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Short communication

Development of probiotic drink from cucumber using *Lactobacillus* sp.

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ABSTRACT

Probiotics are health-friendly microflora, which exert beneficial effect on our body when taken in appropriate doses. Cucumber (*Cucumis sativus*) is a good source of minerals and is generally used as salad. Cucumbers were gently washed and immersed in 2 per cent brine solution containing 0.1 per cent coarsely ground mustard seeds. It was fermented at 37 ± 2°C in air tight container for 7 days. The filtered liquid was stored at low temperature (4°C) for 28 days. The product had *Lactobacillus* count of >10^6 CFU per ml. The drink was supplemented separately with capsicum, ginger, sugarbeet and mint. Drink supplemented with capsicum obtained maximum sensory scores with maximum ascorbic acid content (1.89 mg/ 100 ml) and lowest pH (3.76). The product could be stored safely for 28 days at low temperature (4°C).

Key words: Cucumber, drink, fermentation, *Lactobacillus*, probiotic.

A wide number of products like curd, cheese, pickle, vinegar, wine, cider, yoghurt, etc. are produced through microbial fermentation. Fermented food products, besides having good nutritional profile, provide good resistance to human body against certain hardcore diseases. The probiotics come under the category of functional food, which benefit the human health beyond the level of basic nutrition (Suvarna and Boby, 9). Rivera-Espinoza and Gallardo-Navarro (6) described health benefits of probiotics on gastrointestinal infections, antimicrobial activities *etc*. Lactic acid bacteria are among the most common types of microbes used as probiotics. These help in maintaining health-promoting microbial population in the digestive tract and protect the body from various forms of pathogenic infections and intestinal diseases.

In recent years, research outcomes have revealed that fermented fruit and vegetable products are equally good as probiotic materials. Dairy products are generally associated with problems of cholesterol and lactose intolerance, while fruit and vegetable products are rich in vitamins and minerals. Fruits and vegetables like carrot, sugarbeet, cabbage, tomato, capsicum and cucumber can be made probiotic using fermentation technology. Yoon *et al*. (11) prepared probiotic tomato juice using different strains of *Lactobacillus*. Similarly, Kyung *et al*. (3) found *L. acidophilus* and *L. plantarum* suitable for preparation of probiotic beet juice.

Cucumber is known for it's highly refreshing and health promoting qualities. It is a very low calorie vegetable that provides just 15 calories per 100 g fruit. Cucumber contains good amount of vitamin K (17 µg/100 g) and potassium (147 mg/100 g), very low sodium (2 mg/ 100 g) and no saturated fat or cholesterol. Lactic acid fermentation of cucumber for pickle preparation is a common practice in western countries (DI Cagno *et al*., 2). In the present investigation, protocol was standardized for obtaining an organoleptically acceptable probiotic drink from brine of pickled cucumber by utilizing *Lactobacillus* sp. bacteria, present on the surface of the fruit and improving its sensory qualities through addition of sugarbeet, mint, capsicum and ginger.

The present study was carried at the Division of Post Harvest Management, Central Institute for Subtropical Horticulture, Lucknow. The fresh harvested cucumber fruits were procured from wholesale market and processed for making juice as presented in flow sheet.

1 kg fresh, medium size, firm, green cucumbers
↓
Gentle washing with soft hands
↓
Dipping in 5 l cap. glass jar containing 4 l of 2% brine + 0.1% coarsely ground mustard seeds
↓
Fermentation at 37 ± 2°C for 7 days under anaerobic condition
↓
Separation of cucumbers & filtration of liquid
↓
Addition of sugarbeet slices (5%)/ capsicum shreds (5%)/ mint leaves (2%)/ ginger shreds (2%)
↓
Incubation at low temperature (4°C) for 3 days
↓
Separation of added plant materials
↓
Filling of drink in glass bottles and sealing
↓
Storage at low temperature (4°C) for 28 days

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The total soluble solids of beverages were recorded by using hand refractometer (Erma, Japan). Titratable acidity (as lactic acid), ascorbic acid and total phenolics were determined as per the methods described by Ranganna (5). Ascorbic acid content of beverage was measured using Folin-Ciocalteau's reagent. The pH value of the drink was taken using pH meter. Quantification of nisin was done as per the method described by Pongtharangkul and Demirci (4). The microbial examination of fermented beverage was carried out as per method detailed by Speck (8). The organoleptic evaluation of beverage was carried out on the basis of appearance, colour, acidity, aroma and taste by a panel of seven semi-skilled judges, using a 9-point Hedonic scale as prescribed by Shukla *et al*. (7). Microbial isolate was identified as per method of Bergey *et al*. (1). The experiment was laid out in a Completely Randomized Design (CRD) along with three replicates for each treatment. The data was analyzed statistically using software OPSTAT developed by OP Sheoran of CCSHAU, Hisar, India.

The *Lactobacillus* sp. count (identified up to genus level) of the prepared beverage was worked out to be more than 10^6 as also reported by Yoon *et al*. (11, 12) in probiotic tomato and beet juices. There were non-significant changes in the microbial counts of pobiotic drinks during storage. It was found to be more than 10^6 in all the treatments after 28 days of storage. The beverage possessed anti-microbial properties owing to presence of nisin produced by *Lactobacillus*. It restricted growth of *Aspergillus niger* colony in culture. The nisin content of the drink, calculated on the basis of colony size, was found to be 250 µg per ml.

The total soluble solids (TSS) of the product were found to be between 2.4 to 2.6°Brix at zero day. Negligible changes were observed in TSS values during storage up to 28 days. The initial titratable acidity of the drink (0.16%) increased to 0.2 per cent in case of capsicum fortified samples, while, remained almost unchanged in other treatments at the end of storage period (Table 1). Yoon *et al*. (12) reported increase in acidity of lactic acid fermented beet juice during storage. Decrease in pH of the drink from 3.76 to 3.60 was observed in capsicum added samples, while it decreased in case of control and sugarbeet fortified samples and increased in mint and ginger added samples (Table 1). Tantipaibulvut *et al*. (10) also reported decrease in pH value of probiotic roselle juice during storage. The acid and pH data clearly indicated that addition of capsicum accelerated development of acidity in the beverage. Addition of capsicum also had enhancing effect on the level of ascorbic acid. The initial value of ascorbic acid in the drink rose from 0.32 mg/100 ml to 1.89 mg/100 ml on addition of capsicum (Table 2). Sugarbeet, mint or ginger, on the other hand, did not cause any change in ascorbic acid level. A decrease in ascorbic acid content was observed in all the samples during storage. Loss in ascorbic acid content might be attributed to gradual oxidation of ascorbic acid molecules during storage. At zero day, control sample possessed 3.04 mg/100 ml of total phenolics (Table 2). All the additives contributed to increase in the phenolic content of beverage i.e. mint contributed most (11.5 mg/100 ml), while sugarbeet least (4.71 mg/100 ml). A gradual decrease in the total phenolics content was noticed in all the samples till the end of storage period, mint still had highest phenolics (10.31 mg/100 ml, while control lowest (2.05 mg/100 ml). The decrease in phenolic might be due to break down of these compounds with the storage period.

The sensory evaluation of the beverage on the basis of appearance, colour, acidity, aroma and taste revealed that addition of plant materials enhanced the sensory acceptability of the drink, ranging from 7.3 to 8.6 compared to control with 6.9 (Fig. 1). The highest score of 8.6 was obtained in capsicum added sample due to its pleasant and soothing flavour. The score increased with increase in the storage period in case of control, sugar beet and capsicum added beverages. Mint and ginger, however, exhibited continuous decrease in organoleptic scores up to 28 days of storage. This might be due to deterioration in the flavouring compounds in these treatments. Capsicum added beverage scored highest throughout

Table 1. Changes in titratable acidity and pH value of probiotic cucumber drink during storage.

Treatment	Titratable acidity (%)			pH value		
	0 day	14 days	28 days	0 day	14 days	28 days
Control	0.16	0.13	0.14	3.84	3.84	3.81
Sugarbeet 5%	0.16	0.14	0.16	3.79	3.73	3.73
Capsicum 5%	0.16	0.17	0.20	3.76	3.62	3.60
Mint 2%	0.16	0.14	0.14	3.89	3.98	3.97
Ginger 2%	0.16	0.14	0.14	3.81	3.90	3.86
CD at 5%	T = 0.0025, P = 0.0019, T × P = 0.004			T = 0.0114, P = 0.0088, T × P = 0.020		

Table 2. Changes in total phenolics and ascorbic acid contents of probiotic cucumber drink during storage

Treatment	Total phenolics (mg/100 ml)			Ascorbic acid (mg/100 ml)		
	0 day	14 days	28 days	0 day	14 days	28 days
Control	3.04	2.32	2.05	0.32	0.16	0.11
Sugarbeet 5%	4.71	3.93	3.77	0.32	0.16	0.11
Capsicum 5%	5.95	5.33	5.25	1.89	0.54	0.30
Mint 2%	11.50	10.57	10.31	0.32	0.16	0.11
Ginger 2%	5.35	4.57	4.49	0.32	0.16	0.11
CD at 5%	T = 0.1368, P = 0.1060, T × P = 0.237			T = 0.0144. P = 0.0111, T × P = 0.025		

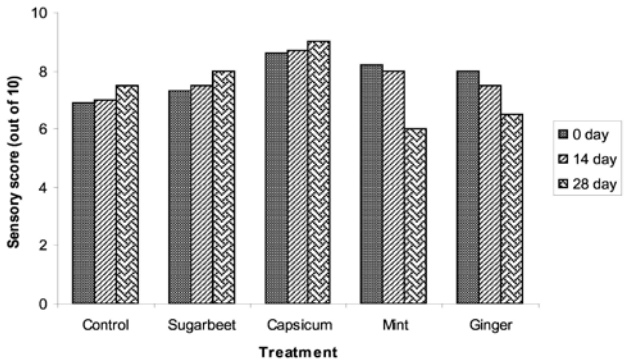

Fig. 1. Changes in sensory scores of probiotic beverage during storage

(Levels: Sugarbeet 5%, Capsicum 5%, Mint 2%, Ginger 2%)

the storage period and rose to as high as 9.0 after 28 days.

It may be inferred from the present study that acceptability of probiotic drink from cucumber can be enhanced by blending it with capsicum. The resultant beverage has pleasant flavour and taste besides containing good amounts of ascorbic acid and phenolics. It can be stored up to 28 days at low temperature (4°C) without any deterioration in its sensory qualities.

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