Production of Fermented Red Beet Juice using Probiotic Lactobacilli Bacteria

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Abstract

During this experiment, the juice of red beets were extracted and fermented by two species of probiotic bacteria :(*Lactobacillus plantarum* and *Lactobacillus paracasei*). Both species of lactic cultures can use beet juice for the producing of lactic acid and the synthesis of cells.Theprocess of fermentation was carried at 30°C for 24 hr. by inoculated of beet juice using lactic cultures. *Lactobacillus plantarum* and *Lactobacillus paracasei* grew well on the vegetable juice of ((Beet plant)) and reached nearly (12.5± 4.12×10) and (8.6± 5.20×10) CFU/ml respectively followingfermentation for 24 h at 30°C. After cold storage for 6 weeks at 4°C, PH, Acidity as (Lactic acid), viable cell counts were detected weekly.As a result the viability of *L. plantarum* and *L. Paracasei* were increased (9.03-9.69 log CFU / ml) respectively on the 42 day of cold storage while, PH of these bacteria lowered to (2.1± 0.020) respectively therefore, increased acidity of fermented beet juice whilst the viability of cell counts of *L. plantarum* and *L. paracasei* were not lost during cold storage at 4°Cconditions.

Keywords: Red Beet Juice; Probiotic; Lactobacillus sp.; Fermentation

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1. Introduction

Probiotics are described as selected, viable microbial nutritional supplements, which are beneficial by their impacts on the intestinal tract when introduced in sufficient amounts. *Lactobacillus* species could eliminate high levels of cholesterol; the probiotics market continues to develop as awareness of their health benefits rises, along with their science support (Manju and Garima, 2013). In food products, some strains of (LAB) such as: *Lactobacillus, Bifidobacterium, Streptococcus,* and *Lactococcus* were promoted due to their renowned health advantages (Gamage*et al*., 2016).Several researchers have stated that the use of multiple vegetables, fruits and cereals in numerous countries around the globe to produce synbiotic drinks or beverages (Marla*et al.*, 2012).

Healthy foods these days imply "functional products," and in general we label a food as functional. It has beneficial impacts or more particular benefits for the body in comparison to traditional impacts.Common instances of functional food are those containing or that are produced with bioactive compounds like nutritional fibers, oligosaccharides and active "friendly" bacteria that enhance the balance of bacterial intestinal strain. Besides well-established functional components like micronutrients, vitamins and minerals, probiotics are component of active ingredients' developing generation involving lipids, prebiotics and phytonutrients (Marianne et al., 2015).

Probiotic products have been divided into (2) kinds: Dairy probiotic products such as cheese, yogurt, cream, ice cream, butter, whey drinks, milky desserts and baby foods (Mohammadi et al, 2012). While others, non-milk products containing probiotics such as grain, sweets, multiple drinks such as juices and non-alcoholic beer, foods for children and meat products (Sohrabvandi et al, 2010).

Probiotics Viability and stability for industrial producers have both been a technological and marketing challenge in order to be functional, probiotics must be viable and at sufficient dosage levels.Probiotic food / feed supplements are produced for needs that the strains retain an adequate amount of viable cell count during processing, which is the shelf-life of the product, should sustain stress conditions of gastrointestinal and

processing and they should preserve its biological function inside the host prior to the delivery of probiotic strains to food / feed. All these criteria must be taken into consideration when selecting a probiotic strain(Kosin and Rakshit, 2006)

Food today plays a major role in health and human nutrition in most societies; probiotic pharmacological products are applied as tablets or capsulesto treatvariousillnessessuch as intestinal andvaginal infection, diarrhea, constipation and lactose intoleranceimproved immune systems, and reducedcolon cancer risk (Mohammad et al, 2016).

Probiotics traditionally have been added to yogurt and to other dairy products that are fermented as well, but there are two drags associated with their consumption which are lactose intolerance and cholesterol content. Lately, Consumers ' attraction for non-milk products with probiotics has risen, and probiotics have been combined into drinks (Shah, 2001). Vegetables and Fruit contain high amount of functional components of food like dietary fibers, antioxidants (phytochemicals), minerals and vitamins. Moreover, vegetables and fruit do not have dairy allergens that mightavoid the use of specific groups of the population (Luckow and Delahunty, 2004).A part from consumption in its fresh form of red beet plant (Beta vulgaris) is also an invaluable crop used in the food industry for the manufacturing of dried and frozen food, non-concentrated and concentrated juices and natural coloring agents (betalains) used as food additives. (Gamage *et al*., 2016). According to (Altman & Dittmer, 1968), beets contain around 87.3% humidity, 9.1% carbohydrate, 1.6% protein0.1% fat, 0.8% fiber, and 1.1% ash. Beet small plants are are tasty and tender and could be processed to pickled or canned products or fresh for human consumption (Pederson, 1979). However, owing to their fibrous or difficult texture, larger/oversized red beets are underused at present. This research aims to determine the bigger beets' suitability as a raw material for the production of probiotic beet juice by *L. paracasei* and *L. plantarum* as valuable lactic acid bacteria.

2. Materials &Methods

2.1. Preparation of red beet juice

Red beets were bought from a market and prepared by hacking the vegetables, after which the juice was separated in a juice manufacturer, the juices were heated at 70 ^{o}C for 20 minutes and kept at 4 ° C until they were used. (Marica et al,2004).

2.2. Fermentation of probiotic red beet juice

Two types of lactic acid bacteria, *L. plantarum and L.paracasei* (SelçukÜniversitesi, Fen fakultesi, Biyoloji culture collection. Konya-Turkiye) were utilized in this experiment. Inoculum has been prepared for 24 h by growing the culture at 30°C in MRS broth mediumcontaining gram per liter: (dextrose 20.0, meat peptone 10.0, beef extract 10.0, yeast extract 5.0, C2H3NaO2 5.0, Na2HPO4.2H2O 2.0, C6H8NH4 2.0, Tween 80 1.0, MgSO4 0.1, MnSO4 0.05) and final PH (at 25°C) is 6.5±0.2 (deMan and Rogosa, 1960).Following incubation for 24 hoursat 30°C, with the purpose of counting a viable cell (Log cfu / mL) of *L. paracasei* and *L. plantarum* as an inoculum, the strategy of standard plate method with MRS agar media was chosen. Fermentation was conducted with 100 ml of beet juice in Erlenmeyer flask (6×250 mm). The inoculation of the whole samplewasconducted with a 24-hour (LAB) (10^6 CFU / ml) culture and incubated then for 24 hours at 30 ° C. The fermented samplewas subsequently stored at 4 ° C for 42 days, samples were drawn for chemical and microbiological analysis every week (1, 7, 14, 21, 28, 35, 42 days) (Mousavi et al, 2011).). PH measurement was performed by using a PH meter, Viable cell count (Log cfu / mL) was determined after 24 hours of incubation at 30 ° C using the Standard Plate Methods (S.P.M) with the Lactobacilli MRS medium to examine the impact of cold storage on cell viability in fermented beet juice as logarithmic colony creating units (Log cfu / mL).

2.3. Statistical analysis

The experiments of fermentation have been performed in triplicate; also the findings are shown as mean± standard deviation (S.D). The experimental information was analyzed using the SAS statistical machine. The values in rows which do not possess a prevalent superscript are considerably distinct ($p < 0.05$) based on the various range test of Duncan.

3. Results and discussion

Table 1 of data shows time course (h) of beet juice fermentation by *L. plantarum* and *L. paracasei* , the number of cell counts increased significantly by 12,5×10 CFU / mL and 8,6 ×10 CFU / mL after 24 h of incubation at 30 ° C and was discovered to be able to grow well on beet juices without addition of nutrients.

The time course (weekly) beet juice fermentation by *L plantarum* was explored in Table 2 result and its impact on PH and acidity values during cold storage, viability of *L. Plantarum* increased from 7.80 during the first week to 9.03 Log cfu / mL at the end of storage*L. plantarum* reduced the PH of fermented red beet juice from 4.6 to 2.1 after 42 days of cold storage at 4 ° C owing to its capacity to generate a big quantity of lactic acid resulting in acidity of 17.10 g/100ml after 6 weeks of cold storage.(kyuong et al., 2005; Fadhil, 2015)Proposed that juices of vegetable and fruit could be used as a medium for probiotics growing and stated that probiotic cultures *Bifidobacterium bifidum L. Reuteri* and *L. acidophilus* have grown properly in products based on non-dairy oats.

The impact of cold storage on feasible cell counts, PH and acidity of L. paracasei is demonstrated in Table 3. The number of bacterial cell counts elevated from 7,97 to 9,69 Log cfu / ml during the 1st to the 6th weeks of cold storage at 4 ° C fermented beet juice. Nevertheless, *L. paracasei* lactic cultures in the fermented juice rapidly decreased the PH value from 4.8 to 2.1, resulting in rising of fermented juice acidity from 9.06 to 16.20 g/100 ml following 42 days of 4 ° C cold storage. The minimum amount of probiotic bacteria needs to be 106 CFU / ml in a food item. Consequently, the lactic acid bacteria viability during refrigerated or frozen condition was the most significant agent.

Table 1:Time course (h) of fermentation of beet juice by *L. plantarum* and *L. paracasei*

	Viability (CFU/mL)	
Time (h)	*L. plantarum*	*L.paracasei*
0	7.2± 2.34 × 10	3.2± 1.00 × 10
12	9.3± 3.11 × 10	5.1± 2.30 × 10
24	12.5± 4.12× 10	8.6± 5.20 × 10

Table 2: Time course (week) of fermentation of beet juice by *L. plantarum* during cold storage

Time (week)	PH value	Acidity (g/100ml)	(Log cfu/mL)
1	4.6± 0.020	8.24± 0.200	7.80± 0.080
7	3.6± 0.020	10.73± 0.200	8.98± 0.010
14	3.5± 0.020	12.83± 0.200	8.70± 0.055
21	3.5± 0.020	13.70± 0.200	8.57± 0.025
28	3.4± 0.020	16.80± 0.200	8.46± 0.010
35	2.2± 0.020	15.72± 0.200	8.41± 0.020
42	2.1± 0.020	17.10± 0.200	9.03± 0.055

Table 3: Time course (week) of fermentation of beet juice by *L. paracaesi* during cold storage

Time (week)	PH value	Acidity (g/100ml)	(Log cfu/mL)
1	4.8± 0.000	9.06± 0.2000	7.97± 0.035
7	4.7± 0.000	10.12± 0.2000	8.87± 0.105
14	3.6± 0.010	10.35± 0.2000	8.88± 0.035
21	3.5± 0.020	11.75± 0.2000	8.90± 0.040
28	3.3± 0.000	12.90± 0.2000	8.11± 0.000
35	2.3± 0.010	14.00± 0.2000	9.24± 0.020
42	2.1± 0.020	16.20± 0.2000	9.69± 0.020

Probiotic organismsviability depends on product oxygen levels, package oxygen permeation, time of fermentation, and the storage temperature (Shah, 2000). Other factors for the loss of probiotic bacteria viability were linked to decrease medium PH level and the organic acid accumulation as a development and fermentation consequence (Kyung et al. 2006; Kyung et al, 2005). We discovered in this research that *L. plantarum* and *L. paracasei* both can live the Low PH and elevated acidity in fermented red beet juice.

4. Conclusion

According to the findings of this experiment, it is possible to conclude that red beet plants can be considered for fermentation with *L. plantarum* and *L. paracasei* as the finest raw materials for producing probiotic beet juices. These lactic cultures grew well in this fermented juice. the PH value of the fermented red beet juice is 2.1 (high acidity), also it contains a great amount of beneficial lactic acid (9.03, 9.69 Log CFU/ml) on the 42 day during cold storage at 4°C without loss viability, we can therefore use these crops to create healthy beet drinks for customers who have allergy to lactose that is found in dairy products.

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