02^c$	$8.54{\pm}0.02^a$	$8.04{\pm}0.02^a$	7.83 ± 0.03^{b}	7.94 ± 0.03^{b}



Figure 1: Ascorbic acid content (mg/100mL) of fibre-enriched strawberry juice (a) and sonicated juice (b)

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Parameters					Trea	atments					
		T0		T1		T_2		T3		T4	
		Т	UN	Т	UN	Т	UN	Т	UN	Т	UN
Dietary fibre (%)		$12.56{\pm}0.02^{b}$	$12.56 {\pm} 0.02^{b}$	13.56 ± 0.0^{a}	$13.56 {\pm} 0.02^{a}$	$15.46 {\pm} 0.02^{b}$	$15.46 {\pm} 0.02^{b}$	$17.6 {\pm} 0.02^{c}$	$17.6 {\pm} 0.02^{c}$	$19{\pm}0.02^c$	$19{\pm}0.02^c$
Total phenolic content (m	g GAE / 100 mL)	$150.88 {\pm} 0.02^{e}$	$140.99{\pm}0.02^{e}$	$214.07{\pm}0.02^{d}$	198.65 ± 0.02^{d}	$228.04{\pm}0.02^{c}$	$210.26 {\pm} 0.02^c$	$239.54 {\pm} 0.03^{b}$	227.05 ± 0.03^{b}	$249.54{\pm}0.03^{a}$	$235.04{\pm}0.03^{a}$
Antioxidant Activity (% I	OPPH assay)	$75.06 {\pm} 0.02^{e}$	$68.88 {\pm} 0.02^{e}$	$86.57{\pm}0.02^d$	$80{\pm}0.02^d$	$86.98 {\pm} 0.02^{c}$	$81.03 {\pm} 0.02^c$	$87.02 {\pm} 0.03^{b}$	$82.13 {\pm} 0.03^{b}$	$87.03 {\pm} 0.03^a$	$83.55 {\pm} 0.03^{a}$
Colour L*		$84.24{\pm}0.02^{b}$	$81.03 {\pm} 0.02^{b}$	84.25 ± 0.02^{a}	82.29 ± 0.02^{a}	$84.30 {\pm} 0.03^{b}$	$83.99 {\pm} 0.03^{b}$	$84{\pm}0.02^{b}$	$84.21 {\pm} 0.02^{b}$	$83.25 {\pm} 0.02^c$	$84.35 {\pm} 0.02^{c}$
a*		$42.54{\pm}0.02d$	52.0 ± 0.022^{d}	$52.34{\pm}0.02^{b}$	52.50 ± 0.02^{b}	52.29 ± 0.02^{a}	52.89 ± 0.02^{a}	$51.39{\pm}0.02^{c}$	$52.38 {\pm} 0.02^c$	$51.39 {\pm} 0.02^c$	$52.35{\pm}0.02^{c}$
*а		$33.90{\pm}0.01^{e}$	$34.01{\pm}0.01^{e}$	$34.66 {\pm} 0.02^a$	$34.90 {\pm} 0.02^{a}$	$34.64{\pm}0.02^{c}$	$34.64{\pm}0.02^{c}$	$34.14{\pm}0.02^{d}$	$34.75 {\pm} 0.02^d$	$34.69 {\pm} 0.02^{b}$	$34.68{\pm}0.02^{b}$
Sensory Col	our	7.52 ± 0.01^{d}	7.50 ± 0.01^{d}	$8.12{\pm}0.03^{c}$	$8.13{\pm}0.03^{c}$	$8.24 {\pm} 0.01^{b}$	8.23 ± 0.01^{b}	$8.44{\pm}0.01^{b}$	$8.42{\pm}0.01^{b}$	$8.55 {\pm} 0.02^{a}$	$8.52 {\pm} 0.02^{a}$
Flav	vour	$8.52{\pm}0.02^{e}$	$8.13 {\pm} 0.02^{e}$	$8.64{\pm}0.02^{d}$	$8.22{\pm}0.02^{d}$	$8.72 {\pm} 0.02^c$	$8.33 {\pm} 0.02^{b}$	$8.81 {\pm} 0.02^{b}$	$8.46 {\pm} 0.02^{b}$	$8.91{\pm}0.02^{a}$	8.56 ± 0.02^{a}
Tex	ture	$8.64{\pm}0.03^{d}$	$8.54 {\pm} 0.03^{d}$	$8.66 {\pm} 0.02^{d}$	$8.59 {\pm} 0.02^{d}$	$8.81 {\pm} 0.02^{c}$	8.74 ± 0.02^{c}	$8.91{\pm}0.022^{b}$	$8.87 {\pm} 0.02^{b}$	$8.97{\pm}0.02^{a}$	$8.94 {\pm} 0.02^{a}$
Ονε	rall accept ability	$8.54{\pm}0.03^{e}$	$8.04 {\pm} 0.03^{e}$	$8.42{\pm}0.02^d$	$8.13{\pm}0.02\mathrm{d}^d$	$8.44{\pm}0.02^c$	$8.23 {\pm} 0.02^{c}$	$8.44 {\pm} 0.01^{b}$	$8.42 {\pm} 0.01^{b}$	8.55 ± 0.02^{a}	$8.52 {\pm} 0.02^{a}$

Table 2: Physicochemical, sensory, and microbial properties of prebiotic fiber-enriched strawberry juice after sonication. Treatments T0, T1, T2, T3 and T4: sonicated for 0, 15, 30, 45 and 60 minutes respectively. T - preservative added, UN - no preservative added. Means within rows with different superscripts (a – d) differed significantly (p < 0.05)



Figure 2: Anthocyanin content (mg P3G/mL) of fiber-enriched strawberry juice (a) and sonicated juice (b)

in T_3 in untreated samples. Similarly in treated samples, the phenolic content increased and followed the same pattern, from 120.02 ± 0.03 to $150.88 \pm 0.02 \text{ mg GAE}/100 \text{mL} (\text{p} < 0.05) \text{ for } \text{T}_0$ to T_2 (Figure 2a). The reason for this was that in T_3 (in both treated and untreated samples) the concentration of pulp became higher than the water content, the solid portion settled out and phase separation of water and solids happened. The total phenolic content of treated samples was higher compared to untreated because of the presence of preservative. The preservative inhibited the growth of spoilage microorganisms that can reduce the concentration of phytochemicals (Ahmed et al., 2021). Similar findings were reported by (Tomadoni et al., 2016).

Antioxidant activity (DPPH assay)

The concentration of apple pomace and preservative both affected the antioxidant content of the juice. The control (T₀) had a value of 67.96 ± 0.01 % DPPH assay, which increased to 75.06 ± 0.01 % DPPH assay (p < 0.05) in T₂ due to increasing apple pomace content that also has higher antioxidants. The treated samples showed higher antioxidant values as compared to the untreated samples (Table 1) because the preserva-

tive inhibited microbial spoilage. Tomadoni et al. (2016) used untreated juice and two preservatives, vanillin and geraniol, and reported similar results.

Anthocyanin

Anthocyanins are the colour pigment that contribute to the increase in the organoleptic properties of fruit juices. The vibrant colour of strawberry juice is an indication of a high level of anthocyanins. Quantitatively, anthocyanins are the most important polyphenols in strawberry. The main anthocyanin in strawberry is pelargonidin-3-glucoside and in apple pomace it is cyanidin-3-O-galactoside (Sirijan et al., 2020). In the fortified juice, the anthocyanin content is contributed by both the strawberry fruit and apple pomace. The anthocyanin content of juices increased by increasing apple pomace concentration (Figure 2a). However, the highest increase of anthocyanin 51.02 ± 0.02 mg P3G/mL and $50.01\pm0.02 \text{ mg P3G/mL}$ (p < 0.05) was observed in T₂ in treated and untreated sample respectively. The addition of preservative helped to maintain juice stability so higher content was observed in treated samples. According to Teribia et al. (2021) the preservative prevented oxidation

or browning and stabilized the colour for longer times.

Colour

The addition of apple pomace affected the red colour of strawberry juice. In T_0 the colour of pure strawberry juice was red, but with increasing the concentration of apple pomace, the colour began to change. Because the apple pomace is brown in colour, it decreased the intensity of the red colour of strawberry juice. The a* (redness) value and L* (lightness) values decreased while b* (yellowness) values became higher with the increasing concentration of apple pomace. More change was observed in untreated samples as oxidative browning occurred rapidly in apple pomace, while the preservative delayed browning for a longer period. The results were similar to the findings of (Mir et al., 2017).

Total plate count

The addition of apple pomace and preservative both affected the microbial count. In T_0 the microbial load was $1.5 \ge 10^4 \pm 0.02$ that increased to 4.3 x $10^4 \pm 0.02$ cfu/mL (p < 0.05) in T₃ in untreated sample indicating that increasing the concentration increased the total plate count significantly. However, in treated samples the microbial load decreased from $1.4 \ge 10^4 \pm 0.02$ in T_0 to 1.9 x 10³±0.03 cfu/mL (p < 0.05) in T_3 . The preservative reduced the microbial population (Figure 3a). The preservative inhibits the microbial population by its effect on the cytoplasmic membrane integrity of microbes resulting in loss of ion gradients and pH homeostasis that negatively affect their activity. Similar results were reported by (Tomadoni et al., 2016).

3.2 Phase-II: Effect of sonication

The best-selected treatment 8% of the pomace was subjected to sonication with (P1) and without (Po) preservative and results are discussed in the following sections. The effect of sonication on pH, acidity, and TSS was non-significant thus results are not shown in table 2. Similar results were obtained in cape gooseberry juice (Ordóñez-Santos et al., 2017).

Dietary fiber

Sonication significantly improved the fiber content from $12.66\pm0.01\%$ to $19\pm0.01\%$ from T₀ to T₄ respectively (Table 1). The sonication effect on the fiber content of prebiotic juice was due to the phenomenon of cavitation. The mechanical effect of ultrasound improved the solubility of components and in this case, the fiber present in apple pomace became more soluble. Moreover, an increase in sonication time increased the fiber content (Table 1) because of the release of soluble compounds due to the disruption of cells. The results of the present research were similar to the literature (Cassani et al., 2018).

Ascorbic acid

Initially, when sonication treatment was given, it showed an increase in ascorbic acid content. But further increasing the application time gradually decreased the ascorbic acid content in both treatments as shown in Figure 1(b). Still, it was higher than the control but less than T_1 . The degradation of ascorbic acid was due to collapsing of bubbles or due to the formation of oxidative products like hydrogen ions (H+), hydrogen peroxide (H_2O_2) , and free radicals on the surface of bubbles and the production of hydroxyl radicals due to cavitation. The treated sample showed better results (Figure 1b). So, sonication for lower times can improve the ascorbic acid content. The present work was similar to the findings of (Aguilar et al., 2017).

Total phenolic content

Sonication conditions such as long time, high power, and low frequency improved the bioactive compounds i.e., phenolic content. In this study the frequency was kept low at 20 kHz, therefore the increase of phenolic content occurred. Increasing the sonication time increased the phenolic content due to the release of hydroxyl radicals during cavitation as described above. The concentration values of phenolic content in treated samples with preservative were higher because the preservative inhibited microbial activity that causes the reduction of bioactive compounds (Figure 2b). The result of the



Figure 3: Total plate count (cfu/mL) of fibre-enriched strawberry juice (a) and sonicated juice (b)

present work was similar to the findings of (Yikis, 2020).

Antioxidant potential

The results of treated and untreated samples sonicated at different times indicated that the DPPH radical scavenging activity of fiberenriched strawberry juices increased with a longer sonication time. Regarding preservative addition, higher values from 75.06 ± 0.01 to 87.03 ± 0.01 (% DPPH assay) were obtained in treated samples as compared to the untreated samples i.e., 68.88 ± 0.01 to 83.55 ± 0.01 (% DPPH assay) (Table 2). Similar results reported that at low ultrasound power at a constant temperature, the DPPH radical scavenging activity increased (Khandpur & Gogate, 2015). The antioxidant activity increased with increasing sonication time, due to an increase in polyphenolic content and anthocyanin because of cavitation. Increasing sonication time increased the concentration of antioxidants released (Khandpur & Gogate, 2015).

Anthocyanin

Sonication time also increased the anthocyanin concentration up to a certain time. At a shorter time, the anthocyanin content increased due to the release of bound anthocyanin in the pulp. When a long time is applied, anthocyanin degradation occurs due to the presence of other organic acids (Figure 2b). The results are supported by the findings of (Radziejewska-Kubzdela et al., 2020).

Total plate count

As the sonication time increased the microbial load started to decrease due to the phenomenon of mechanical cavitation. Sonication, at a lower frequency and shorter times, is insignificant but as time increased, a significant reduction in microbial load occurred as shown in Figure 3b, similar to results reported by (Bhat & Goh, 2017).

3.3 Sensory evaluation

In Phase-I, T₂ showed the most liked because of the optimal percentage of apple pomace. Similarly, treated juice was preferred over untreated. The formulations with high levels of apple pomace received lower scores for colour ($\rho < 0.05$) which is associated with brown colouration of apple pomace. There was not much difference in the flavour profile. However, the texture and overall acceptability scores were different. The sensory scores were analysed statistically and were related to chemical analysis results. As T₂

showed better results for total phenols, antioxidants and anthocyanin that impacted the colour and flavour profile of juice. That might be the reason that the panel preferred the T_2 juice in both treated and untreated sample as shown in Table 1, which was similar to Silva et al. (2021). In phase-II, the treated and untreated mean did not show much variation. Sonication reduced the microbial activity thus no browning occurred. prevented the development of off flavour, improved texture with increasing time by preventing separation and fractionation of prebiotic fibre. The preservative in the presence of sonication treatment had a non-significant impact on the juice texture. The sensory results showed better acceptability of both the juices. The sensory parameters i.e., colour, flavour, texture and overall acceptability were not significantly different as shown in Table 2. The results were similar with the findings of Cassani et al. (2018).

4 Conclusions

Strawberry juice enriched with apple pomace showed improved nutritional profile and sensory acceptability with T_2 being the best treatment with good content of apple pomace. The treated samples showed better results as compared to the untreated samples as the preservative prevented microbial spoilage and loss of bioactive components. Similarly, sonication improved physicochemical, microbial, and sensory parameters with increasing time of sonication in both treatment batches. Further research could be conducted by changing ultrasound power and temperature and measuring their effect on product quality.

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