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Effect of tarragon on the growth of *Lactobacillus acidophilus* and *Bifidobacterium bifidum* in probiotic dairy products

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ABSTRACT

The objective of this study was to investigate the effects of different doses of tarragon on the growth of two probiotic bacteria, *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together) in milk and yoghurt produced in the first and second phases, respectively. The products were then examined in terms of PH, acidity and microbe counting during the incubator setting period and their respective permanence. In day 14, the production quantities of the products were evaluated by sensory method. The results of the questionnaires filled by 30 people in statistical-descriptive test were analyzed using SPSS software version 17 system. In the samples containing both bacteria, *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together), it was observed that the increased concentration of tarragon create a favorable taste of yoghurt and the yoghurt sample with %1 tarragon was with the best taste. The testifier and the yoghurt containing %1 tarragon were of greater permanence than the other samples. The bioability of probiotic bacteria was measured by direct counting method. Duration of the product permanence was determined within 21 days. Upon examination of the results, it was revealed that the increased concentration of tarragon had a positive effect on the growth of the probiotic bacteria, *Lactobacillus acidophilus* and *Bifidobacterium bifidum* in probiotic milk and yoghurt. To evaluate the effect of tarragon on the amount of the probiotic yoghurt protein, the control sample and the yoghurt sample containing %3 tarragon were tested, it was observed that the amount of protein in the sample with %3 tarragon was more than the control sample. © 2014 Trade Science Inc. - INDIA

KEYWORDS

Probiotic;
Lactobacillus aciophilus;
Bifidobacterium bifidum;
Tarragon;
Milk;
Yoghurt.

INTRUDUCTION

The growing health awareness in the consumption

of microorganisms as probiotic has encouraged consumers worldwide. To attain health benefits from health friendly organisms, certain instructions were on

FULL PAPER

demand hence a number of organizations to recommend guidelines for their use^[5,9]. Probiotics are dietary supplements and live microorganisms which when administered in adequate amounts confer a health benefit on the host^[4,10].

Strains of the genera *Lactobacillus* and *Bifidobacterium*, are the most extensively used probiotic bacteria. LAB has been used in the food industry for many years, because they are able to convert sugars (including lactose) and other carbohydrates into lactic acid^[7]. Recently, the design and production of plant based probiotic products have received much attention chiefly due to their natural health benefits (protein, fiber, vitamin and salts) and also because of the Variety in their production. Therefore, it seems that the issue of producing probiotic foods with appropriate qualities will be a major research topic for prospective researchers^[1].

Although, in the past decades, the synthetic chemical drugs that make use of separation mechanisms have been much in demand, their corresponding side effects are being gradually observed so much so that their irregular and improper consumption has turned out to be a critical issue.

On the contrary, the benefits of medicinal plants and their little or zero side effects have made them a proper substitute, highly appreciated by physicians and patients. Iran possesses a very rich source of such plants and herbs in the world in terms of variety and amount. The essence plants play a critical role in human life, and have been used for long by Iranians.

Tarragon is a perennial member of the Asteraceae family, related to the herb wormwood. The herb is widely used as food seasoning and as a primary flavor for some brands of carbonated beverages. The plant contains compounds that help alleviate pain associated with dental conditions, and that promote bile production and detoxification by the liver, thus aiding digestion, and they may act as a mild sedative^[6,8].

Tarragon may be useful in management of dysregulated glucose metabolism, including hyperglycemia, diabetes, and related metabolic syndromes^[3,8,14].

This aims of the present study was to evaluate the effect of tarragon on the growth of *Lactobacillus acidophilus* and *Bifidobacterium bifidum* in probiotic milk and yoghurt.

MATERIALS AND METHODS

Materials

Materials included tarragon plant extract, Low-fat milk and yoghurt from supermarket (% 1/5 fat), lyophilized *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together) (CHR Hansen Company, Denmark) and MRS Agar (Merk Company, Germany).

Effect of tarragon on the production of probiotic *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together) milk as first passage:

In order to produce the milk containing the probiotic bacterium *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together), four containers each containing 1 liter of low-fat sterilized milk (% 1/5 fat) were considered as our four groups. 0/33 gram starter (*Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together)) was added directly to all the containers, followed by adding tarragon powder 0 (the control sample), 1, 2 and %3 to all the containers, respectively and finally they were placed in an incubator at 38 °C. The acidity test was performed approximately every 2 hours until reaching 42 °Dornic. The samples were then taken out of the incubator and transferred to a refrigerator and stored at 2 °C. The produced probiotic milk was evaluated once every 7 days by counting the microbes using direct counting method.

Effect of tarragon on the production of probiotic *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together) yoghurt at the second passage:

To Produce *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together) yoghurt in this stage, after providing four containers, 1 liter of low-fat sterilized probiotic milk (% 1/5 fat) from the control group at first passage, 15 gram (% 1/5) starter of low-fat yoghurt and 0/33 gram (%33) starter (*Lactobacillus acidophilus* and *Bifidobacterium bifidum* (together)) was added to each container. Different concentrations of tarragon (0, 1, 2 and %3) were added respectively to the containers and mixed properly so that tarragon was uniformly dissolved. After wards, all the containers were placed in the incubator at 38 °C. Approximately

every 2 hours, the acidity and pH tests were done until acidity reached 70 °Dornic. Then the samples were taken out of the incubator and transferred to the refrigerator and stored at 2 °C. The produced probiotic tarragon yoghurt was evaluated every 7 days by counting the microbes using direct counting method and after 7 days the yoghurt was evaluated for sensory properties, using questionnaires filled by 30 people. The respondents were asked to rate the factors of scent, taste and thickness on a scale ranging from very good, good, medium, to weak. He results were analyzed in a statistical descriptive test by SPSS version 17 software. To evaluate the effectiveness of probiotic yoghurt,

kjeldahl method was used.

RESULTS

TABLE 1 shows the pH level in the tarragon *L. acidophilus* and *B. bifidum* (together) milk and yoghurt during incubation and TABLE 2 shows the acidity level in the tarragon *L. acidophilus* and *B. bifidum* (together) milk and yoghurt during the same time. TABLE 3 shows the PH degree in tarragon milk and yoghurt *L. acidophilus* and *B. bifidum* (together) during the same cooling period, TABLE 4 shows the acidity degrees of tarragon milk and yoghurt *L.*

TABLE 1 : The pH level in the tarragon *L. acidophilus* and *B. bifidum* (together) milk and yoghurt during incubation

		pH Level							
Tarragon Milk (%)	00:00(h)	02:00(h)	04:00(h)	Tarragon Yoghurt (%)	00:00(h)	02:00(h)	04:00(h)	04:20(h)	
0	5/49	5/26	4/22	0	5/49	5/38	4/33	3/43	
1	5/44	4/66	3/91	1	5/43	4/89	3/29	---	
2	5/31	4/51	---	2	5/34	4/86	3/23	---	
3	5/24	4/89	---	3	5/31	4/91	3/31	---	

TABLE 2 : The acidity level in the tarragon *L. acidophilus* and *B. bifidum* (together) milk and yoghurt during incubation

		Acidity Level in Dornic degree							
Tarragon Milk (%)	00:00(h)	02:00(h)	02:15(h)	Tarragon Yoghurt (%)	00:00(h)	02:00(h)	04:00(h)	04:20(h)	
0	20	22	44	0	20	31	45	70	
1	21	35	42	1	30	44	95	---	
2	25	42	---	2	34	47	105	---	
3	23	46	---	3	42	52	110	70	

TABLE 3 : The PH level in the tarragon *L. acidophilus* and *B. bifidum* (together) milk and yoghurt wit in 21-day storage in the refrigerator

		PH Level						
Tarragon Milk (%)	7 day	14 day	21 day	Tarragon Yoghurt (%)	7day	14day	21day	
0	5/42	5/51	5/69	0	5/22	4/87	4/84	
1	5/38	5/47	5/74	1	5/17	5/01	5/01	
2	5/86	5/97	6/34	2	5/12	5/04	5/03	
3	6/35	6/49	6/72	3	5/24	5/19	5/15	

TABLE 4 : The acidity level base on Dornic degree in the tarragon *L. acidophilus* and *B. bifidum* (together) milk and yoghurt within 21-day storage in the refrigerator

		Acidity Level in Dornic degree						
Tarragon Milk (%)	7 day	14 day	21 day	Tarragon Yoghurt (%)	7day	14day	21day	
0	63	74	82	0	107	110	100	
1	85	92	98	1	122	113	118	
2	81	89	94	2	114	130	143	
3	75	80	84	3	145	151	145	

FULL PAPER

acidophilus and *B. bifidum* (together), during storage time in the refrigerator, TABLE 5 shows the growth rate of microbes in the tarragon *L. acidophilus* and *B. bifidum* milk and yoghurt and TABLE 6 shows the sensory evaluation of tarragon *L. acidophilus* and *B. bifidum* (together) and yoghurt.

TABLE 5 : Growth of microbes in the tarragon *L. acidophilus* and *B. bifidum* (together) milk and yoghurt

Tarragon Milk (%)	10 ⁻⁵ cfu/gr	Tarragon Yoghurt (%)	10 ⁻⁵ cfu/gr
0	55 × 10 ¹¹	0	75 × 10 ¹¹
1	27/5 × 10 ¹¹	1	42/5 × 10 ¹¹
2	30 × 10 ¹¹	2	50 × 10 ¹¹
3	31/25 × 10 ¹¹	3	27/5 × 10 ¹¹

TABLE 6 : Sensory evaluation of tarragon *L. acidophilus* and *B. bifidum* (together) and yoghurt

Tarragon yoghurt (%)	taste	aroma	thickness	color
0	120	110	70	123
1	98	101	68	90
2	81	98	63	81
3	62	80	60	74

DISCUSSION

The probiotic concept is today widely spread in the scientific and industrial fields. However, further scientific input is required. Probiotic cultures are described as live microbial feed supplements that improve intestinal microbial balance and are intended for maintenance of health or prevention, rather than curing of disease. The demand for probiotic foods is increasing in Europe, Japan and the U.S. reflecting the heightened awareness among the public of the relationship between diet and health^[1,10].

In recent years, the probiotic bacteria, as the food additives, have been introduced into numerous foods, of which the dairy products especially yoghurt has played an important role in carrying these bacteria (such as *Lactobacillus acidophilus* and *Bifidobacterium bifidum*). Eating regularly the sufficient amounts of the living cells called "the minimum treatment" is required if the consumer is to benefit from the probiotic products. The daily recommended amount of the yoghurt containing 10⁶ CFU.ml⁻¹ probiotic bacteria is 100 gr. It is also very important to investigate the survival of these

microorganisms within the interval between storage in the refrigerator and consumption^[2].

Essence medicinal plants and herbs play a significant role in the human life and have been very popular for long among the Iranians. The tarragon as a medicinal plant, has extensive effects of which the most notable include appetite stimulant and treating gastrointestinal diseases^[8].

In the present study, the effects of tarragon on the growth of the bacteria *L. acidophilus* and *B. bifidum* in probiotic milk and yoghurt were investigated. The acidity, PH and survival of the bacteria in the tarragon probiotic milk and yoghurt were evaluated at 2 h interval till reaching 42 °Dornic acidity degrees for milk and 70 °Dornic for yoghurt in the incubator at 38 °C and also within 21-day period of storage in the refrigerator.

The probiotic milk containing %3 tarragon reached 42 °Dornic acidity much earlier than other samples, which was transferred to a refrigerator and stored at 2. So, this sample had the most effect on the growth of bacteria during incubation.

The sample with %2 tarragon and subsequently the control sample and finally the sample with 1% tarragon reached 42 °Dornic and transferred to the refrigerator. So, the sample with 1% tarragon had a minimal effect on the growth of bacteria during incubation. The acidity of the milk product with plant (no bacteria) did not change. This sample was transferred to the refrigerator, too.

During the 21 days storage of milk samples in the refrigerator, the acidity levels in the sample with %1 tarragon was higher than others, and subsequently the samples with %2, %3, %0 (the control sample) and finally the milk product with plant (no bacteria) were higher, respectively. So, during refrigeration milk product with plant (no bacteria) and the control sample had the most persistence. The milk sample containing %1 tarragon had a minimal persistence.

In the direct counting method of bacteria, maximum number of microbes were observed in the control sample and subsequently milk product with plant (no bacteria), the sample with %3, %2 and finally %1 tarragon, were more, respectively, in the first week. Maximum colony growth were observed in the sample with %3 tarragon and subsequently the sample with %1, %2 and %0 tarragon were more, respectively. The milk product with

plant (no bacteria) had no colony growth. The samples containing probiotic yoghurt were transferred to the incubator at 38 °C to let the acidity reach 70 °Dornic. The probiotic yoghurt containing %3 tarragon reached 70 °Dornic acidity earlier than others which was transferred to a refrigerator and stored at 2 °C. So, this sample had the most effect on the growth of bacteria during incubation. The sample with %2 tarragon and subsequently the sample with %1, yoghurt product with plant (no bacteria) and finally the control Sample reached 70 °Dornic and transferred to the refrigerator. So, the control Sample had a minimal effect on the growth of bacteria during incubation.

During the 21 days storage of yoghurt samples in the refrigerator, the acidity levels in the sample with %3 tarragon was higher than others, and subsequently the samples with %2, yoghurt product with plant (no bacteria), the sample with %1 and finally %0 tarragon were higher, respectively. So, during refrigeration the control sample had the most duration and the yoghurt sample containing %3 tarragon had a minimal duration.

In the direct counting method of the bacteria, maximum numbers of microbes were observed in the control sample and subsequently the sample with 2%, yoghurt product with plant (no bacteria) and finally the sample with %3 tarragon were more, respectively, in the first week.

Maximum colony growth were observed in the sample with %2 tarragon and subsequently yoghurt product with plant (no bacteria), the sample with %1, %0 and %3 tarragon were more, respectively.

Although, the basic feature of the probiotic products consumption is their medicinal effects (biovalue), their associated sensory properties are also important. In other words, sensory properties rather than medicinal effects play the most important role in their daily consumptions.

Among the probiotic products, fermented ones especially the probiotic yoghurt is popular worldwide for its unique sensory properties^[1]. In the present study, the sensory properties investigated in all the samples including the controls, were aroma, scent, color, thickness and taste.

The results of the questionnaires filled by 30 people showed that the control sample and subsequently the sample with %1 and %2 tarragon, yoghurt product with

plant (no bacteria) and the sample with %3 tarragon were more popular in aroma, scent, color, thickness and taste, respectively.

To evaluate the effect of tarragon on the amount of propioteic yoghurt protein, the control sample and the yoghurt sample containing %3 tarragon were tested, it was observed that the effect of tarragon on the amount of probiotic yoghurt protein in the yoghurt sample containing %3 tarragon was more than testifier.

The results of the studies addressing the probiotic bacteria have demonstrated the following: The increased concentration of malt and soya caused increase in the microorganism growth and rising acidity level which in turn resulted in shorter incubation time for the desired acidity^[11].

The effect of honey on the growth of the above mentioned bacteria introduced simultaneously into dairy products and drinks was investigated, and the results indicated that the yoghurt with only *Lactobacillus acidophilus* tasted sourer than the yoghurt with both bacteria. The products containing *Bifidobacterium bifidum*, compared to those with *Lactobacillus acidophilus*, were with slower growth rate and also tasted less sour and were of longer permanence.

They were not of favorable taste when honey concentration increased and the control was of the best taste among all the samples^[13]. In another study addressing the effect of cinnamon on the bacterial growth, it was demonstrated that the increased cinnamon concentration promoted the growth of the bacteria in probiotic milk and yoghurt^[12].

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FULL PAPER

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