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Research Article



PREPARATION OF CURD IN THE PRESENCE OF EASILY AVAILABLE PREBIOTIC SOURCES AND STUDY OF THEIR EFFECT ON PHYSIOCHEMICAL, SENSORY AND MICROBIOLOGICAL PROPERTIES OF THE CURD

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Accepted on: 06-09-2012; Finalized on: 31-10-2012.

ABSTRACT

Dahi or curd is one of the most important fermented milk products in India and have highly beneficial effects on human health. Probiotics are live friendly microorganisms in the gut microflora and Prebiotics are non-digestible oligosaccharides. They have been demonstrated to positively modulate the intestinal microflora and when used together they confer a synergistic effect on the host's health. The aim of this study was to determine the effect of easily available prebiotic sources on the physiochemical, sensory and microbiological properties of curd. The curd was prepared in the presence of four different kinds of prebiotic sources which were wheat, oat, barley and soybean. The effects on Organoleptic, chemical and microbiological properties were studied. The chemical properties were indicated in terms of pH and acidity and the microbiological properties were indicated by the growth of *Lactobacillus* spp. in the MRS media. All the Prebiotic materials were shown to enhance the growth of *Lactobacillus* spp. The Organoleptic and chemical properties of the curd containing prebiotic was found to be acceptable, when compared to the normal curd.

Keywords: Curd, Microflora, Organoleptic, Oligosaccharides, Prebiotics, Probiotics.

INTRODUCTION

Dahi or curd is one of the most important fermented milk products in the Indian sub-continent and the neighbouring countries and have been used since the time immemorial. In Ayurvedic system, curd has been strongly recommended for curing ailments like dyspepsia, dysentery and other gastrointestinal disorders. This product is also believed to improve appetite and vitality. Some of the beneficial effects of curd are attributed to the antibacterial components formed during the fermentation, low pH that prevents the growth of putrefactive and other undesirable organisms including potential pathogens and increased digestibility.¹⁻³ Curd is a good source of B vitamins, proteins, and calcium which are much easier for the body to digest than when they are present in fresh milk. It is, therefore, an ideal diet for those with sensitive digestive systems particularly young children and elderly persons.

A diversity of microorganisms inhabits our gastrointestinal environment. These different microorganisms play a key role in maintaining microbial balance in the intestine and host's overall health.⁴ Probiotics were defined as "Live micro organisms which when administered in adequate amounts, confer a health benefit on the host".⁵ The probiotic organism has the property of being non-pathogenic, non-toxic, resistant to gastric juice and should produce antimicrobial substances to prevent the growth of unwanted/harmful microorganisms. The potential benefits of Probiotics include lactose digestion in lactose intolerant people, prevention of colon cancer, lowering cholesterol, lowering blood pressure, improving immune functions, vitamin production and preventing infections.⁶

Prebiotics on the other hand are non-digestible oligosaccharides which are resistant to direct metabolism by the host and reach the caeco-colon intact where they are preferentially utilized by selected groups of beneficial bacteria and enhance their colonization. Among the identified prebiotic components, inulin and Fructooligosaccharide (FOS) is found as a natural component in several edible fruits and vegetables. Traditional dietary sources of prebiotic include soybeans, inulin sources (such as Jerusalem artichoke, jicama, and chicory root), raw oats, unrefined wheat, unrefined barley and yacon.^{7,8}

Our study combines the beneficial effects of both Curd (Dahi) and the Prebiotics. The curd was prepared in the presence of wheat, oat, barley and soybean prebiotic sources and then the effects were studied.

MATERIALS AND METHODS

Preparation of Prebiotic Powder: The seeds were collected from the local market of Vellore. Four kinds of seeds were collected which were Soybean, Barley, Wheat and Oat. All the collected seeds were weighed in equal amount and then washed with distilled water twice. The washed seeds were soaked in water for 12-18 hours. After soaking, the water was drained and the seeds were sun dried for 2-3 days. For complete drying, the seeds were kept in hot air oven at 65-70°C for 24 hours. Complete drying of the seeds was ensured. All the dried seeds were ground to powder in a grinder and were sieved to uniform size. Also the powders of all the four seeds (Barley, Soybean, Wheat and Oat) were mixed in equal amount to make a mixture. The powders were stored at room temperature in sterile air tight containers for further use in experiment.



Curd Preparation: The inoculum was obtained from the Nilgiri's curd for the setting of curd in the presence of prebiotics. The buffalo's milk was used because of its high fat content. The milk was filtered and then boiled at 80-90°C for 10-15 minutes. Then it was allowed to cool down to temperature of 45-50°C. 40 mL of milk was taken in different vessels and prebiotic powder of each of the seeds was added and mixed properly. 0.5% of inoculum was added in each and every vessel and also in vessel without any prebiotic, labelled as control. The caps of the vessels were tightly closed and incubated for 6-8 hours at 37-40°C.

Chemical Properties of Curd Samples

pH of Curd Samples: The pH of the curd samples obtained was measured by using the pH meter.

Acidity of Curd Samples: The acidity was measured by adding 1 mL of phenolphthalein indicator to 9 gm of curd and titrating it against 0.1 N NaOH.

Organoleptic Properties of Curd Samples: A taste panel of 15 local tasters were formed and an organoleptic test was conducted. The panel included 6 females and 9 males in the age group of 18-20 years. A blind test was performed and each member in the panel rated the eleven curd samples including the control on a 5-point scale (5= extremely like to 1=extremely dislike) for 5 attributes (appearance, aroma, texture, taste, overall acceptability). Water was provided in between two samples to rinse the mouth. A fresh batch of each curd sample was prepared for this test.

Microbiological Properties: 1mL of curd from the samples was serially diluted up to 10^{-4} dilutions. Then 0.1 mL from 10^{-3} or 10^{-4} dilution was used for pour plate method of enumeration of bacterial colonies. The media used was MRS Agar media which favours the growth of *Lactobacillus* spp. The plates were incubated at 37°C for 36 hours.

RESULTS AND DISCUSSION

Chemical Properties of curd samples

A good-quality Dahi has a pH of 4.4-5.0 and the titratable acidity of 0.6-1.0 %. The pH and acidity of control was 4.56 and 0.70 % respectively. The minimum pH was found for curd containing wheat 0.5gm (4.27) and the maximum was for curd containing oat 0.25gm (4.48). All the pH values are in normal range. The reason for the acceptability of the oat samples was due to its less acidity. Samples containing more amount of prebiotic material are comparatively more acidic than those containing lesser amount. This may be due to greater availability of proteins and more growth of lactic acid bacteria in the samples. The pH and acidity values for all the samples are given in table 1.

Organoleptic Properties of curd samples

One way ANOVA (Analysis of Variance) was carried out, in order to know if the differences observed among

different curd samples with different prebiotic materials are statistically significant. Through the organoleptic analysis of the curd samples, it was found that the curd sample containing oat as the prebiotic source was the best sample with the highest rating for overall acceptability and taste. For the other samples the overall acceptability was in the range of 2.73-3.8. The soybean containing curd sample got the least rating for taste and consumer's overall acceptability. It was found that the data were not significant and all the samples were comparable to that of control. The results are shown in table 2.

Table 1: pH and acidity values for different samples

Samples	pH	Titratable Acidity (%)
Curd control	4.56	0.70
Curd with Wheat 0.25gm	4.33	0.82
Curd with Oat 0.25gm	4.48	0.72
Curd with Soybean 0.25gm	4.47	0.73
Curd with Barley 0.25gm	4.40	0.76
Curd with Mixture 0.25gm	4.40	0.75
Curd with Wheat 0.5gm	4.27	0.83
Curd with Oat 0.5gm	4.29	0.80
Curd with Soybean 0.5gm	4.33	0.81
Curd with Barley 0.5gm	4.37	0.77
Curd with Mixture 0.5gm	4.34	0.79

Microbiological Properties of Curd samples

The starter or Khatta (Jaman) used for the preparation of dahi is a mixture of lactic streptococci and lactobacilli. The organisms commonly found in the inoculum are *Lactococcus. cremoris*, *L. lactis*, *S. thermophilus*, *Lactobacillus acidophilus*, *L. bulgaricus* and *L. helveticus*. Population and types of lactic acid bacteria occurring in samples of dahi vary in different region of India. MRS media used was specific for the growth of *Lactobacillus* spp. The colonies on MRS Agar plate were round shaped, irregular and white. The CFU/mL of curd sample for the control was found to be 46×10^4 . The colonies in the plates containing prebiotics were higher than the control. The maximum number of colonies was found in the curd containing 0.5 gm of soybean powder and the minimum number of colonies was found in the curd containing 0.25gm wheat. Overall, the number of colonies was more for samples containing greater amounts of prebiotic sources. All the values are given in table 3.

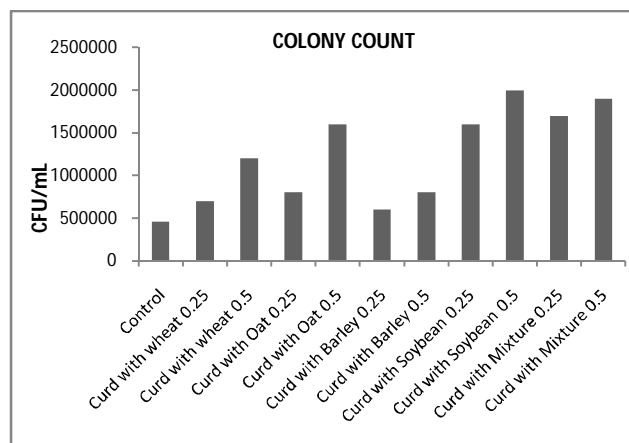


Table 2: Rating chart for different samples showing the mean values

PREBIOTIC SOURCE	CONTROL	0.25gm POWDER	0.5gm POWDER	F-RATIO ($\alpha=0.05$)
ATTRIBUTE: 1. APPEARANCE				
WHEAT	3.66±0.723*	3.53±1.091	3.67±0.861	NS**
OAT	3.66±0.723	3.73±0.798	3.6±1.121	NS
BARLEY	3.66±0.723	3.46±1.112	3.2±1.320	NS
SOYBEAN	3.66±0.723	3.46±0.915	4.0±1.069	NS
MIXTURE	3.66±0.723	3.33±0.736	4.26±0.743	NS
ATTRIBUTE: 2. AROMA/FLAVOUR				
WHEAT	4.0±0.845	3.53±1.246	3.33±0.723	NS
OAT	4.0±0.845	3.73±0.961	3.4±1.121	NS
BARLEY	4.0±0.845	3.27±1.099	3.2±1.014	NS
SOYBEAN	4.0±0.845	3.33±1.112	3.13±1.055	NS
MIXTURE	4.0±0.845	3.53±0.941	3.87±0.798	NS
ATTRIBUTE: 3. TEXTURE				
WHEAT	4.07±0.798	3.47±1.060	3.33±0.899	NS
OAT	4.07±0.798	4.0±0.593	3.4±0.991	NS
BARLEY	4.07±0.798	3.33±0.723	3.06±1.22	NS
SOYBEAN	4.07±0.798	3.46±0.833	3.26±0.990	NS
MIXTURE	4.07±0.798	3.33±0.828	4.06±0.676	NS
ATTRIBUTE: 4. TASTE				
WHEAT	3.8±0.861	3.13±1.302	3.06±0.798	NS
OAT	3.8±0.861	3.67±0.828	3.33±1.234	NS
BARLEY	3.8±0.861	3.13±0.883	2.86±1.125	NS
SOYBEAN	3.8±0.861	2.93±1.162	2.73±1.437	NS
MIXTURE	3.8±0.861	3.06±0.816	3.46±0.985	NS
ATTRIBUTE: 5. OVERALL ACCEPTABILITY				
WHEAT	3.8±0.676	3.13±1.187	3.13±0.703	NS
OAT	3.8±0.676	3.80±0.861	3.26±1.032	NS
BARLEY	3.8±0.676	3.13±0.961	3.06±0.961	NS
SOYBEAN	3.8±0.676	3.67±0.925	2.73±1.221	NS
MIXTURE	3.8±0.676	3.06±0.975	3.73±0.676	NS

*Mean±Standard Deviation; **NS-Not-Significant (P<0.05)

Table 3: Colony count for the different samples

Samples	CFU/mL
Curd control	46 x 10 ⁴
Curd with Wheat 0.25gm	7 x 10 ⁵
Curd with Oat 0.25gm	8 x 10 ⁵
Curd with Soybean 0.25gm	16 x 10 ⁵
Curd with Barley 0.25gm	6 x 10 ⁵
Curd with Mixture 0.25gm	17 x 10 ⁵
Curd with Wheat 0.5gm	12 x 10 ⁵
Curd with Oat 0.5gm	16 x 10 ⁵
Curd with Soybean 0.5gm	20 x 10 ⁵
Curd with Barley 0.5gm	8 x 10 ⁵
Curd with Mixture 0.5gm	19 x 10 ⁵

CONCLUSION

Dahi is one of major indigenous fermented milk products, which is palatable, refreshing, and has characteristic taste which is liked by the people of all age group. People who are lactose intolerant can also consume these products, if not for their therapeutic quality but for the essential and satisfying nutrients required in a daily diet. In addition, it has also been recommended for replenishing the normal flora in the intestine after having heavy doses of antibiotics.

Combining Dahi or curd with the prebiotics will be beneficial for human health. This will not only enhance the growth of beneficial microorganisms in the gut but also improve the gut health. The consumer's overall acceptability for the product was satisfying and also the

pH and acidity was in normal range. The best quality curd was the one containing oat as the prebiotic source. The oat also contains higher percentage of dietary fibres. The least liking of soybean curd may be because of its bitter taste and beany smell but it has shown higher growth of *Lactobacillus spp.* as compared to others.

By either developing new and innovative products or just reformulating existing ones, nutritional food ingredients enable manufacturers to meet and exceed the expectations of today's health-conscious consumer. Cereals not only have the ability to grow and deliver probiotic lactic acid bacteria to the human gut, but also contain potentially prebiotic compounds whose functionality should be explored.



Acknowledgements: The Authors would like to express their gratitude towards Honorable Chancellor, VIT University, Dr. G. Viswanathan, for providing necessary infrastructural lab facilities and encouragement to carry out this research work.

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