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Development and Physico-Chemical Analysis of Fortified Vegan Probiotic Fruit Yogurt with Ashwagandha (*Withania Somnifera*)

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ABSTRACT

The study of Herb fortified Probiotic Vegan Fruit Yogurt development was carried out at the Department of Food Technology, Parul Institute of Applied Science, Parul University, Vadodara. The main idea of study was to develop a herb fortified fruit yogurt as it can be served to the Lactose-Intolerant group of people with minor or no symptoms as well those who have diverted their diet towards vegan. Herb fortified yogurt was developed using Ashwagandha (*Withania somnifera*) and Fig (*Ficus carica*). Yogurt provides immunity, helps in reducing high blood pressure and improves digestion and in addition, ashwagandha is a condiment that provides Immuno- boosting, anti-inflammatory and anti-cancer properties. It helps in reducing blood sugar level and cholesterol levels. This yogurt was prepared by adding five strains of probiotic culture (*Lactobacillus acidophilus, Lactobacillus Rhamnosus, Bifidobacterium Bifidum, Bifidobacterium Longum, Streptococcus thermophilus*) in pasteurised milk in addition, Ashwagandha condiment and Fig pulp were further added. Three distinct formulations (T_0, T_1 and T_2) were developed with different formulation of Ashwagandha (0.5%, 1% and 2%) and Fig pulp (5%, 20% and 30%). The named expression was further analysed for physio- chemical and microbiological characteristic. Herbal probiotic fruit yogurt with T_1 formulation with 1gm of Ashwagandha and 20 ml of fig crush was set up to be best on the basis of sensory evaluation like taste, texture, colour, mouth feel, and overall acceptability. The herbal fruit yogurt contains fat 0.69%, Protein 1.20% and Ash 0.17%. The overall acceptability of herbal fruit yogurt was found to 8.03.

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1. INTRODUCTION

It is essential to create innovative products that can catch consumers' imaginations, have greater convenience, and offer superior nutrition due to rising consumer awareness and changing lifestyles (Sarkar, 2008). In contrast to the traditional definition of a vegetarian, a vegan diet excludes dairy products such as milk, cheese, butter, and eggs, as well as dishes containing those ingredients. Vegans now have access to a wider variety of substitutes, albeit not all of them may taste exactly like traditional milk and dairy products (Hunt, 1992).

Demand for yoghurt and other fermented products has recently increased as consumers become more health conscious. In India, the yoghurt market expanded at a CAGR of 28.9% between 2011 and 2015 (Tech Sci Research Report). Yogurt supports the growth of beneficial microorganisms that enhance intestinal health. Numerous bodily processes, including digestion, are influenced by the microbial ecology of the gastro-intestinal tract (GIT). Yogurt is a wonderful option for the development of functional foods because of all these factors (Jaziri *et al.* 2009). Yogurt production begins with the careful selection of raw materials and the precise formulation of a liquid mix that is specific to the type of yoghurt being produced. (United States Department of Health and Human Services, 2013).

Ashwagandha (*Withania somnifera*, fam. Solanaceae) is commonly known as "Indian Winter cherry" or "Indian Ginseng". It has been used for millennia as a rasayana for its extensive range of health effects, making it one of the most significant herbs in Ayurveda (India's traditional medical system). The anti-stress compounds sitoindosides and acyl steryl glucosides are found in ashwagandha (Verma, K. C.,2010). Ashwagandha is a well-known herb, it helps with memory and the way the brain and nervous system work. By making the reproductive system more effective, it encourages a balanced sexual and reproductive system. It boosts the body's capacity to withstand stress as a result of its powerful adaptogenic capabilities. Additionally, it has strong antioxidant qualities that aid in preventing cellular damage brought on by free radicals (Narendra Singh, Mohit Bhalla, Prashanti de Jager and Marilena Gilca,1938). Ashwagandha was used to make herbal yoghurt that also contained probiotic bacteria. (Behrad, S., Yusof, M. Y., Goh, K. L., Baba, A. S,2009).

Agar, a plant-based, gel-like product made from red algae, is the ideal vegan alternative to gelatin. Agar is primarily used as a gelling agent and secondarily as a stabilising agent and for viscosity control in the human food business. It is used to make candies, marshmallows, and jellies for confectionery. (Yudkin, 1986).

According to USDA data, anjeer is a good source of fibre, copper, manganese, magnesium, potassium, calcium, and vitamin K. Figs are frequently recommended to hydrate, tone, and act as a natural laxative on the intestines due to their high fibre content. Additionally, they are a good source of flavonoids and polyphenols (Vinson et al. 2005).

Coconut milk is a milky white, oil-in-water emulsion made from coconut kernel products, with water and lipids making up the majority of its ingredients (Tansakul and Chaisawang, 2006). Due to their more environment friendly manufacture than milk from cows, plant-based milk substitutes have seen a huge increase in popularity among consumers (Raikos et al., 2020). Coconut milk is a lactose-free and healthy component, is a wonderful substitute for dairy milk in yoghurt. Because medium-chain triglycerides from fatty acids make up more than 50% of the fat, it is easily digested. Additionally, glutamic acid, aspartic acid, arginine, and antioxidants are found in coconut milk, along with calcium, phosphorus, potassium, and vitamins C, E, and B (Yuliana *et al.,* 2010).

Coconutmilk-based yogurt is produced by incorporating the Ashwagandha and Anjeer followed by fermentation of coconut milk using probiotic (*Lactobacillus acidophilus, Lactobacillus Rhamnosus, Bifidobacterium Bifidum, Bifidobacterium Longum, Streptococcus thermophilus*) bacteria.

2. MATERIALS AND METHODS

2.1. Materials

The research work was conducted in the Food Processing Laboratory, at Parul Institute of Applied Science, Parul University, Vadodara, Gujrat.

Coconut Milk (Pantai norasingh, Thailand), Ashwagandha (Patanjali, Uttarakhand), Anjeer Crush (Shree Yogi Anjeer Crush) and Agar (Bacsgo Agar Agar Powder) were obtained from a local supermarket in Vadodara, Gujrat. According to the labeled composition, 100 ml of coconut milk are composed of 19 g fat, 2 g carbohydrate and 1 g protein. The yogurt starter culture, Urban Platter Yogurt Culture Capsules, Contains a blend of bacteria like Lactobacillus acidophilus, Lactobacillus Rhamnosus, Bifidobacterium Bifidum, Bifidobacterium Longum, Streptococcus thermophilus and Fructo-oligosaccharides.

2.2. Preparation of Yogurt from Coconut Milk

The mixtures of coconut milk, Ashwagandha and Agar were heated to 90 °C for 3min (Yaakob et al., 2012), cooled to 43^{0}C, and the probiotic culture was added and incubated at 43^{0}C and after fermentation the product was stored at 4^{0}C. Using the Sensory, Chemical and Microbial evaluation the product was analysed and three discrete formulations were formed for the Analysis as given in Table 2.2.1.

Table.2.2.1.:- Formulations used for preparation of yogurt sample.

Constituents	Coconut Milk (ml)	Ashwagandha (gm)	Agar (gm)	Anjeer Crush (ml)	Probiotic Culture
T_0	100	0.5	6	5	1 Tablet
T_1	100	1	4	20	1 Tablet
T_2	100	2	3	30	1 Tablet

Note: 1 Tablet of probiotic culture contains 0.7 billion % of each species values based on 2320 calories diet.

2.3. Physical Analysis

Food preferences have been measured using a variety of techniques. The most popular technique involves creating a questionnaire with randomly selected items or food groups and using a hedonic scale to score the

degree of enjoyment. An organoleptic measure known as the hedonic scale allows the judge to describe his level of preference.

A 9 point balanced scale was used. Hedonic nine-point scales were used for the overall tests. The general form of the scale: 1. Dislike extremely, 2. Dislike very much, 3. Dislike moderately, 4. Dislike slightly, 5. Neither like nor dislike, 6. Like slightly, 7. Like moderately, 8. Like very much, 9. Like extremely. Appearance, colour, texture, Taste and overall acceptance tests of the samples were accomplished. Both trained and unskilled panellists had used this test. The 8 untrained panelists were teachers and students of Department of Food Technology, Parul Institute of Applied Science, Vadodara, Gujrat. Hedonic scaling uses a rating scale to quantify response, or the state of liking and disliking. The hedonic scale's fundamental characteristics were its premise of a continuum of preferences and its straightforward definition of the response categories in terms of likes and dislikes.

2.4. Chemical Composition Analysis

Chemical quality evaluation of the chosen sample was performed. Ash was determined using the incineration method, fat was determined using the modified Rose- Gottlieb Method, and protein was determined using the Kjeldahl method. The oven-drying technique was used to measure moisture.

2.5. Microbiological Analysis

The sample was conducted by various microbial parameters. Total Plate Count, Total Coliform Count (TCC), Total Yeast and Mould Count (TFC), Detection of E.Coli was done according to the standard methods for the examination (H. Michael Wehr and Joseph F. Frank.,2004)

2.6. Data Analysis

Data were collected, compiled, tabulated, and examined from all angles. With the aid of Microsoft Excel 2021, a statistical analysis of the data produced by the chemical analysis of sample was carried out. The regional units were changed to international units. The qualitative information was converted into quantitative information. For data analysis and presentation, Microsoft Excel 2021 and Word 2021 were used. The variable was categorised and described using a variety of descriptive statistical measures, including percentage, mean, standard deviation (SD), and others. To present the results, various tables, graphs, charts, etc. were used.

3. RESULTS AND DISCUSSION

3.1. Physical Assessments of Different Types of Prepared Yogurt

Organoleptic tests of the prepared yogurt depend on its appearance, colour, texture, taste and overall acceptability of the sample. The comparison among prepared yogurt of their organoleptic quality factors are presented on Table-3.1.1.

All types of prepared yoghurt had acceptable band scores overall, although the test panel board found modest differences in some of their specific qualities.

Table-3.1.1.: Comparison of Physical Qualities of Different Types of Prepared Yogurt.

Constituent	T_0	T_1	T_2
Appearance	8	9	7
Colour	8	9	8
Texture	7	9	8
Taste	7	9	8
Overall Acceptability	7.5	9	7.75

3.1.1. Appearance

In Table 3.1.1., the physical rating and a comparison of the visual appeal of several varieties of prepared yoghurt are provided. Average appearance score of prepared yogurt samples T_0, T_1 and T_2 were 7.5, 9 and 7.75 respectively. Highest appearance score was recorded for T_1 sample of prepared yogurt respectively. The milk, ashwagandha, agar, and processing method can all affect how yoghurt looks. In the appearance acceptability test, the hedonic scale showed that all sorts of yoghurt samples were liked very much (>7) by the judge, as shown in Table 1.

3.1.2. Colour

Colour is a crucial visual component of food products. Table 3.1.1. provide the physical rating and a comparison of the colour of several varieties of prepared yoghurt. Average colour score of prepared yogurt samples T_0, T_1

and T_2 were 8, 9 and 8 respectively. The colour of the milk or other colouring agents, as well as the colour of the crushed anjeer, determine the colour of the finished yoghurt. The results of the colour acceptability test, which are displayed in Table 3.1.1., indicate that the judge enjoyed all types of yoghurt samples very well (>7).

3.1.3. Texture

Table 3.1.1., provide the physical scores for the texture of various varieties of prepared yoghurt. Average texture score of prepared yogurt samples T_0, T_1 and T_2 were 7, 9 and 8 respectively. By using a hedonic scale, the judges gave the texture of all varieties of prepared yoghurt an average rating of "very much liked" (>7). Highest (9) texture score was recorded for T_1 type of prepared yogurt. Texture of the yogurt may be depend upon the rate of development of the acidity as well as on the amount of agar added.

3.1.4. Taste

Table 3.1.1. and Figure 3.1.5.1. contain the physical rating and a comparison of the flavours of several varieties of prepared yoghurt. Average taste score of prepared yogurt samples T_0, T_1 and T_2 were 7, 9 and 8 respectively. Highest and lowest taste score were recorded for T_1 and T_2 types of prepared yogurt respectively. It was observed that, the addition of Anjeer crush increased the flavour score.

3.1.5. Overall Acceptance

Consumer approval was also influenced by the quality of the raw ingredients used in the production of prepared yoghurt. Table 3.1.1. and Figure 3.1.5.1. show the physical score for the general acceptance of several varieties of prepared yoghurt. Average overall taste score of prepared yogurt samples T_0, T_1 and T_2 were 7.5, 9 and 7.75 respectively. The T1 sample of prepared yoghurt outperformed the T0 and T2 samples according to the hedonic scale. The sensory evaluation found that yoghurt containing up to 1% Ashwagandha and 4% Agar was acceptable. Finally, the study demonstrated that the panellists preferred and accepted the T_1 (1% Ashwagandha, 4% Agar) sample.

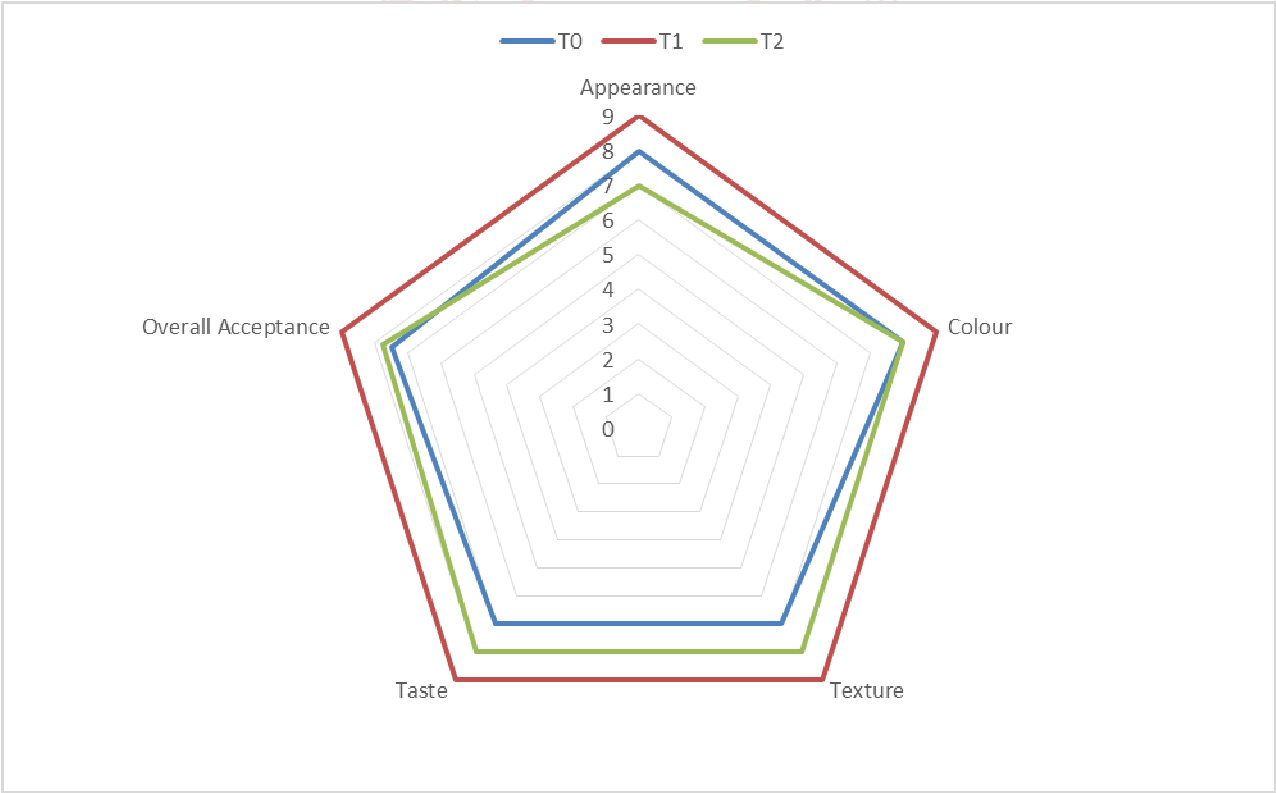

Fig. 3.1.5.1.: Comparison of the Sensory Evaluation of Different Yogurt Samples.

Based on the results from the sensory evaluation using 9 point hedonic scale, the yogurt made with addition of 1% Ashwagandha and 4% Agar received the highest score in overall acceptability from untrained penalists. So, the product with 1% Ashwagandha and 4% Agar selected for the proximate analysis.

3.2. Physico-Chemical Assessments of Prepared Yogurt.

Chemical composition of prepared yogurt is given in Table 3.2.1. Moisture, protein, fat, ash, sample was analysed for their chemical qualities.

Table-3.2.1.: Chemical Composition of Prepared Yogurt.

Sr. No.	Quality Characteristic	Units	Results
1.	Moisture	%	77.84
2.	Total Ash	%	0.17
3.	Fat	g/100g	0.69
4.	Protein	g/100g	20.1
5.	Carbohydrate	g/100g	20.01
6.	Energy	KCal/100g	91.41
7.	Lactose	%	4.21

3.3. Microbiological Assessments of Prepared Yogurt.

In this study 4 different tests Total Plate Count, Yeast and Mould Count, E.coli Detection and Total Coliform Count were conducted to analyse the microbiological quality of prepared yogurt sample. Microbial composition of prepared yogurt is given in Table 3.

Table-3.3.1.: Microbial Composition of Prepared Yogurt

Sr. No.	Quality Characteristic	Units	Results
1.	Total Plate Count	Cfu per g	82,900
2.	Yeast and Mould Count	Cfu per g	400
3.	Total Coliform Count	Per g	Absent
4.	Escherichia Coli (Detection)	Per g	Absent

4. CONCLUSION

Based on organoleptic, chemical, and microbiological examination, the study was conducted with different concentrations of Ashwagandha (0.5%, 1.5%, and 2%), as well as Anjeer crush (5%, 20%, and 30%). The appearance, colour, flavour, and texture of yoghurt with 1% Ashwagandha and 20% Anjeer Crush were acceptable than those containing 0.5% and 2% Ashwagandha and 5% and 30% Anjeer Crush, according to a nine-point hedonic scale of organoleptic qualities. According to result obtained T_1 was the most effective sample out of all formulations with varying amount of ashwagandha and anjeer crush which contains Energy 91.41 Kcal, fat 0.69%, Protein 1.20% and Ash 0.17%. The safety of all products is further ensured by microbiological quality measures, and in the case of total plate count, yeast and mould count, and total coliform count, these values were within permissible limits. So, all of the yoghurt samples that had been created were safe to eat. From the above research it can be concluded that the developed vegan probiotic yogurt can also be consumed with peoples with lactose intolerance, normal peoples as well as peoples with vegan diet.

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