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Effect of Sugar Beet Fiber and Inulin on Survival and Activity of *Lactobacillus acidophilus*, Chemical and Sensorial Properties of Pineapple Synbiotic Drink

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Abstract

Probiotics, especially lactic acid bacteria, are very useful in the production of health drinks, and increasing the nutritional value of beverages. Probiotic bacteria along with prebiotics are used to produce synbiotic drinks. In this study, the effect of inulin at three levels (1, 2 and 3%), sugar beet fiber (0.75, 1.5 and 2%) and bacteria (*Lactobacillus acidophilus*) in the production of synbiotic pineapple juice was investigated. The treatments were stored at 4 °C for 25 days and tests for pH, bacterial viability, brix, colorimetry and sensory evaluation were performed at 5-day intervals. With increasing the amount of inulin, sugar beet fiber as well as shelf life, bacterial viability and red-green color (a*) increased, and the values of brix, pH, transparency (L*) and yellow-blue (b*) decreased. The results showed that storage time is one of the most important factors affecting the studied parameters. Based on the results of Brix, pH, colorimetric (L*, b* and a*) and bacterial viability on day 25, treatments containing 3% inulin and 0.75% sugar beet fiber were 19.17%, 3.6, 68.91, 14.3, -2.19, 12.28 CFU/g, respectively and was selected as the ideal treatment.Therefore, it can be concluded that in order to produce a useful pineapple drink, *Lactobacillus acidophilus* can be used with inulin and sugar beet prebiotics.

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Keywords
Inulin
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Synbiotic

Introduction

Nowadays, the acceptance and consumption of functional food products in the world is on the rise so that different additives are used to enhance the functional of food products. These food types have a special nutritional and medicinal value; thus, the consumption of foods containing beneficial microorganisms, called probiotics, significantly help the survival and maintenance of native intestinal microbes and therefore have many benefits for human health. The products containing a mixture of probiotic and prebiotic bacteria are called synbiotics leading to positive effects on the body (Champagne, Ross, Saarela, Hansen, & Charalampopoulos, 2011; Lin, Hwang, Chen, & Tsen, 2006; Tripathi & Giri, 2014). Prebiotics are indigestible compounds selectively fermented and

cause marked changes in the composition and activity of microorganisms like bifidobacteria and lactobacilli in the gastrointestinal tract that are very beneficial to the host health. In the food industry, these compounds are usually used to meet technological goals and enhance the nutritional value of the product (Akusu, Kiin-Kabari, & Ebere, 2016; Wang, 2009). Inulin is one of the prebiotics, which is a linear polysaccharide with beta-bonds, one to two fructoses (Canbulat & Ozcan, 2015; Tárrega, Rocafull, & Costell, 2010), with a polymerization degree from 2 to 60 and in two commercial forms: short chain inulin with 2-7 units and long chain with more than 30 units of oligofructose. The solubility and intensity of inulin hydration decreases with increase in chain length or degree of polymerization; thus, they are used to improve and thicken beverage texture (Arcia, Navarro, Costell, & Tárrega, 2011). Sugar beet pulp is another prebiotic that is a by-product of sugar factories making up about 4-5% of the fresh weight of sugar beet root (Poel, Schiweck, & Schwartz, 1998) and contains 1.1% fat, 8.92% protein, 3.72% ash and 86.26% polysaccharides, of which 22-30% are cellulose (Toğrul & Arslan, 2003). Most of the probiotics belong to the gut flora and the family of lactic acid-producing bacteria like *Lactobacillus acidophilus*, *Lactobacillus. plantarum*, *Lactobacillus casei* and *Sterptococos lactis* (Shah, Dave, & Roghelia, 2016; Zoghi, Khosravi-Darani, Sohrabvandi, Attar, & Alavi, 2017). Lactic acid-producing bacteria ferment inulin and produce compounds like acetate, propionate, butyrate, and lactate, involved in regulating cellular metabolism, especially division and differentiation. Synbiotic compounds increase the growth and survival of probiotic bacteria in the large or small intestine by acting on them. Studies on animal models indicated that bifidobacteria and fructooligosaccharides alone cannot prevent bowel cancer, but their simultaneous use in synbiotic products can prevent bowel cancer. Synbiotics outperform probiotics and prebiotics and strengthen the immune system better than them, which shows a synergistic effect between probiotics and prebiotics (Goderska, Czarnecka, & Czarnecki, 2007). Pineapple, scientifically called Comanus Ananas, belongs to the Bromeliaceae family, a perennial herbaceous plant native to Brazil and Colombia, with only one genus, Pitcairnia felicana, native to West Africa. It has long lanceolate and leathery leaves with longitudinal stripes of cream and white and red spines (Nguyen, Bujna, Fekete, Tran, Rezessy-Szabo, Prasad, & Nguyen, 2019). This plant has prickly and pleasant fruits that use its water and flesh in the production of canned food and its leaves in the production of textiles. Pineapple fruit contains vitamins A, C, B, calcium, phosphorus, iron, citric and malic acids, and a proteolytic enzyme called bromelain or bromelain (Costa, Fonteles, de Jesus, & Rodrigues, 2013). The studyhas used sugar beet fiber and inulin to prepare the synbiotic drink pineapple juice.

Materials and methods

Preparation of treatments

Lyophilized *L. acidophilus* was obtained from DSM Co., Australia and activated in 20 ml of MRS broth (Merck Germany) medium under anaerobic conditions (anaerobic jar and gas pack type C) and temperature of 37 °C for 24 h and in the next step, the activated bacteria from the first culture were inoculated in 95 mL of liquid MRS culture medium and propagated under the same conditions. Bacterial growth biomass was isolated by centrifuge (DM0412, Chinese Dragonlab) at 1500 rpm for 15 min at 25 °C and combined in two steps with 0.1% sterile saline solution (Mahdian, Mehraban, Karazhian, & Vaghei, 2014). Sugar beet pulps prepared from sugar factory were stored in the freezer at -18 °C until consumption and were cleaned by hand and dark parts removed and homogenized

in a mixer with 96% ethyl alcohol at the time of consumption after defrosting to make the extract of smoothing completely colorless. Then it was placed in an oven at a temperature of 50 ℃ for 12 h until its humidity reached 9-11%. The dried pulp was ground, and finally screened with a 250-300 micron mesh sieve (Özboy & Köksel, 2000). Pineapple drink is prepared from Sanich Factory and then short chain inulin with 8 degree of polymerization and 99.5% purity (Roosendaal company, Netherlands) is added in proportions of 1, 2 and 3% to the pineapple juice sample (Natural concentrate, Sanich Factory) and and then beet fiber at levels of 0.75, 1.5 and 2% was added to the beverage samples using a homogenizer (Iran, Fan Azma) in homogeneous conditions (60, C, MPa20) and homogenized. Ultimately, the juice containing inulin and sugar beet fiber prebiotics was enriched with *L. acidophilus bacterium* with a population of 10^6 CFU/mL and the produced synbiotic juice was kept at 4 ℃ for 25 days (Table 1), qualitatively evaluated by microbial, chemical and sensory tests at 5-day intervals.

Table 1. Coding the treatments

Treatment code	*L. acidophilus* (CFU/mL)	Sugar beet fiber (%)	Inulin (%)
In1Be0.75	10^6	0.75	1
In1Be1.5	10^6	1.50	1
In1Be2	10^6	2.00	1
In2Be0.75	10^6	0.75	2
In2Be1.5	10^6	1.50	2
In2Be2	10^6	2.00	2
In3Be0.75	10^6	0.75	3
In3Be1.5	10^6	1.50	3
In3Be2	10^6	2.00	3
Control	10^6	0.00	0

Experiments

pH, brix and colorimetry evaluation

pH and brix values of synbiotic fruit juice were measured by pH meter made (PL-700PC model, Taiwan Gondo Co.) and digital desktop refractometer (model PR-32-α, Atago Japan), respectively, during storage during days 0, 5, 10, 15, 20 and 25. All the experiments were performed in three replications (Iranian Standardization Organization national [ISIRI], 2008) and from Hunterlab colorimeter (Colorimeter, Minolta CR-400, Japan) was used to evaluate the color of fruit juice and to measure the parameters a* (green-red), b* (blue-yellowness) and L* (transparency).

Microorganisms' survival assessment

MRS agar medium was used to count *L. acidophilus* using purplate method with dilutions of 10^{-1} to 10^{-6}, temperature 37 °C‹, duration 48 h (Yoon, Woodams, & Hang, 2004).

Sensory evaluation

Sensory evaluation of fruit juice samples was done by 5-point hedonic method and the treatments were evaluated for properties like color, taste, texture and general acceptance by 20 untrained evaluators. The scores for the very good, good, average, bad and very bad samples as 5, 4, 3, 2 and 1, respectively (Krasaekoopt & Kitsawad, 2010).

Statistical analysis

SPSS version 24 and Two-way ANOVA test were used to compare means and Excel software 2016 was used to draw graphs to study the effect of storage time, inulin and sugar beet fiber values on the quality characteristics of pineapple juice and symbiotic.

Results and discussion

Microorganisms' survival

One of the major indices in the production of synbiotic juices is the viability of probiotic bacteria in these products. The results of the effect of variables like storage time, inulin and sugar beet values on the survival of *L. acidophilus* bacteria in Table (2) showed that with increase in storage time, the number of *L. acidophilus* increased and the highest growth rate in the treatment of 2% inulin and 0.75% sugar beet fiber has been seen. Inulin and sugar beet provide the conditions for the

growth of these bacteria by providing energy and organic acids. The control treatment had the lowest bacterial viability. Although there were no significant differences (P>0.05) between the control treatment and the treatments containing inulin and sugar beet fiber up to day 10 of storage, in other storage times there was a difference between samples containing inulin and sugar beet fiber with control treatment (P<0.05). Contrary to the results of this study, Zoghi *et al.* (2017) and some researchers on the survival and growth of probiotic bacteria Such as *L. acidophilus*, *L. plantarum* have found that with increasing the shelf life, growth and number of these bacteria have decreased for some reason (Nualkaekul & Charalampopoulos, 2011; Shisheh, Hashemiravan, & Pourahmadjaktaji, 2014; Tripathi & Giri, 2014; Zoghi *et al.*, 2017). Factors affecting the survival and activity of bacteria in probiotic juices are dietary parameters such as pH, acidity, oxygen, aqueous activity, the presence of salt and sugar, chemical and artificial flavors and colors, and process parameters like pasteurization, cooling rate, closed materials, storage methods, oxygen level and volume and microbiological factors like bacterial strains and their incubation ratio (Tripathi & Giri, 2014).

Brix value

According to Fig. (1), the value of soluble solids (Brix˚) in all treatments decreased with increase in storage time and a significant difference (P<0.05) was established. The effect of adding sugar beet fiber and inulin on soluble solids in the treatments examined was irregular and the treatment of 2% inulin and 2% sugar beet fiber had the highest value of Brix compared to other treatments during the storage period. The growth and activity of *L. acidophilus* during storage ends in the consumption of bacterial substrates and reduces Brix (Costa *et al.*, 2013; Tárrega *et al.*, 2010). The value of brix in pineapple juice with probiotic *L. casei* has been reported in the range from 17 to 22, where the Brix value decreased in pineapple juice samples over time, which is in line with the results of this study. Moreover, with the results of grape juice containing *L. paracasei*, the Brix value in the control treatment was higher than other treatments. Increasing inulin and sugar beet fiber values led to an increase in soluble solids in the samples, yet with the growth of *L. acidophilus* bacteria in the environment, the fermentation process started and reduced the value of sugars in the product, ending in reduced Brix (Silva & Ferrari, 2016). Such results were seen in Totonchi, Hesari, Moradi, & Fathi (2015) and Shaykhgasemi & Zomorodi (2014) in probiotic drinks of red grapes and probiotic apple juice.

Table 2. The effect of storage time, sugar beet percentage and inulin on bacterial viability (Log CFU/mL) *L. acidophilus*

Treatment	Storage time (days)					
	0	5	10	15	20	25
In3Be2	6.01±0.1 ^{Ae}	8.62±0.52^{DEd}	10.3±0.32 ^{Dec}	12.3±0.35^{Ab}	13. 05±0.9^{Aab}	14±1.02^{ABa}
In3Be1.5	6.1±1.22 ^{Ae}	9.7 ±0.25 ^{Bd}	10.75±0.25^{CDcd}	11.12±0.25^{BBC}	12.4±0.35^{Bab}	13.06±0.15^{Ca}
In3Be0.75	6.1±1.02 ^{Ad}	8.32 ±0,05^{EFd}	9.99± 0.42 ^{Ec}	11.21±0.25^{Bb}	11.99±0.6^{BCab}	12.0±28.34^{EFa}
In2Be2	6.0±1.5 ^{Ae}	9.0±37.42^{BCd}	12.0± 94.28^{Aa}	11.0±07.31^{BCc}	11.0±74.8^{BCbc}	12.0±79.3^{DEa}
In2Be1.5	6.0 ±0.12 ^{Ae}	8.51±0.23^{EBd}	10.09±0.61^{DEc}	11.12±0.8 ^{BCab}	11.93±0.28^{BCab}	12.05±0.9F^{Ga}
In2Be0.75	6.0±0.52 ^{Af}	10.26±0.82^{Ae}	11.68± 0.42^{Bd}	12.1±3.2^{Acd}	13.45±0.9^{Ab}	14.52±0.85^{Aa}
In1Be2	6.01±0.21^{Ae}	7.8 ±0.2^{Ghd}	9.38± 0.5 ^{EFc}	10.34±0.72^{CDbc}	11.39±0.34^{Cab}	11.58±0.5^{Ga}
In1Be1.5	6.0±1.22 ^{Af}	9.23±0.32^{BCe}	10.72±0.52^{CDd}	10.86±0.45^{Ccd}	12.08±1.2^{BCab}	12.92±0.7^{CDa}
In1Be0.75	6.0±0.14 ^{Ad}	7.93 ±0.2 ^{FGc}	8.0± 64.35^{FGbc}	8.0±82.28^{EFb}	9.37±0.5^{DEab}	9.9±0.3^{HIa}
Control	6.0±0.1 ^{Ae}	7.74 ±0.28^{Hd}	8.37± 0.3 ^{Gc}	7.52±0.6^{Fbc}	7.06±0.6^{Eb}	6.68±0.2^{Ia}

* The difference between uppercase letters shows a significant difference (P<0.05) in the column and the difference of lowercase letters a significant difference (P<0.05) in the row (Mean ± standard deviation, three repetitions).

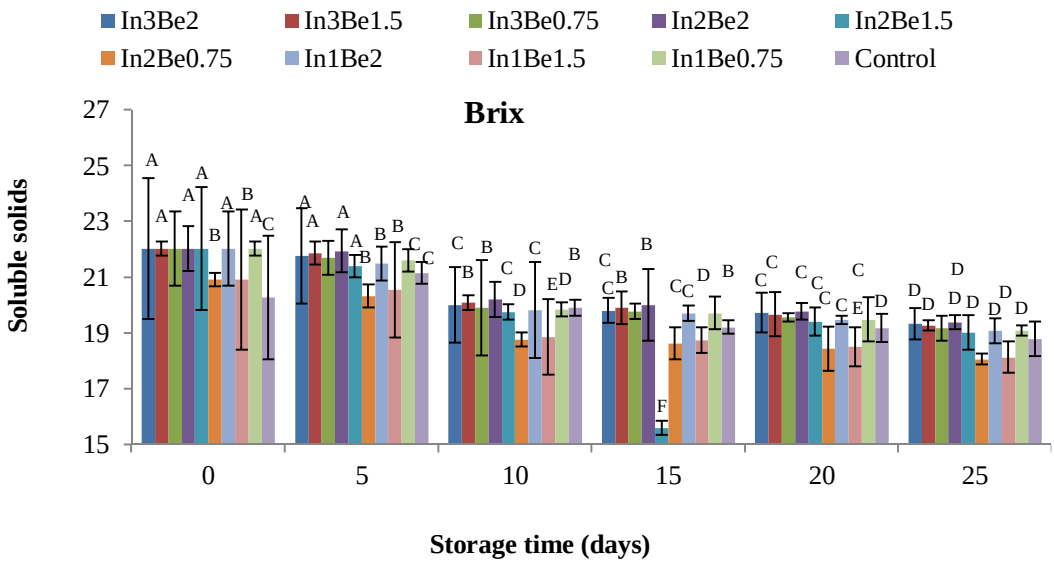

Fig. 1. The effect of storage time, percentage of sugar beet fiber and inulin on soluble solids (BX°) of pineapple juice

pH analysis

According to Fig. (2), with increase in storage time, pH decreased in all treatments and the control treatment had the highest pH during the storage period. The difference between control and other treatments except day zero was significant (P<0.05) on other storage days. Treatments with equal values of inulin increased the pH with decrease in the value of sugar beet fiber. The cause of decreasing pH and increasing acidity is the growth of *L. acidophilus* in all treatments containing inulin and sugar beet. Such results have been observed in research into the production of fermented bean juice with *L. delbrueckii* and *L. plantarum*, as well as the production of tomato probiotic drinks with lactobacilli (Yoon, Woodams, & Hang, 2006). Decreased pH during storage has been seen in probiotic beverages like grape juice, apple juice, carrot milk and kale containing two bacteria *L. plantarum* and *L. delbrueckii* (Shaykhgasemi & Zomorodi, 2014; Silva & Ferrari, 2016; Yoon *et al.*, 2006).

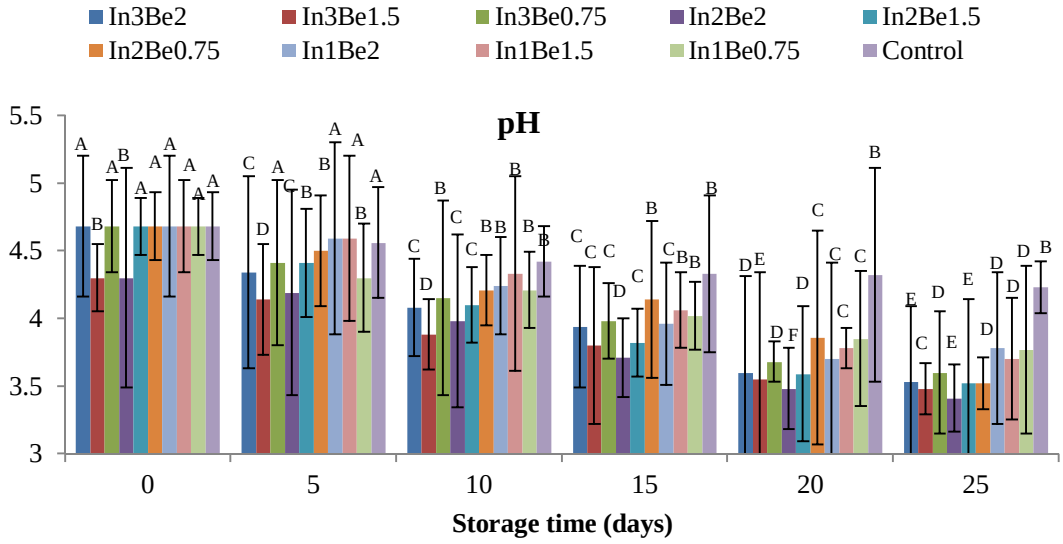

Fig. 2. The effect of storage time, sugar beet content and inulin on the pH of synbiotic pineapple juice

Color evaluation

With increase in storage time, transparency (L*) and yellow-blue (b*) indices according to the reduced Tables (3) and (4), the control treatment without inulin and sugar beet fiber had the highest value of L* and b*, but the amount of color red-green (a*) that has increased with increasing storage time, according to Table (5). The lowest value of a* index was observed in the control treatment.

Table 3. The effect of storage time, sugar beet and inulin percentage on the transparency (L*) of synbiotic pineapple juice

Treatment	Storage time (days)					
	0	5	10	15	20	25
In3Be2	77.3 ± 1.1^{Ca}	76.09 ± 0.2^{Bab}	74.5 ± 1.2^{Dc}	72.02 ± 0.5^{BEd}	68.6 ± 0.21^{EFe}	67.9 ± 0.15^{Eef}
In3Be1.5	77.3 ± 0.12^{Ca}	$76.\ 6\pm0.52^{Bab}$	75.4 ± 1.4^{CDb}	72.7 ± 0.42^{DEc}	69.2 ± 0.24^{DEde}	68.4 ± 0.23^{DEe}
In3Be0.75	77.3 ± 2.2^{Ca}	75.4 ± 1.30^{Bb}	74.7 ± 0.85^{Dbc}	72.3 ± 1.20^{Ec}	69.6 ± 0.12^{Dde}	68.9 ± 0.35^{De}
In2Be2	77.6 ± 1.15^{BCa}	73.5 ± 0.92^{Bbc}	72.9 ± 1.02^{Ec}	70.3 ± 0.35^{Fd}	67.3 ± 0.54^{Fef}	66.6 ± 0.27^{Ff}
In2Be1.5	80.2 ± 1.2^{ABa}	77.4 ± 1.85^{Bbc}	76.1 ± 1.10^{Bc}	73.7 ± 0.38^{CDd}	70.6 ± 1.20^{Cef}	69.8 ± 0.52^{CDf}
In2Be0.75	80.1 ± 2.5^{ABa}	79.3 ± 2.1^{Aab}	78.2 ± 1.2^{Ab}	76.06 ± 1.5^{Ac}	72.5 ± 0.32^{ABde}	71.8 ± 0.25^{ABe}
In1Be2	80.1 ± 1.29^{ABa}	77.8 ± 1.35^{Bbc}	76.5 ± 3.1^{Bc}	74.5 ± 0.61^{BCd}	70.8 ± 0.31^{Ce}	70.1 ± 0.35^{BCe}
In1Be1.5	80.6 ± 1.4^{Aa}	79.02 ± 1.5^{ABab}	75.07 ± 0.48^{CDcd}	74.4 ± 1.03^{BCd}	72.7 ± 1.2^{Aef}	71.3 ± 1.4^{ABf}
In1Be0.75	80.1 ± 1.14^{Ba}	77.7 ± 1.3^{Bbc}	76.7 ± 1.5^{Bc}	74.6 ± 1.1^{BCd}	71.09 ± 1.3^{BCef}	70.3 ± 1.20^{Bf}
Control	80.2 ± 3.1^{Ba}	77.8 ± 0.4^{Bbc}	76.4 ± 1.31^{Bc}	74.4 ± 0.34^{BCd}	72.5 ± 0.71^{ABef}	72.1 ± 0.41^{Af}

* The difference between uppercase letters shows a significant difference (P<0.05) in the column and the difference of lowercase letters a significant difference (P<0.05) in the row (Mean ± standard deviation, three repetitions)

Table 4. The effect of storage time, percentage of sugar beet and inulin on the amount of yellow-blue (b*) color of pineapple juice

Treatment	Storage time (days)					
	0	5	10	15	20	25
In3Be2	15.1 ± 0.10^{Da}	14.7 ± 0.20^{Db}	14.5 ± 0.20^{Cb}	14.1 ± 0.30^{Ec}	13.4 ± 1.2^{Bd}	13.3 ± 0.25^{Bd}
In3Be1.5	16.0 ± 0.12^{BCa}	15.5 ± 0.20^{BCbc}	15.2 ± 0.20^{Bc}	14.7 ± 0.25^{Dd}	14.1 ± 1.2^{Bef}	13.9 ± 0.70^{Bf}
In3Be0.75	16.0 ± 0.21^{BCa}	15.8 ± 0.15^{Bab}	15.6 ± 0.50^{ABab}	15.2 ± 0.35^{ABc}	14.5 ± 0.4^{Bde}	14.3 ± 0.20^{Be}
In2Be2	16.0 ± 0.15^{BCa}	15.8 ± 0.41^{Bbc}	15.3 ± 0.40^{Bc}	14.9 ± 1.20^{CBd}	14.1 ± 0.34^{Bef}	14.03 ± 0.52^{Bf}
In2Be1.5	16.1 ± 0.20^{Ba}	15.8 ± 0.20^{Bab}	15.09 ± 0.20^{Bcd}	14.8 ± 0.28^{CDde}	14.5 ± 0.6^{Bef}	14.27 ± 0.20^{Baf}
In2Be0.75	16.0 ± 0.28^{BCa}	15.5 ± 0.31^{Bbc}	15.36 ± 0.18^{Bc}	14.9 ± 0.90^{Cde}	14.24 ± 0.24^{Bae}	14.07 ± 0.51^{Be}
In1Be2	16.1 ± 0.29^{Ba}	15.8 ± 0.80^{Bab}	15.6 ± 0.22^{ABb}	15.0 ± 0.45^{Bac}	14.38 ± 0.28^{Bde}	14.24 ± 0.32^{Be}
In1Be1.5	16.1 ± 0.22^{Ba}	15.9 ± 0.20^{ABab}	15.73 ± 0.20^{ABab}	15.2 ± 0.25^{ABc}	14.5 ± 0.18^{Bde}	14.41 ± 0.32^{Be}
In1Be0.75	16.1 ± 0.15^{Ba}	15.7 ± 0.20^{Bab}	15.6 ± 0.37^{ABb}	15.1 ± 0.21^{Bc}	14.62 ± 0.29^{Bde}	14.45 ± 0.20^{Be}
Control	16.5 ± 0.10^{Aa}	16.2 ± 1.10^{Aab}	15.84 ± 0.38^{Acd}	15.6 ± 0.40^{Ad}	15.08 ± 0.70^{Aef}	15 ± 0.32^{Bf}

* The difference between uppercase letters shows a significant difference (P<0.05) in the column and the difference of lowercase letters a significant difference (P<0.05) in the row (Mean ± standard deviation, three repetitions)

Table 5. The effect of storage time, percentage of sugar beet and inulin on redness - vegetable (a*) pineapple juice synbiotic

Treatment	Storage time (days)					
	0	5	10	15	20	25
In3Be2	-3.1 ± 0.15^{Aad}	-3.02 ± 0.4^{Aae}	-2.83 ± 0.28^{Aad}	-2.59 ± 0.5^{Aac}	-2.19 ± 0.3^{Aab}	-2.15 ± 0.6^{Aa}
In3Be1.5	-3.12 ± 0.25^{Aad}	-3.4 ± 0.15^{Aad}	-2.86 ± 0.2^{Aacd}	-2.62 ± 0.23^{cA}	-2.2 ± 0.34^{Aba}	-2.18 ± 0.4^{Aa}
In3Be0.75	-3.2 ± 0.14^{Aad}	-3.2 ± 0.1^{Aad}	-2.84 ± 0.2^{Adc}	-2.6 ± 0.2^{Ac}	-2.24 ± 0.2^{Aab}	-2.19 ± 0.2^{Aa}
In2Be2	3.21 ± 0.25^{AeB}	-3.14 ± 0.3^{Ade}	-2.85 ± 0.25^{Acd}	-2.62 ± 0.2^{Ac}	-2.23 ± 0.17^{Aab}	-2.21 ± 0.5^{Aa}
In2Be1.5	-3.2 ± 0.2^{AeB}	-3.17 ± 0.34^{Ae}	-2.99 ± 0.19^{Ad}	-2.75 ± 75.36^{Acd}	-2.3 ± 0.24^{ABab}	-2.32 ± 0.24^{Aa}
In2Be0.75	-3.2 ± 0.4^{AaBe}	-3.15 ± 0.7^{Aade}	-3.06 ± 0.3^{AaBd}	-2.83 ± 0.16^{ABc}	-2.45 ± 0.2^{ABab}	-2.39 ± 0.47^{ABa}
In1Be2	-3.2 ± 0.29^{AcB}	-3.18 ± 0.5^{Ac}	-3.01 ± 0.28^{AaBc}	-2.7 ± 0.35^{ABb}	-2.37 ± 0.5^{Aba}	-2.33 ± 0.14^{ABa}
In1Be1.5	-3.22 ± 0.22^{AcB}	-3.18 ± 0.2^{Ac}	-2.95 ± 0.4^{ABbc}	-2.7 ± 0.67^{ABb}	-2.45 ± 0.8^{Aba}	-2.37 ± 0.27^{ABa}
In1Be0.75	-3.2 ± 0.14^{AdB}	-3.18 ± 0.2^{Ad}	-3.01 ± 0.35^{AcB}	-2.78 ± 0.15^{ABb}	-2.39 ± 0.3^{Aba}	-2.34 ± 0.38^{ABa}
Control	-3.2 ± 0.1^{Aae}	-3.18 ± 0.25^{Ae}	-3.2 ± 0.1^{AdB}	-2.81 ± 0.4^{ABc}	-2.45 ± 0.7^{ABab}	-2.41 ± 0.2^{ABa}

* The difference between uppercase letters shows a significant difference (P<0.05) in the column and the difference of lowercase letters a significant difference (P<0.05) in the row (Mean ± standard deviation, three repetitions).

The highest transparency was in the control treatment and treatments containing 2 and 3% inulin and 0.75% of sugar beet fiber, respectively. The transparency increased in fiber value equal to inulin. The results of the effect of variables like storage time, percentage of inulin and sugar beet on b* index in Table (4), can be seen that with the passage of storage time, the value of b* index decreased in all treatments and the highest value of b* index was in the control treatment. There was a significant difference with other treatments during the maintenance period. As the sugar beet fiber increased, b* index increased. Moreover, in treatments containing equal amount of beet fiber, those with less inulin had a higher b* index. Table (5) shows the results of the effect of storage time, inulin and sugar beet values in synbiotic pineapple juice on index (a*). Over time, a* index increased. The lowest value of a* was in the control sample. Furthermore, the increase in the value of inulin and sugar beet fiber led to a decrease in a* index, where the difference between the samples was not significant at the beginning of the storage period (P>0.05), which became significant (P<0.05) over time. The results of L*, b* and a* colorimetric test in this study are in line with the results obtained from the evaluation of the color characteristics of pineapple juice containing *L. casei*, with the retention time of L* and b* indices decreased and a* index increased * (Costa *et al.*, 2013), one of the reasons for which was the growth of probiotic bacteria during the storage period.

Sensory evaluation

Of the most significant characteristics of food products are its sensory properties, including taste, flavor, color, texture and smell. The first condition for acceptance of the product by the consumer is its sensory properties. Factors like taste, color, texture and overall acceptance were evaluated by a 5-point hedonic method by 20 untrained evaluators, and treatments with very good, good, average, bad and very bad quality, scores of 5, 4, 3, 2, and 1, respectively (Krasaekoopt & Kitsawad, 2010; Nualkaekul & Charalampopoulos, 2011). Sensory evaluation of taste factor, shown in Table 6, has decreased with increase in storage time, popularity and taste acceptance in all treatments except treatments containing 3% inulin. The treatments of 2% inulin, 0.75% sugar beet fiber and 1.5% sugar beet fiber obtained the highest taste points, respectively. The difference between treatments was significant (P<0.05). Tables (6) and (7) show the results of color and texture evaluation of synabiotic pineapple juice in, where with increase in storage time, the desirability and acceptance of color and texture for evaluators decreased and the amount of reduction in inulin-containing treatments was 3% less that is insignificant in the treatments (P>0.05).

The results of evaluation of pineapple juice of synbiotic show that during the storage period, the color factor decreased in various treatments and the highest color factor score in the treatment of 2% inulin and 0.75% sugar beet fiber and the lowest score in the inulin treatment of 1% and sugar beet fiber was observed to be 0.75%. The results of tissue evaluation in Table (7) indicate that the increase in the storage time has led to a decrease in tissue sensory score in the treatments and fruit juice samples have a significant difference (P<0.05) with each other in terms of tissue changes during storage. With increasing the shelf life of pineapple juice samples containing 3% inulin, which contained 2, 1.5 and 0.75% of sugar beet fiber, respectively, had the highest sensory score of the tissue, respectively. The results of general acceptance in Table (7) showed that in all samples, with increasing the shelf life, the amount of general acceptance decreased, among which the treatment of 3% inulin and 0.75% sugar beet fiber was more acceptable than the other treatments and had the highest score of general acceptance sensory evaluation. Moreover, the difference between treatments is significant (P<0.05). Such results were seen in the study of probiotic apple juice (Shaykhgasemi & Zomorodi, 2014), pineapple juice and probiotic mango juice (AdebayoTayo & Akpeji, 2016; Hossain, Hoque, Hossain, Kabir, Yasin, & Islam, 2020; Hossain, Hoque, Kabir, & Yasin, 2019).

Table 6. The effect of storage time, amount of inulin and sugar beet on the taste of pineapple synbiotic juice

Sensory factor	Day	Treatments									
		In3Be2	In3Be1.5	In3Be0.75	In2Be2	In2Be1.5	In2Be0.75	In1Be2	In1Be1.5	In1Be0.75	Control
Taste	0	3.4±0.11^{Eg}	3.6±0.4^{Ff}	3.8±0.2^{Ee}	4.4±9.31^{Ac}	4.7±0.2^{Aa}	4.7±0.1^{Aa}	4.4±0.3^{Ac}	4.3±0.2^{Ad}	4±0.7^{Aa}	4.5±0.5^{Ab}
	5	3.3±0.3 ^{Fe}	3.7±0.2^{Ed}	4±0.2^{Dc}	4.0±0.1^{Cc}	4.5±0.4^{Ba}	4.5±0.3^{Ba}	4.2±0.2^{Bb}	4.2±0.1^{Bb}	3.7±0.1^{Bd}	4.5±0.3^{Aa}
	10	3.6±0.2 ^{Cf}	3.9±0.1^{De}	4.2±0.3^{Cc}	3.9±02Da	4.2±0.3^{Dc}	4.3±0.2^{Cb}	4.0±0.7^{Cd}	4.2±0.5^{Bc}	3.6±0.3^{Cf}	4.4±0.6^{Ba}
	15	3.8± 0.5^{Be}	3.4 ±0.5^{Bb}	4.4±0.7^{Ba}	4.2±0.14^{Bc}	4.3±0.5^{Cb}	4.3±0.7^{Cb}	3.8±0.4^{De}	4±0.4^{Cd}	3.2±0.7^{Df}	4.3±0.1^{Cb}
	20	3.9± 06^{Ae}	4.5±0.7^{Ab}	4.7±0.3^{Aa}	3.5±0.25^{Ea}	3.8±0.9^{Ef}	4.2±0.4^{Dc}	3.8±0.8^{Df}	4±0.7^{Cd}	3.1±0.4^{Eg}	4.2±0.2^{Dc}
	25	3.5±0.2 ^{Dc}	4.0±0.3^{Ca}	4.0±0.4^{Da}	3.4± 0.4^{Fd}	3.7±0.1^{Fb}	4±0.6^{Ea}	3.5±0.6^{Ec}	3.5±0.6^{Dc}	2.8±0.2^{Fe}	4.0±0.1^{Ea}
Color	0	3.9±0.5 ^{Ad}	2.3±0.9^{Ad}	4.7±0.2^{Aa}	4.4±0.5^{Ac}	4.7±0.5^{Aa}	4.7±0.4^{Aa}	4.7±0.2^{Aa}	4.5±0.1^{Ab}	3.7±0.3^{Ae}	4.5±0.5^{Ab}
	5	3.8± 0.2^{Be}	3.8±0.6^{Be}	4.4±0.2^{Bb}	4.2±0.17^{Bd}	4.3±0.6^{Bc}	4.5±0.3^{Ba}	4.4±0.7^{Bb}	4.3±0.2^{Bc}	3.6±0.5^{Bf}	4.5±0.6^{Aa}
	10	3.9±0.6 ^{Ae}	3.6±0.6^{Cf}	4.2±0.5^{Cb}	3.9±0.12^{Ca}	4.2±0.2^{Cb}	4.1±±0.7^{Dc}	4.2±0.3^{Cb}	4.0±0.6^{Cd}	3.3±0.1^{Cg}	4.4±0.7^{Ba}
	15	3.5±0.8 ^{Cf}	3.3±0.5^{Eg}	4.0±0.4^{Dc}	3.5±0.4^{Df}	3.8±0.3^{De}	4.3±0.6^{Ca}	4.0±0.8^{Dc}	3.9±0.9^{Dd}	3.1±0.3^{Dh}	4.2±0.2^{Cb}
	20	3.4±0.6^{De}	3.4±0.9^{De}	3.8±0.6^{Ec}	3.4±0.9^{Ee}	3.7±0.4^{Ed}	4.1±0.9^{Da}	4.0±0.9^{Db}	3.8±0.4^{Ed}	3.0±0.4^{Ef}	4.0±0.1^{Db}
	25	3.2±0.3^{Ee}	3.2±0.7^{Fe}	3.03±0.5^{Fd}	3.5±0.15^{Dd}	3.5±0.4^{Fd}	4.0±0.5 ^{Ea}	3.8±0.1^{Eb}	3.6±0.5^{Fc}	3.0±0.2^{Ef}	3.8±0.8^{Eb}

* The difference between uppercase letters shows a significant difference (P<0.05) in the column and the difference of lowercase letters a significant difference (P<0.05) in the row (Mean ± standard deviation, three repetitions).

Table 7. The effect of storage time, amount of inulin and sugar beet on texture and general acceptance of synbiotic pineapple juice

Sensory factor	Day	Treatments									
		In3Be2	In3Be1.5	In3Be0.75	In2Be2	In2Be1.5	In2Be0.75	In1Be2	In1Be1.5	In1Be0.75	Control
Texture	0	4.3±0.2^{Ac}	4.5±0.5^{Ab}	5.7±0.3^{Aa}	4.2±0.3^{Ad}	4.5±0.42^{Ab}	3.6±0.7^{Af}	4.5±0.3^{Ab}	3.9±0.2^{Ae}	3.4±0.4^{Ag}	4.5±0.6^{Ab}
	5	4.0±0.1^{Be}	4.3 ±0.3^{Bc}	4.6±0.5^{Ba}	4.2±0.1^{Ad}	4.3±0.6^{Bc}	3.5±0.2^{Bg}	4.3±0.21^{Bc}	3.8±0.15^{Bf}	3.3±0.1^{Ba}	4.4±0.1^{Bb}
	10	3.9±0.5^{Ce}	4.3±0.2^{Bb}	4.4±0.2^{Ca}	4.0±0.8^{Cd}	4.2±0.2^{Cc}	3.2±0.4^{Cg}	4.2±0.3^{Cc}	3.6±0.3^{Cf}	3.1±0.3^{Ch}	4.2±0.3^{Cc}
	15	3.8±0.2^{Da}	4.3±0.4^{Ba}	4.3±0.7^{Da}	4.1±0.1^{Bb}	3.8±0.3^{Da}	3.0±0.1^{Df}	3.8±0.14^{Da}	3.5±0.7^{De}	2.8±0.4^{Dg}	4.0±0.5^{Dc}
	20	3.4±0.6^{Ee}	4.2±0.6^{Cb}	4.3±0.2^{Da}	3.6±0.5^{Da}	3.7±0.1^{Ed}	3.0±0.6^{Df}	3.7±0.2^{Ed}	3.4±0.4^{Ee}	2.6±0.6^{Eg}	3.8±0.2^{Ec}
	25	3.2±0.4^{Fd}	4.0±0.2^{Db}	4.1±0.11^{Ea}	3.5±0.7^{Ec}	3.5±0.9^{Fc}	3.0±0.2^{De}	3.5±0.1^{Fc}	3.2±0.12^{Fd}	2.4±0.9^{Ff}	3.5±0.4^{Fc}
general acceptance	0	3.8±0.3^{Ag}	4.2±0.3^{Ae}	4.6±0.15^{Aa}	4.3±0.41^{Ad}	4.3±0.2^{Ad}	4.4±0.1^{Ac}	3.5±0.15^{Ch}	4.0±0.4^{Cf}	3.3±0.5^{Bi}	4.5±0.3^{Ab}
	5	3.7±0.5^{Bf}	4.1±0.2^{Bd}	4.4±0.42^{Bb}	3.8±0.17^{Ca}	4.2±0.5^{Bc}	4.2±0.7^{Bc}	3.8±0.19^{Be}	4.5±0.16^{Aa}	3.5±0.1^{Ag}	4.4±0.9^{Bb}
	10	3.7±0.2^{Bf}	4.0±0.8^{Cd}	4.4±0.13^{Ba}	3.9±0.1^{Be}	4.2±0.4^{Bc}	4.0±0.1^{Cd}	4.0±0.2^{Ad}	4.2±0.3^{Bc}	3.3±0.1^{Bg}	4.3±0.4^{Cb}
	15	3.6±0.7^{Ce}	4.1±0.21^{Bb}	4.3±0.2^{Ca}	3.8±0.2^{Ca}	4.0±0.3^{Cc}	3.8±0.3^{Dd}	3.5±0.3^{Cf}	3.5±0.2^{Df}	3.3±0.3^{Bg}	4.0±0.2^{Dc}
	20	3.5±0.3^{De}	4.1±0.3^{Bb}	4.3±0.32^{Ca}	3.7±0.6^{Dd}	4.0±0.25^{Ca}	3.8±0.5^{Dc}	3.2±0.24^{Dg}	3.3±0.11^{Ef}	3.2±0.6^{Cg}	3.8±0.5^{Ec}
	25	3.3±0.1^{Ed}	4.0±0.42^{Cb}	4.1±0.25^{Da}	3.2±0.12^{Ee}	3.5±0.21^{Dc}	3.5±0.4^{Ec}	3.0±0.4^{Ef}	3.0±0.1^{Ff}	3.0±0.2^{Df}	3.5±0.7^{Fc}

* T The difference between uppercase letters shows a significant difference (P<0.05) in the column and the difference of lowercase letters a significant difference (P<0.05) in the row (Mean ± standard deviation, three repetitions).

Conclusions

According to the results of sensory evaluation, Brix, pH and colorimetry (a*, b* and L*), samples containing 3% inulin and 0.75% sugar beet fiber were selected as the best treatment in this study. As with increase in the storage time of synbiotic pineapple juice at refrigerator temperature, the viability of the probiotic bacterium *L. acidophilus* has increased. Hence, the probiotic bacterium *L. acidophilus* can be used with 3% inulin prebiotics and 0.75% sugar beet one can conclude that to produce a useful pineapple drink with appropriate nutritional and therapeutic properties.

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اثر فیبر چغندرقند و اینولین بر بقاء و فعالیت *لاکتوباسیلوس اسیدوفیلوس*، خواص شیمیایی و حسی نوشیدنی سین‌بیوتیک آناناس

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چکیده

پروبیوتیک‌ها به‌ویژه باکتری‌های اسیدلاکتیک در تولید نوشیدنی‌های فراسودمند بسیار مفید بوده و باعث افزایش ارزش غذایی نوشیدنی‌ها می‌شوند. برای تولید نوشیدنی سین‌بیوتیک از باکتری‌های پروبیوتیک همراه با پری‌بیوتیک‌ها استفاده می‌شود. در این مطالعه، به بررسی تأثیر اینولین در سه سطح (1، 2 و 3 درصد)، فیبر چغندرقند (0/75، 1/5 و 2 درصد) و باکتری (*Lactobacillus acidophilus*) در تولید آب آناناس سین‌بیوتیک پرداخته شد. تیمارها به مدت 25 روز در دمای 4 درجهٔ سانتی‌گراد نگهداری و آزمون‌های تعیین pH، قابلیت زنده‌مانی باکتری‌ها، بریکس، رنگ‌سنجی و ارزیابی حسی در فواصل زمانی 5 روزه انجام شد. با افزایش مقدار اینولین، فیبر چغندرقند و همچنین زمان نگهداری، قابلیت زنده‌مانی باکتری‌ها و رنگ قرمزی-سبزی (a*) افزایش، و مقادیر بریکس، pH، شفافیت (L*) و رنگ زرد-آبی (b*) کاهش یافت. نتایج نشان داد که زمان نگهداری یکی از مهم‌ترین عوامل تأثیرگذار بر پارامترهای موردبررسی است. براساس نتایج مقادیر Brix، pH، رنگ‌سنجی (L*، b* و a*) و قابلیت زنده‌مانی باکتری‌ها در روز 25 تیمار حاوی 3 درصد اینولین و 0/75 درصد فیبر چغندرقند به‌ترتیب 19/17 درصد، 3/6، 68/91، 14/3، 2/19-، 12/28 واحد تشکیل کلنی/گرم بوده است و به‌عنوان تیمار ایده‌آل انتخاب شد. بنابراین می‌توان نتیجه گرفت که به‌منظور تولید نوشیدنی فراسودمند آناناس می‌توان از باکتری *لاکتوباسیلوس اسیدوفیلوس* با پری‌بیوتیک‌های اینولین و چغندرقند استفاده نمود.

واژه‌های کلیدی: آناناس، اینولین، فیبر چغندرقند، نوشیدنی سین‌بیوتیک