PREPARATION OF WHEY-BASED BEVERAGES; AN ALTERNATIVE TO SOFT DRINKS

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A production report submitted as per approved styles and contents

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TABLE OF CONTENTS

Contents	Page no

List of Tables

Abstract

The disposal of whey, a byproduct of the cheese-making process, poses significant environmental challenges due to its high Biological Oxygen Demand (BOD) value and stringent regulatory requirements. However, recent efforts have focused on repurposing whey due to its potential applications in various industries. Whey proteins, known for their foaming, emulsification, nutritional, and biological attributes, have gained attention. Whey proteins, with unique nutritional benefits and neutral pH characteristics, are employed in ready-to-drink beverages like sports drinks, smoothies, and meal replacements. Despite their relatively small quantities, whey proteins exhibit high protein efficiency ratio, net protein utilization, and biological value, rivaling egg protein. The composition of whey proteins includes various bio-active compounds with potential health benefits. The exceptional solubility of whey proteins across a wide pH range further enhances their utility. In this study, milk and whey samples were comprehensively analyzed. Milk composition parameters such as lactometer reading, specific gravity, acidity, protein, fat, solid-not-fat percentage, and total solids percentage were measured. Whey samples were assessed for acidity, protein, fat content, as well as the volumes of vinegar and water present. Furthermore, the sensory characteristics of two different flavors, cardamom and vanilla, were evaluated based on color, flavor, taste, and odor. The study's findings underscore the potential of whey proteins for use in various beverages, owing to their unique properties. However, limitations include sample size and representativeness, methodology constraints, external validity, and potential environmental influences. Recommendations include expanding sample diversity, refining methodologies, exploring broader product ranges, considering environmental factors, and conducting longitudinal studies for more robust results. In conclusion, the study contributes to the understanding of whey's applications and underscores the need for further research to address its limitations and promote its sustainable utilization in the beverage industry.

Keyword: Whey Production, Environmental Sustainability, Health Benefits, Sensory Evaluation

Introduction

Whey or milk plasma is a greenish yellow, semi-translucent liquid that separates from the curd during the cheese making process. It is one of the major problematic disposals for dairy industry because of high Biological Oxygen Demand (BOD) value ranging from 39,000 to 48,000 ppm according to Divya and Kumari et al. (2009) and its stringent environmental regulatory acts. The world produces more than 80*10^9 L of whey, which is dumped into rivers and lakes, according to Smithers et al. (1996). Recent years have witnessed considerable efforts directed towards redefining the potential applications of whey to mitigate environmental pollution (González-Martínez et al., 2002; Douaud, 2007; Jeličić et al., 2008). Whey and its protein concentrates have found valuable roles as ingredients within the food industry, primarily due to their capacity for foaming and emulsification (Hall and Iglesias, 1997; Ji and Hauque, 2003; Jovanović et al., 2005), as well as their nutritional and biological attributes (Mistry et al., (1996); Smithers et al., (1996); Kenny et al., (2001); Carunchia Whetstine et al., (2005); Herceg et al., (2008); Akpinar-Bayizit et al., (2009). Notably, whey proteins are believed to encompass biological functions such as cancer prevention, enhancement of glutathione levels, antimicrobial capabilities, and stimulation of the satiety response (Valli and Trail, 2005; Madureira et al., 2007).Tryptophan, an important amino acid that helps increase brain serotonin levels, is particularly abundant in the proteins made from whey (Delgado-Andrade et al., 2006). Probiotic bacteria have been shown to provide nutritional and health advantages, including quicker recovery from diarrhea, protection against lactose intolerance, lowering cholesterol risk, and protection against urogenital infection (Anukam et al., 2006). The creation of these products requires meticulous sensory, physical, and chemical characterization to ensure quality control and facilitate effective product development (Gallardo-Escamilla et al., 2007). Whey proteins possess distinctive nutritional and functional attributes that can be harnessed to produce high-protein offerings for consumers. Their neutral pH characteristics make dairy proteins particularly well-suited for the production of beverages with diverse nutritional and functional profiles.The development of ready-to-drink (RTD) beverages hinges on the inherent heat stability of caseins at neutral pH levels. In contrast, whey proteins exhibit lower heat stability under similar conditions, explaining the prominence of caseins or milk proteins in ingredient lists. Whey protein chemical constituent percentages are α-Lactalbumin 11.3–16.5, β-Lactalbumin Lactoglobulin 37.9–49.0, IgG 5.0–8.0, Glycomacropeptides 15.0–20.0, Lactoferrin 1.3–1.8, Bovine serum albumin 3.0–5.0 (Gangurde, H., Chordiya, M., Patil, P., & Baste, N. (2011). Despite their smaller quantities, whey proteins possess a high protein efficiency ratio (3.6), net protein utilization (95), and biological value (104). When compared to other protein sources, they closely follow egg protein in terms of nutritional value. Whey proteins contain compounds like α-lactalbumin, β-lactoglobulin, bovine serum albumin, caseinomacropeptides, immunoglobulins, lactoferrin, and lysozyme, often associated with health benefits, including enhanced immunity, anticancer properties, resistance against pathogens, antiviral and antimicrobial effects (through iron binding), as well as antihypertensive properties. The advantages of whey proteins can be strategically harnessed by beverage manufacturers due to their remarkable solubility within a pH range of 2 to 10. Furthermore, they offer stability and clarity even within the pH range of 3.0 to 3.2. Developers require minimal additional processes or ingredients at these lower pH levels, allowing them to optimize the inherent characteristics of whey proteins.

Objectives:

1. To evaluate the composition of milk and whey.
2. To test the acceptability of prepared whey-based beverages.

Materials and methods

Study area

The goal of the study was to reduce the amount of whey that confectioneries and sweet shops discharge into the environment. Whey proteins are commonly utilized in the creation of products such as sports drinks, beverages, smoothies, and meal replacements. Development of whey based beverages will decrease the environmental pollution and create an alternative source of income. Experiments and laboratory tests were conducted in the Laboratory of department of dairy and poultry science, CVASU. For the testing and creation of the whey-based drinks, Nahar Agro's milk sample was used.

Study period

Data and test results were gathered over the course of two weeks, and the whey beverages were subjected to a three-day sensory review.

Laboratory tests

Specific gravity of milk

The milk sample was thoroughly mixed. The milk was poured into the lactometer jar. The lactometer was placed inside the jar with a rotating motion, and the reading was taken when it reached a stationary position. The temperature of the milk was recorded using a dairy thermometer. The Corrected Lactometer Reading (CLR) was calculated by adding 0.2 for each degree Fahrenheit below 84°F. The lactometer reading was adjusted to 60°F by adding 0.1 degree Quevenne for each degree Fahrenheit above 60°F and subtracting 0.1 degree Quevenne for each degree Fahrenheit below 60°F.

Acidity of milk

The milk sample was mixed thoroughly. 10 ml of milk was taken in a porcelain beaker. 2-3 drops of phenolphthalein indicator were added to the sample. Titration was performed using 0.1N NaOH solution until a faint pink color appeared. The procedure was repeated three times. The volume of alkali used for each titration was recorded.

Protein and casein percentage of milk

10 ml well-mixed milk was taken in a conical flask. 0.4 ml potassium oxalate was added and mixed, and the mixture was allowed to rest for two minutes. 2-3 drops of phenolphthalein indicator were added, and titration was performed against 0.1N NaOH solution until a faint pink color appeared. 2 ml of formaldehyde solution was added, and the mixture was allowed to rest for 30 minutes. Titration was repeated following the same procedure after adding 2-3 drops of phenolphthalein indicator. The amount of alkali used was recorded, and protein and casein percentages were calculated.

Determination of fat percentage of milk by Gerber Methods

10 ml H_2SO_4 was taken in a butyrometer. 10.75 ml well-mixed milk sample was added. 1 ml amyl alcohol was added. The opening was closed with a lock stopper and shaken until the white particle disappeared. Centrifugation was done at 1100 RPM for 5 minutes. The fat column reading was recorded.

Estimation of SNF% and Total solids percentage in milk

Mix the milk sample well. Determine the fat % of milk by the Gerber method. Calculate the Corrected Lactometer Reading (CLR) at 77°F.Estimate the SNF% and TS% using the provided formula.

Preparation of Whey

Preparation

The milk is poured into a large pot and placed on the stove over medium heat. While the milk is being heated, the vinegar is prepared by measuring out approximately 1/4 to 1/2 cup of vinegar. The amount is adaptable based on the milk quantity being used.

Heating the Milk

The milk is gradually heated with occasional stirring to prevent adhesion to the pot's base. Temperature monitoring is facilitated using a thermometer. The milk is heated until it attains a temperature of around 180°F (82°C). This process serves to induce the denaturation of milk proteins, a precursor to curd formation.

Adding Vinegar

Once the desired temperature of the milk is reached, the vinegar is gently introduced while implementing stirring motions. The interaction of milk proteins with the acid in the vinegar initiates the aggregation of curds, characterized by their solid composition.

Curd Formation

Gentle stirring is sustained for a few minutes, ensuring a complete segregation of curds from the liquid component known as whey. Whey constitutes the aqueous fraction of the milk.

Draining the Curds

A colander is lined with a layer of cheesecloth or muslin fabric. Positioned above a bowl or sink, the colander serves as the site for whey collection.

Results

Descriptive analysis of milk composition

Lactometer Reading

The average corrected lactometer reading is 31.77, indicating the density or composition of the milk. The small standard deviation of 0.147 suggests that the readings are relatively close to the mean. The readings range from 31.61 to 31.9.

Table 1: Composition of milk

Variable	**Mean**	**Standard deviation**	**Min**	**Max**
Corrected Lactometer reading	31.77	0.14	31.61	31.9
Specific Gravity	1.0317	0.000152	1.0316	1.0319
Acidity	0.11	0.098	0.0017	0.18
Casein	2.28	0.62	1.58	2.77
Protein	2.94	0.80	2.04	3.57
Fat	3.93	0.057	3.9	4
SNF%	9.52	0.023	9.50	9.55
TS%	13.45	0.039	13.42	13.50
Whey (L)	1.41	0.028	1.4	1.45
Curds (kg)	0.26	0.016	0.25	0.28

Specific Gravity

The mean specific gravity of the milk is 1.0317, reflecting its density compared to water.The small standard deviation of 0.00015 indicates low variability in specific gravity.Values range from 1.0316 to 1.0319.

Acidity

The average acidity level is 0.114, representing the milk's acid content. The larger standard deviation of 0.098 suggests variability in acidity levels. Acidity values range from 0.0017 to 0.18.

Casein

The mean casein content is 2.28, indicating the presence of this protein in the milk. The relatively high standard deviation of 0.62 suggests some variation in casein content. Casein content ranges from 1.58 to 2.77.

Protein

The average protein content is 2.94, reflecting the protein concentration in the milk. The larger standard deviation of 0.80 indicates variability in protein levels. Protein content varies from 2.04 to 3.57.

Fat

The mean fat content is 3.93, representing the fat concentration in the milk. The small standard deviation of 0.057 suggests relatively consistent fat levels. Fat content ranges from 3.9 to 4.

Solid-Not-Fat (SNF)

The average SNF% is 9.52, indicating the portion of solids in the milk excluding fat. The small standard deviation of 0.023 suggests consistent SNF% values. SNF% ranges from 9.50 to 9.55.

Total Solids (TS)

The mean TS% is 13.45, reflecting the total solids content in the milk. The standard deviation of 0.039 indicates some variability in TS%. TS% values range from 13.42 to 13.50.

Whey

The average whey volume is 1.41liters, representing the liquid portion of whey. The small standard deviation of 0.028 suggests consistent whey volume. Volume ranges from 1.4 to 1.45 liters.

Curd

The average curds weight is 0.268 kilograms, representing the weight of curds formed. The small standard deviation of 0.0160 suggests relatively consistent curds weight. Weight ranges from 0.25 to 0.28 kilograms.

Overall, these statistics provide insights into the composition and characteristics of the milk sample, including its density, acidity, protein, fat, and other components. The variability indicated by standard deviations can offer an understanding of how consistent or varied these components are within the sample.

Whey composition

Whey Acidity

The average whey acidity is 0.28 %, indicating the acid content in the whey. The standard deviation of 0.137 suggests some variability in acidity levels. Whey acidity values range from 0.27 to 0.29.

Table 2: Composition of Whey

Variable	**Mean**	**Standard deviation**	**Min**	**Max**
Whey acidity	0.28	0.137	0.27	0.29
Whey protein	0.44	0.136	0.34	0.6
Whey fat	0.43	0.15	0.3	0.6
Vinegar (ml)	71	1.73	70	73
Water (ml)	71	1.73	70	73
Sugar (g)	78.67	6.1	72	84
Flavor (ml)	3.26	2.96	0	5.8

Whey Protein

The mean whey protein content is approximately 0.44 %, reflecting the protein concentration in the whey. The standard deviation of 0.136 indicates variability in protein levels. Whey protein content varies from 0.34 to 0.6.

Whey Fat

The average whey fat content is approximately 0.43 %, indicating the fat concentration in the whey. The standard deviation of 0.15 suggests variability in fat levels. Whey fat content ranges from 0.3 to 0.6.

Vinegar

The average volume of vinegar in the whey sample is 71 milliliters. The standard deviation of 1.73 suggests variability in the volume of vinegar. The volume of vinegar ranges from 70 to 73 milliliters.

Water

To create the whey sample, an average of 71 milliliters of water was added to the milk sample. The standard deviation of 1.73 indicates variability in the volume of water. The volume of water also ranges from 70 to 73 milliliters.

Sugar

This variable represents the amount of sugar in grams. The mean value is 78.67 g, and the standard deviation of 6.1 suggests some variability around the mean.

Flavor

This variable represents flavor in milliliters. The data seems to be more spread out, as indicated by the larger standard deviation. The values range from 0 to 5.8 ml.

Overall, these statistics provide insights into the composition and characteristics of the whey sample, including its acidity, protein, fat content, and the volumes of vinegar and water present. The standard deviations offer an understanding of how consistent or varied these components are within the whey sample.

Acceptability of prepared whey-based beverages

Flavor 1: Cardamom

Table 3: Summary of score-card judging of prepared whey-based beverages with cardamom Flavor

Variable	**Mean**	**Standard deviation**	**Min**	**Max**
Color	11.75	0.5	11	12
Flavor	15.5	2.64	12	18
Taste	30.5	1	30	33
Odour	13.5	2.38	10	15

Flavor 2: Vanilla

Table 4: Summary of score-card judging of prepared whey-based beverages With vanilla Flavor

Variable	**Mean**	**Standard deviation**	**Min**	**Max**
Color	12	1	11	13
Flavor	20.66	1.15	20	22
Taste	32	3.46	30	36
Odour	16	1.73	15	18

Flavor 2 generally had higher ratings in terms of flavor, taste, and odor compared to Flavor 1. Flavor 2 also showed slightly higher variability in taste, as indicated by the larger standard deviation for taste ratings. Flavor 1 had relatively consistent and less variable ratings for color, flavor, taste, and odor compared to Flavor 2.

Discussion

In this study, a comprehensive analysis of milk and whey samples was conducted using various methods to determine their composition and characteristics. Additionally, two different flavors, namely Cardamom and Vanilla, were evaluated for color, flavor, taste, and odor. Before testing, prepared drinks with different mixtures are refrigerated. A trained panel assessed the sensory qualities, which were specifically outlined by Poste et al. (1991), including color, taste, flavor, odour, and overall acceptability.

Milk and Whey Analysis

The determination of specific gravity, acidity, protein, casein, fat, SNF%, and total solids in milk involved a series of meticulous steps. The precision in handling the samples was evident from the small standard deviations in most parameters, indicating consistency within the sample set. Specific gravity, a measure of milk's density, demonstrated a remarkably low variability with a mean of 1.0317. The calculated Corrected Lactometer Reading (CLR) allowed the characterization of the milk's composition with a mean CLR of 31.77. The slight variations observed in acidity, protein, casein, and fat contents could be attributed to natural variations in milk composition, as seen in the standard deviations. This results are almost similar with (Jain et al. 2013) .The estimation of SNF% and total solids provided an understanding of the non-fat components in milk. The close agreement between SNF% and TS% results validated the accuracy of the methods used. The Gerber method for fat determination yielded consistent and precise results, showcased by the small standard deviation in fat content. This indicated a high level of accuracy in measuring the fat content of milk (James et al 1995).

Whey Analysis

The whey analysis highlighted the parameters of acidity, protein, and fat content. The whey's acidity, an important factor in determining its quality, exhibited minor variations as indicated by the standard deviation. The composition varies greatly depending on the availability of milk and the whey production technique. The whey sample had a mean acidity percentage of 0.28 %. Wheys are easily categorized into the following groups: Sweet whey: titratable acidity 0·10-0·20 per cent, Medium acid whey titratable acidity 0·20-0·40 per cent, Acid whey titratable acidity greater than

0·40 per cent Zadow, J. G. (1994). As a result, the whey sample falls into the category of medium-acid whey. Similarly, the protein and fat content in whey showed reasonable consistency despite some variability. The whey sample had a mean protein percentage of 0.44 % which slightly differs from study conducted by (Tsakali, E., (2010, June). The whey samples' mean fat content was 0.43%, which is just under the standard 0.5%. There should be more testing for the vitamins and minerals in the whey content as the water-soluble vitamins are incorporated into the whey during production in varied percentages: 40 – 70% of vitamin B12, 55% of pantothenic acid and vitamin B6, 70% – 80% of riboflavin and biotin, and 80 – 90% of thiamine, nicotinic acid, folic acid, and ascorbic acid. In the instance of vitamin B12, rennet coagulation rather than acid coagulation was utilized, which resulted in a greater transfer of the vitamin into the whey (Zadow, 1992). Whey is a good source of electrolytes. including sodium and potassium, which are required during recovery from diarrhea. Minerals such as calcium, magnesium, and phosphorus are present in the solution and are also partly bound to proteins. Zinc is present in trace amounts (Zadow, 1992). Additionally, Mg and zinc ions are better absorbed when there is lactose present, even in minimal levels (Ziegler and Fomon, 1983). The volumes of vinegar and water in the whey sample displayed relatively low standard deviations, as the samples were almost the same weight, indicating the careful addition and reproducibility of the process.

Flavor Evaluation

Flavor evaluations were conducted for two distinct flavors, Cardamom and Vanilla. The evaluations were based on color, flavor, taste, and odor. The results revealed insights into the sensory attributes of each flavor.

Flavor 1 (Cardamom) exhibited consistent color, flavor, taste, and odor ratings with relatively low standard deviations. This suggests a uniform sensory experience across the evaluators , making it a reliable choice for a consistent flavor.

Flavor 2 (Vanilla) displayed higher flavor, taste, and odor ratings, albeit with slightly larger standard deviations in taste. But, it appears that flavor 2 was favoured because the mean score across all the factors for flavor 2 is much higher than that for flavor 1.

Limitations

The study might not have included a large and diverse enough sample of milk and whey sources to ensure that the findings are broadly applicable. The samples used might not accurately represent the variability in milk and whey compositions found in different regions or production methods.

The methods used for determining various parameters (such as protein, fat, and acidity) might have their own limitations and potential sources of error. These limitations could impact the accuracy and reliability of the obtained results.

The study's findings might not be directly applicable to different types of whey-based products or to the broader food and beverage industry. Factors such as processing methods, ingredient combinations, and packaging could influence the results in real-world product development scenarios.

The study's focus on specific flavors might limit the generalizability of its findings to other flavors or taste preferences, potentially overlooking a broader range of consumer preferences.

The study might not have considered potential environmental factors that could influence the composition and characteristics of milk and whey samples, such as the diet and health of the animals producing the milk.

Recommendation

To address this limitation, future studies could aim to gather data from a more diverse range of milk and whey sources. This could involve collaboration with multiple dairy farms, production facilities, and regions to ensure a broader representation of milk and whey compositions. Increasing the sample size would enhance the study's external validity and applicability.

To mitigate methodology limitations, researchers should critically evaluate the methods used for parameter determination. Exploring alternative techniques and cross-validation with established methods can help ensure accurate and reliable results. Additionally, employing rigorous quality control measures during sample collection, preparation, and analysis can reduce potential sources of error.

To improve the external validity of the study, researchers could consider studying a wider range of whey-based products beyond the scope of the current research.

To address multiple limitations, longitudinal studies could be conducted over a longer time frame.

Conclusion

The comprehensive analysis of milk and whey samples, along with the sensory evaluation of flavors, provided valuable insights into their characteristics. The low standard deviations observed in most parameters for both milk and whey indicated the precision and reliability of the methods employed. This study serves as a foundation for understanding the composition and sensory attributes of these samples, which can guide future applications in various industries such as food and dairy.

Moreover, the flavor evaluation shed light on the sensory profiles of Cardamom and Vanilla flavors, aiding in the selection of flavors for specific products or preferences. This information can be beneficial for manufacturers aiming to create products with consistent sensory experiences or distinct profiles based on consumer preferences.

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Biography

I am Md Nahid Hasan, son of Md Nur Nobi and Aleya Begum. I completed my Secondary School Certificate from Bangladesh Bank Colony High School in 2014 and Higher Secondary School Certificate from Government Hazi Mohammad Mohsin College in 2016. I am an intern veterinarian at Chattogram Veterinary and Animal Sciences University, Bangladesh under the Faculty of Veterinary Medicine. I am very interested in veterinary medical research, and I want to use my abilities and creativity for the good of the nation. So that we can get past the obstacles we currently encounter with this field.