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STUDY ON OPTIMIZATION OF PROPORTION OF SKIM MILK POWDER AND STABILIZER IN PREPARATION OF WHEY YOGHURT AND EVALUATION OF ITS QUALITY

– Research paper –

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ABSTRACT: This study aims to optimize the utilization of whey, a significant dairy by-product from paneer production, in whey-based yogurt preparation. The primary goal is to identify the optimal ratio of skim milk powder and stabilizer (pectin) to minimize syneresis, a critical quality parameter, while preserving sensory attributes. Six formulations varying in skim milk powder (5-8%) and pectin (0-1%) were meticulously crafted and underwent sensory analysis alongside a control yogurt. The findings reveal that the inclusion of skim milk powder markedly influences syneresis reduction, showcasing a quadratic relationship. By employing response surface methodology and sensory evaluation, an optimal formulation comprising 8% skim milk powder and 0.06% stabilizer emerged, boasting superior sensory properties and mitigated syneresis. Furthermore, the study meticulously analyzed the composition of the optimized formulation, unveiling specific content percentages for total solid, pH, acidity, protein, fat, total ash, and lactose. Additionally, the research assessed the storage stability of the optimized product over a 10-day refrigerated period, tracking alterations in pH, acidity, and syneresis. Results indicated a gradual decline in pH coupled with an increase in acidity and syneresis, highlighting the importance of monitoring product attributes during storage. This investigation contributes valuable insights into maximizing whey utilization in yogurt production, ensuring both product quality and stability. The optimized formulation not only minimizes syneresis but also maintains sensory excellence, offering a promising avenue for the valorization of dairy by-products and enhancing sustainability within the dairy industry.

Keywords: Whey, Paneer, Yoghurt, Stabilizer, Skim milk powder, Syneresis, pH

INTRODUCTION

A major issue in the food industry is the considerable volume of surplus materials and secondary products generated throughout the production process (Comunian, Silva, & Souza, 2021). In dairy industry, the manufacturing of paneer results in the generation of a significant quantity of residual greenish water, commonly identified as paneer whey (Kumari & Rani, 2019). The whey comprises roughly 85-90% of the milk utilized, serving as the primary liquid waste and byproduct (Gupta & Prakash, 2017). The byproduct from the paneer industry is currently acknowledged as a valuable source of nutrients, labeled as a "treasure trove of nutrients." It contains approximately 45-50% of all milk solids, 70% of lactose, 20% of proteins, nearly all water-soluble vitamins, and 70-90% of the crucial minerals found in milk (Kumari & Rani, 2019). Studies indicate that whey demonstrates beneficial effects in managing various chronic conditions, including diarrhea, gallbladder complications, skin disorders, urinary tract problems, cancer, hypertension, and heart diseases (Ashoush, El-Batawy, & El-Shourbagy, 2013; Kerasioti et al., 2014).

Insufficient disposal systems in numerous milk factories result in the direct release of approximately 85% of total whey into the environment, posing a significant challenge for the dairy industry (Gantumur et al., 2024). Whey having the gretest biochemical oxygen demand among all dairy wastes, becomes the most harmful pollutant,promoting significant global environmental worries regarding its disposal (Kumari & Rani, 2019). Hence, it becomes imperative for both industry and the environment to explore innovative methods for economically utilizing waste whey (Gantumur et al., 2024). Utilizing whey and its preparations as substitutes

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may positively influence not just consumers' health but also the financial aspects of numerous companies. This substitution strategy has the potential to reduce raw material costs, consequently lowering overall production costs (Božanić, Barukčić, & Lisak, 2014). In developed nations, methods have been devised to valorize whey by converting it into value-added products like ricotta cheese, whey protein concentrates, isolates, and fermented beverages. However, in developing countries, a significant portion of whey is discharged untreated into water sources (Arshath, Alihath, & Chandran, 2023).

Fermented dairy items are pivotal in the global human diet, with yogurt standing out as a prime instance. Emerging from the symbiotic growth of Streptococcus thermophilus and Lactobacillus bulgaricus, yogurt yields a smooth, viscous gel with a desirable cultured flavor (Tribby, 2009). This fermentation process renders yogurt more easily digestible compared to milk (Olugbuyiro & Oseh, 2011). Yogurt is a nutritionally complete food, containing carbohydrates, protein, fat, vitamins, calcium, and phosphorus (Farahat & El-Batawy, 2013). Its consumption has been associated with various health advantages, including hindering the proliferation of cancer cells, protecting against osteoporosis, averting coronary heart disease, and easing digestive issues like constipation, diarrhea, and dysentery (Kamruzzaman, Islam, Rahman, Parvin, & Rahman, 2002).

The purpose of this study is to optimize the utilization of whey, in the making of yogurt. The aim is to identify the ideal combination of skim milk powder and stabilizer (pectin) to minimize water separation (syneresis) while maintaining the sensory qualities of the yogurt. This research highlights the potential for maximizing the use of whey in yogurt production, improving product quality, and contributing to sustainability efforts within the dairy industry.

MATERIALS AND METHODS

Collection of raw material

Fresh pasteurized milk (3% Fat and 8% SNF), paneer whey and yoghurt culture (50:50- *L. bulgaricus* and *S. thermophilus)* was used from Kamdhenu Dairy Development Corporation, Sunsari Nepal. Skim milk powder (Brand - DDC Nepal) used as the source of milk solid not fat (MSNF), sugar, pectin (Andre Pectin APA103, Grade 150) used as stabilizer, collected from the local market of Itahari.

Methodologies

Preliminary trial for whey-based yoghurt gel formation

An initial experiment was conducted to determine the optimal initial pH adjustment of acid whey for achieving optimal yogurt gel formation. Acid whey was prepared from standardized milk as shown in Figure 1 and used for preparation of yoghurt.

An initial experiment was conducted to determine the optimal pH adjustment of acid-whey for the production of high-quality yogurt gel. Acid-whey with a pH of 5.08 & 6% total solids was adjusted to different pH levels (5.14, 5.38, 5.63, 5.8, and 6) using sodium bicarbonate.

Each pH-adjusted acid whey sample was then mixed with 7% skim milk powder (SMP) and 1% sugar to achieve a total solid (TS) content of 14%. The mixture was heated at 75ºC / 15 seconds, followed by cooling to a temperature range of 43-45ºC.

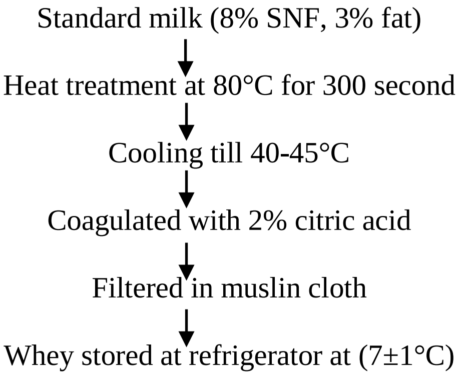

Figure 1. Preparation of whey (Bohora, 2018)

Following the addition of 1% starter culture, the mixture was incubated at 42±1ºC for fermentation. Gel formation was monitored hourly during incubation. It was observed that yogurt produced from whey with an initial pH of 5.08 adjusted to pH 6 exhibited superior gel formation compared to other pH levels, showing desirable consistency after just 4 hours of observation. Therefore, acid whey adjusted to an initial pH of 6 using sodium bicarbonate was selected for further studies.

Preparation of set type (control) yoghurt

Control yoghurt sample was prepared by using pasteurized milk (3% Fat and 8% SNF). The milk was mixed with 2% SMP and 1% sugar at 45°C, heated continuously to reach the temperature around 75°C for 15 seconds. Once cooled to 43-45°C, the starter culture was incorporated into each formulation at a 2% rate. Subsequently, the yogurt mixture underwent incubation at 43°C for a duration of 3.5-4 hours until the coagulum developed and after that it was cooled and stored. The flow diagram for preparation of set type of yoghurt is shown in Figure 2.

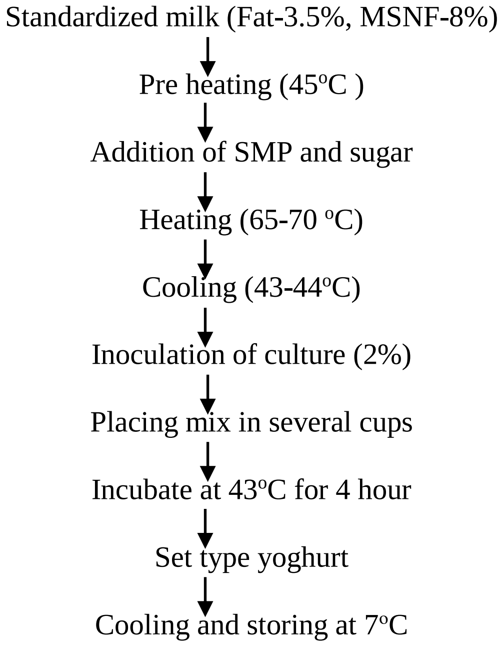

Figure 2. Preparation of set type of yoghurt Aswal, Shukla, and Priyadarshi (2012)

Formulation of yoghurt

Response surface methodology (RSM) by using Design Expert version 11 was used for optimization of level of SMP and stabilizer (pectin) addition for the preparation of whey yoghurt taking % syneresis as response variable. A three-level; two factor (central) composite Face Central Design was used for the experimental design. Two factors were varied as follows; % SMP from 5 to 8% & Stabilizer from 0 to 1% of acid whey (pH 6 adjusted by sodium bicarbonate). The treatment combinations obtained is presented in Table 1.

Table 1. Experimental combination of uncoded level for SMP and Stabilizer

Run	A: SMP (%)	B: stabilizer (%)
1	6.3	0.4
2	8.0	0.6
3	5.0	0.0
4	6.3	0.4
5	5.9	1.0
6	8.0	0.3
7	6.3	0.0
8	6.9	1.0
9	5.0	0.9
10	8.0	1.0
11	7.5	0.0
12	5.0	0.4
13	6.3	0.4
14	6.3	0.4

Preparation of whey-based yoghurt

Acid whey having pH of 4.8±0.8 was first adjusted to pH 6 by using sodium bicarbonate, pasteurized at 75°C for 15s before used for whey-based yoghurt production. The flow diagram for the production of yogurt based on whey is depicted in Figure 3.

Analytical methods

Syneresis: The syneresis analytical method, as outlined by (Lee & Lucey, 2004) involves placing a 100 g sample of yogurt on a filter paper atop a funnel. After 10 minutes of drainage under vacuum, the remaining yogurt is weighed. Syneresis, expressed as the proportion of free whey, is then calculated based on the difference in weight before and after drainage:

% free whey(g/ 100g)= weight of initial sample-weight of final sample ×100/ weight of initial sample

Fat: The Gerber method for fat analysis involves adding sulfuric acid and amyl alcohol to a milk/yoghurt sample, causing fat to separate as butyric esters. The fat content is determined by measuring the volume of separated fat, typically reported as a percentage (AOAC, 2005).

Lactose: Lane and Eynon's method detects glucose by its reaction with Fehling's solution, forming a red precipitate of cuprous oxide (Ranganna, 2004).

pH: The pH was measured at ambient temperature (27˚C) using a digital pH meter (Ecosense pH1000A). Before use, the pH meter underwent calibration using pH 4 and pH 10 buffer solutions. A volume of 50 ml of whey and yogurt was transferred to a beaker, and the pH meter's probe was inserted to record the pH value. Prior to each measurement, the probe was washed meticulously with distilled water.

Titratable acidity: Titration was employed to measure the titrable acidity of the samples. In this method, a 0.1N NaOH solution was used to titrate the samples. For the whey sample, 10 ml was carefully transferred to a conical flask after thorough mixing to prevent air incorporation, and an indicator was added. In contrast, for the yogurt sample, 10 ml was transferred to a flask after proper mixing, Afterwards, an equivalent volume of distilled water was added, and titration ensued with the introduction of an indicator (AOAC, 2005) (formula 1):

Titratable acidity% = (V×N×W)/(V^1×10)[where, V-volume of NaOH,V^1- volume of sample, N-Normality of NaOH, W-equivalent weight of reference acid(for lactic acid W=90)] *[1]*

Protein: Protein was determined Kjeldahl method. 2g sample was digested by adding 2g catalyst mixture and 25ml conc H_2SO_4. After digestion sample was diluted and distillation was carried out. Distilled volume was titrated and calculation was done by using data (AOAC, 2005).

Total ash: The ash content of the dried whey and yogurt samples was assessed at a temperature of 550°C following the AOAC (2005) protocol. Ash content is represented as the percentage of inorganic residue remaining after the incineration of whey and yogurt's total weight.

Total Solids: 10 gramm sample was weighed and after that dried under oven at 105°C for 3 hour and residue weight also noted. After that total solid of sample was calculated using given formula [2] (Olugbuyiro & Oseh, 2011).

Total solids (%) = Weight of residue / Weight of initial sample ×100% *[2]*

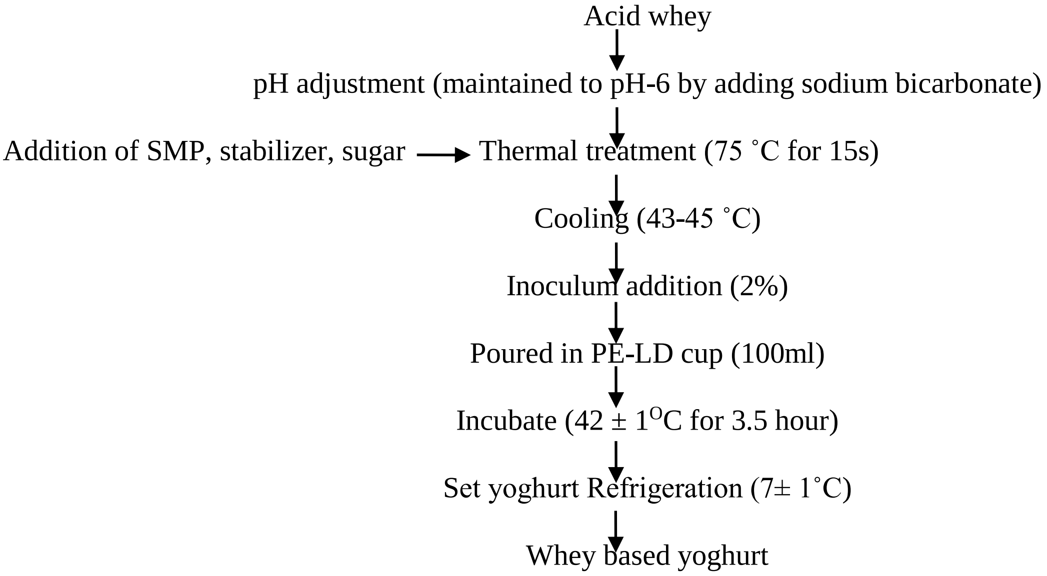

Figure 3. Flowchart for preparation of whey-based yoghurt (Skryplonek, 2018)

Sensory evaluation

Sensory evaluation of the samples were performed by 9 points hedonic scoring test (9 =like extremely, 1= dislike extremely) for appearance, taste, texture, color and overall acceptance. (Ranganna, 2004). The panelist members consisted of students and teachers of Nilgiri College.

Statistical analysis

Design Expert 11's Response Surface Methodology (RSM) was employed to optimize the levels of SMP and pectin. The acquired data were processed as mean values with standard deviations and subjected to Analysis of Variance (ANOVA) using IBM SPSS version 20 for analysis.

RESULT AND DISCUSSION

Physicochemical analysis of whey

The physicochemical composition of whey used in preparation of yoghurt was shown in Table 2. Chemical composition for the fresh whey was found to be 6.08% total solids, 5.18 pH, 0.21% lactic acid, 0.46% fat, 0.48% protein, 0.57% ash and 4.67% lactose respectively. This finding was similar with the results obtained by Raghavendra, Veena, Nath, and Amaladhas (2017) as 6.06% total solids, 5.43 pH, 0.24% lactic acid, 0.13% fat, 0.38% protein, 0.62% ash and 5.08 % lactose respectively. According to Koti and Gayathri (2022), our panner whey was classified as medium acid whey, as our obtained data were in the range 0.20 – 0.40% titratable acidity and 5.0-5.8 pH.

Table 2 Physicochemical composition of whey

Parameter	Panner Whey
TSS (°Bx)	5.50 (0.10)
Total solids (%)	6.08 (0.005)
pH	5.18 (0.01)
Titratable acidity % (as lactic acid)	0.21 (0.011)
Fat %	0.46 (0.005)
Protein %	0.48 (0.005)
Ash %	0.57 (0.006)
Lactose %	4.67 (0.011)

*Values are the mean of triplicates and values in brackets represent standard deviation.

Percentage syneresis of formulated yoghurt

Table 4.2 presents the percentage of syneresis for different formulations analyzed in a design experiment. The factors considered in the experiment include Build Type, Space Type, A:SMP (percentage of skimmed milk powder), B: stabilizer (percentage of stabilizer), and the resulting syneresis percentage.

The table consists of 14 runs, each representing a specific formulation. The Build Type refers to whether the formulation was replicated or part of the model, while the Space Type specifies where the formulation was located (interior, edge, or vertex). A:SMP and B: stabilizer indicate the percentages of skimmed milk powder and stabilizer used in the formulation, respectively. The last column shows the calculated percentage of syneresis for each formulation.

Different methods can be employed to minimize syneresis in milk-based products, including enriching them with various additives like protein-based elements such as skimmed milk powder (SMP), whey protein powders (WP), casein powders (CP), and appropriate stabilizers (Arab et al., 2023) . Similarly in our study we see syneresis percentages fluctuating at different levels of SMP and stabilizer. For instance, at 5% SMP, the syneresis percentage is 42.24%, while at 8% SMP, it is 19.74%.

Table 3. Percentage syneresis of different formulation

			Factor 1	Factor 2	Response
Run	**Build Type**	**Space Type**	**A:SMP, %**	**B:stabilizer, %**	**Syneresis, %**
1	Replicate	Interior	6.3	0.4	53.28
2	Model	Edge	8	0.6	24.67
3	Model	Vertex	5	0	42.24
4	Model	Interior	6.3	0.4	46.88
5	Lack of Fit	Edge	5.9	1	45.03
6	Lack of Fit	Edge	8	0.3	21.28
7	Lack of Fit	Edge	6.3	0	46
8	Model	Edge	6.9	1	35.02
9	Model	Edge	5	0.9	38.39
10	Lack of Fit	Edge	8	1	19.74
11	Model	Edge	7.5	0	36.01
12	Lack of Fit	Edge	5	0.4	43.53
13	Replicate	Interior	6.3	0.4	46.03
14	Replicate	Interior	6.3	0.4	45.22

This comparative analysis can provide insights into the effectiveness of different formulation strategies in reducing syneresis and contribute to the understanding of the underlying mechanisms involved.

Effect of addition of SMP and pectin on syneresis of yoghurt

The content of SMP in yogurt ranged from 5% to 8%, while pectin ranged from 0% to 1%. Tables 4 and 5 display the model coefficients and other statistical parameters for syneresis. The regression model applied to experimental syneresis results revealed a significant model F-value of 28.39 (p<0.0001). Our results accord with the findings of (Akalın, Unal, Dinkci, & Hayaloglu, 2012) , who reported that casein-based samples showed firmer gels with less syneresis than yoghurts enriched with cheese whey. The probability of such a high model F-value occurring due to random variation was only 0.4909%. The model's goodness of fit was also indicated by the coefficient of determination (R2), which was determined to be 0.9466, suggesting that 94.66% of the response variability could be explained by the model. Additionally, the adjusted R2 value of 0.91333 and the predicted R2 value of 0.8556 demonstrated an adequate model fit (see Table 5).

A ratio >4 is desirable and hence this model can be used to investigate the effect of SMP and pectin variation on % syneresis of prepared whey-based yoghurt sample (Myers, Montgomery, & Anderson-Cook, 2016). The equation expressed in terms of coded factors facilitates predictions of the response for specified levels of each factor. Typically, the high levels of the factors are coded as +1, while the low levels are coded as -1. This coded equation proves beneficial in discerning the relative influence of the factors through comparison of the factor coefficients.Which is given in equation [3]:

$$Syneresis = +46.22 - 9.31A - 2.43B - 0.0946AB - 13.09A^2 - 2.38B^2 \qquad [3]$$

where, A and B are the coded values of SMP (%) and stabilizer (%) respectively.

Percentage syneresis in yoghurt had highly significant positive quadratic effect of SMP (A) (p < 0.0001), Puvanenthiran et al. (2002) observed that increasing the ratio of whey protein to casein protein in yogurt formulation resulted in a looser microstructure of the yogurt. This looser structure allowed for more free water to be present in the yogurt. Similar results on the decrease of whey separation by using SMP were also obtained by (Guzmán-González, Morais, Ramos, & Amigo, 1999) (Bhullar, Uddin, & Shah, 2002) and (Remeuf, Mohammed, Sodini, & Tissier, 2003) . whereas percentage stabilizer follows quadratic model but don't have significant effect on % syneresis . Similar result was shown in (Sobhay, Hassan, & El-Batawy, 2019) study, where physochemical properties of drinking yoghurt fortified with different types and ratios of stabilizers during storage showed that the types and concentrations of different stabilizers added to different drinking yoghurt treatments had no significant effect on physochemical properties in all final products. This could be due to the very low quantity of different stabilizers (0.2 and 0.4%) added to various drinking yoghurt treatments and these low quantity had no effect on total chemical composition of final product. The obtained results agree with (Hematyar, Samarin, Poorazarang, & Elhamirad, 2012)who found that, using xanthan and carrageenan at different concentrations had no effect on physochemical composition and pH value of yoghurt. In addition, interaction of both SMP and stabilizer (AB) was found to be had no significant effect on % syneresis. Response surface plot for % SMP and stabilizer as a function of % syneresis which is given in Figure 4.

Table 4. ANOVA for Quadratic model for syneresis

Source	Sum of Squares	Df	Mean Square	F-value	p-value	
Model	1311.52	5	262.3	28.39	< 0.0001	Significant
A-SMP	581.15	1	581.15	62.9	< 0.0001	
B-stabilizer	39.52	1	39.52	4.28	0.0724	
AB	0.029	1	0.029	0.0031	0.9567	
A^2	482.25	1	482.25	52.19	< 0.0001	
B^2	16.06	1	16.06	1.74	0.2239	
Residual	73.92	8	9.24			
Lack of Fit	33.26	5	6.65	0.4909	0.7726	not significant

Table 5. Fit statistics

Std dev.	Mean	C.V%	R2	Adjusted R^2	Predected R^2
3.04	38.81	7.83	0.9466	0.91333	0.8556

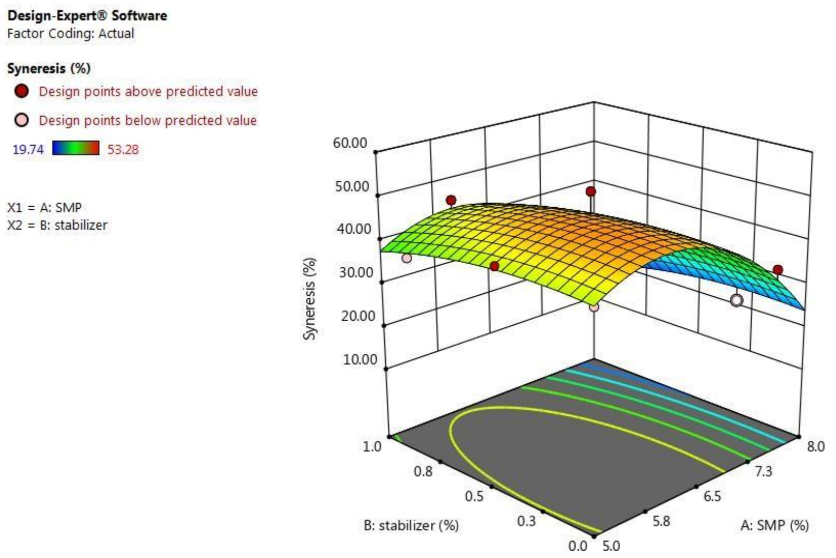

Figure 4. Response surface plot for % SMP and stabilizer as a function of % syneresis

Optimum level of SMP and pectin addition for minimum syneresis of yoghurt

By using equation, optimization of SMP and pectin for whey based yoghurt was carried outkeeping the assumption; SMP in range (5-8%), Pectin in range (0-1%) and % syneresis minimum. The model output provided altogether 26 solutions for % SMP and % Pectin level addition for whey based yoghurt preparation with minimum syneresis (table 6).

Table 6 Twenty-six solution for optimization of SMP and stabilizer for best syneresis

Number	SMP	stabilizer	Fermentation Time (second) to reach pH 4.3	Std Err (Fermentation Time to reach pH 4.3)	Syneresis	StdErr (Syneresis)	Desirability
1	7.986	0.991	188.392	0.859	19.365	2.377	1.000
2	7.999	0.972	188.113	0.837	19.335	2.322	1.000
3	8.000	0.995	188.199	0.893	18.979	2.413	1.000
4	7.993	0.981	188.236	0.840	19.348	2.346	1.000
5	7.984	0.998	188.470	0.885	19.323	2.403	1.000
6	7.995	0.963	188.137	0.828	19.545	2.284	1.000
7	7.995	1.000	188.307	0.907	19.030	2.426	1.000
8	7.980	0.982	188.435	0.823	19.638	2.332	1.000
9	7.997	0.967	188.128	0.831	19.456	2.300	1.000
10	7.985	0.972	188.318	0.815	19.656	2.302	1.000
11	7.984	0.976	188.350	0.818	19.628	2.315	1.000
12	7.971	0.999	188.674	0.878	19.610	2.391	1.000
13	7.973	0.993	188.592	0.851	19.639	2.367	1.000
14	7.993	0.985	188.263	0.849	19.302	2.362	1.000
15	7.993	0.957	188.154	0.826	19.672	2.261	1.000
16	7.999	0.990	188.191	0.872	19.090	2.389	1.000
17	7.997	0.952	188.085	0.838	19.659	2.248	1.000
18	7.998	0.948	188.071	0.844	19.698	2.236	1.000
19	8.000	0.883	188.085	0.996	20.486	2.068	0.977
20	8.000	0.556	190.263	0.751	23.502	1.899	0.883
21	8.000	0.487	190.457	0.740	23.878	1.917	0.872
22	8.000	0.000	176.411	3.002	23.951	3.112	0.869
23	8.000	0.004	176.680	2.926	23.968	3.089	0.869
24	8.000	0.060	180.302	1.970	24.187	2.772	0.862
25	5.000	1.000	194.524	2.793	37.725	3.106	0.442
26	5.000	0.000	185.231	0.883	42.391	2.613	0.298

Among them 18 solutions had desirability of 1 and only six solution having slightly different combination level of SMP and pectin were chosen for post analysis (% Variability and model validation) of model output and for further sensory evaluation.

The combinationlevels of %SMP and pectin taken for evaluation of model output and sensory evaluation are presented in the Table 7. Incorporating pectin as an ingredient in yogurt often yields a pleasing pudding-like consistency (Tribby, 2009). In order to evaluate model performance, all the six levels of SMP and pectin (Table 7) were used for preparation of whey-based yoghurt, analyzed for the % syneresis and compared with the model prediction. The percentage variation between model output and real response found to be ranged from 0.62 to 5.74% showing almost 94% of model validity.

Effect of optimized formulations on sensory characteristics

The six whey yoghurt samples were made by varying the SMP and pectin formulation. The graph of mean scores and significant differences in terms of appearance, color, texture, taste and overall acceptance is shown in Figure 5. The similar alphabet above the bar graph indicates that there is no significant difference, and the error bars show the standard deviation of scores given by 15 panelists.

Table 7 Comparison of % syneresis of selected formulation for model validation

Code	% SMP	% stabilizer (pectin)	% syneresis from lab test (actual)	% syneresis from model predication	% variation
A	7.980	0.991	19.480	19.360	0.620%
B	7.997	0.952	20.670	19.560	5.670%
C	8.000	0.883	21.380	20.480	4.390%
D	8.000	0.556	24.260	23.520	3.140%
E	8.000	0.000	23.800	23.960	0.660%
F	8.000	0.060	22.800	24.187	5.740%

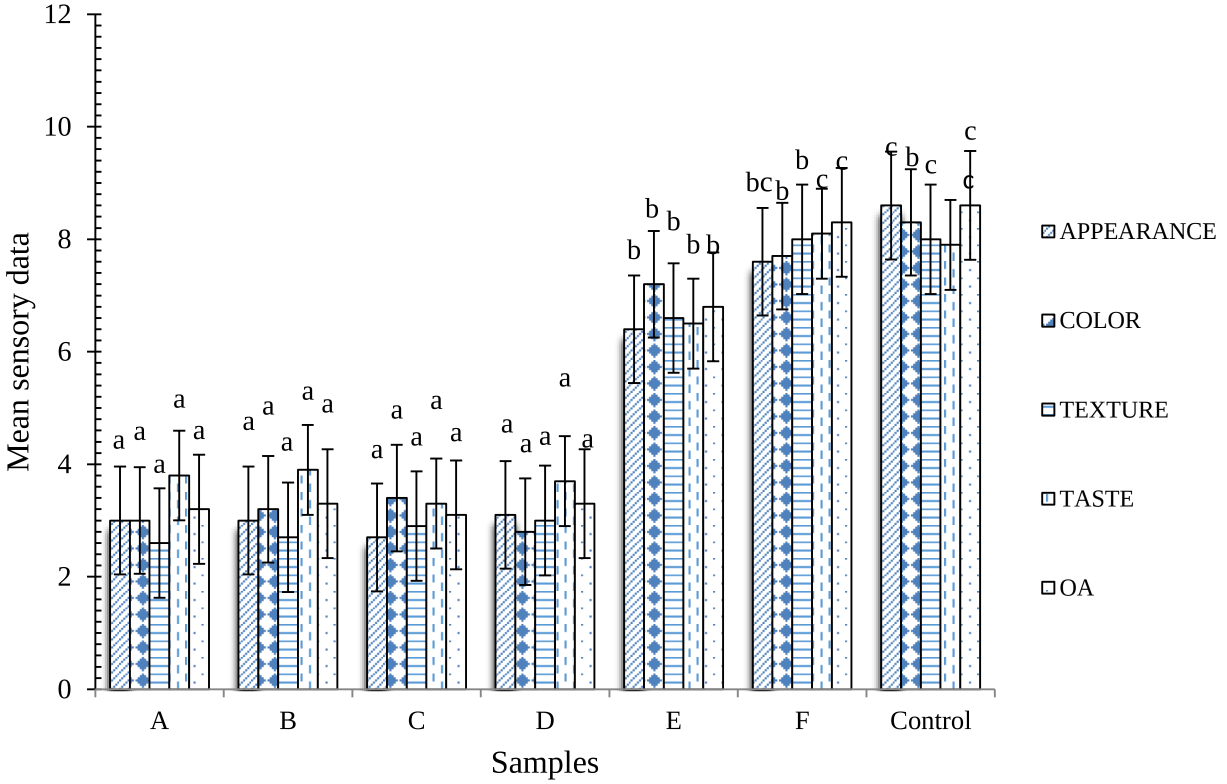

Figure 5. Mean sensory score of prepared whey yoghurt. Error bars represent standard deviation, and those with distinct superscripts differ significantly (P<0.05).
A(SMP:Stabilizer-7.980:0.991), B(SMP:Stabilizer-7.997:0.952), C(SMP:Stabilizer-8.000:0.883), D(SMP:Stabilizer-8.000: 0.556), E(SMP:Stabilizer-8.000: 0.000), F(SMP:Stabilizer-8.000: 0.060).

Appearance

The mean sensory score for the appearance of the yoghurt of seven samples A, B, C, D, E, F and control were determined to be 3, 3, 2.7, 3.1, 6.4, 7.6 and 8.6 respectively. The obtained mean values are represented as bar diagram in Figure 5. Statistical analysis shows that products A, B, C, D, E were significantly different from control sample where sample F was not significantly different. Sample F contain less % of stabilizer and high SMP, it showed that pectin has negative effect on appearance. According to Bhattarai, Pradhananga, and Mishra (2015) An excessive amount of stabilizer can negatively affect the taste of a natural yogurt gel. Therefore, a moderate concentration of stabilizer might be suitable to maintain its appealing appearance. This is the similar results as obtained in our study.The addition of SMP had significant linear effect on increasing average score of appearance of whey-based yoghurt. While, the addition of sugar have significant linear effect on decreasing average score of appearance of whey based yoghurt (Adhikari, 2022).

Color

From the sensory evaluation for the color of the seven distinct samples A, B, C, D, E, F and control the mean sensory score were found to be 3, 3.2, 3.4, 2.8, 7.2, 7.7 and 8.3 respectively. The obtained mean values are represented as bar diagram in Figure 5. The statistical analysis showed that products A, B, C, D was found to be significantly distinct from that of the control sample. but sample E & F was found to not be significantly different from that of the control sample. Bhattarai et al. (2015) concluded that yogurt containing less stabilizer was perceived as the best option by panelists, based on the frequency of occurrence of "best" in each attribute. This suggests a preference among panelists for yogurt with lower stabilizer content over yogurt with higher stabilizer content. This is the similar results as obtained in our study. There was slightly significant difference in color of yoghurt with different stabilizer amount but as the higher stabilizer amount the color was less preferred by panelist.

Texture

During the sensory evaluation, the texture of seven different samples (labeled as A, B, C, D, E, F) and a control sample, the mean sensory score were found to be 2.6, 2.7, 2.9, 3, 6.6, 8 and respectively. The obtained mean values are represented as bar diagram in Figure 5. The statistical analysis showed that products A, B, C, D were significantly different from control while the sample F was significantly not different with control at 5% level of significance. Bhattarai et al. (2015) found that an excessive concentration of stabilizer can diminish the palatability of a natural yogurt gel. Therefore, they suggested that a moderate concentration of stabilizer might be more suitable to ensure good textural quality. These findings align with the results obtained in our study. There was significant difference in texture of yoghurt with different stabilizer amount, as the higher stabilizer amount the texture was less preferred by panelist.

Taste

The mean sensory score for the taste of the yoghurt of seven samples A, B, C, D, E, F and control were determined to be 3.8, 3.9, 3.3, 3.7, 6.5, 8.1 and 7.9 respectively. The obtained mean values are represented as bar diagram in Figure 5. The statistical analysis showed that products A, B, C, D were significantly different from control. Sample F were not significantly different from control sample. There was significant difference in taste of yoghurt with different stabilizer amount, as the higher stabilizer amount the taste was less preferred by panelist.

Overall acceptability

The mean sensory score for the OA of the yoghurt of seven samples A, B, C, D, E, F and control was found to be 3.8, 3.9, 3.3, 3.7, 6.5, 8.1 and 8.6 respectively. The obtained mean values are represented as bar diagram in Figure 5. The statistical analysis indicated that products A, B, C, D, and E were significantly different from the control sample, whereas sample F did not exhibit significant differences at the 5% level of significance. Moreover, there was no significant distinction between the control sample and sample F. Overall, there appears to be similarity between the control sample and sample F in terms of appearance, color, texture, taste, and overall acceptability. The finding from Bhattarai et al. (2015), suggest that yogurt containing less stabilizer was preferred by panelists, as indicated by the frequency of occurrence of "best" in each attribute. Similarly, our study yielded similar results, further supporting the notion that yogurt with lower stabilizer content tends to be preferred over yogurt with higher stabilizer content by sensory panelists. Appearance and taste are significant factors influencing consumer acceptability. In the present study, whey-based yogurts containing 8% SMP and 0.06% stabilizer indicates the most preference by panelist in terms of sensory value. So, from whey-based yoghurt sample F was found to be best and no significantly difference with control sample

Physicochemical composition of optimized whey yoghurt with milk yoghurt

The proximate composition of whey-based yoghurt was found to be different from those of the control sample. The physicochemical composition of yoghurt obtained were presented in Table 8. The values presented are the means calculated from triplicate measurements and values in parenthesis represent standard deviation. Similar alphabets indicate no significance difference where different alphabets indicate significant difference.

The total solids content of sample F was slightly lower than that of the control sample, with values of 14.26% and 14.79% respectively. A significant difference ($p < 0.05$) was observed between them. Total solid of control sample found in our study was similar to that reported by Hofi, El-Dien, and El-Shibiny (1978). The TS and other parameter increase during fermentation. This may be due to thepresence of microbial cells and their metabolic activities in fermented products having highprotein and fat. The other constituents in yoghurt which included in % total solids are-fat associated substance (like lecithin, cholesterol, carotene, vit. A, D, E, K), others constituents like enzymes, vit. B, vit. C and dissolved gas etc. Non reducing sugar (sucrose) also take parts in total solid. Microbial cells also take parts in %TS (Acharya, 2011).

The pH of sample F was slightly lower than that of the control sample, with values of 4.4 and 4.5 respectively. A significant difference ($p < 0.05$) was observed between them. The pH of yoghurt from whole milk was similar to that reported by (Khadka, 2018). The decrease in lactose content and pH, along with the increase in acidity, observed during fermentation can be attributed to the conversion of lactose into lactic acid, as indicated by (Zourari, Accolas, & Desmazeaud, 1992). When fermentation is halted at a pH level above 4.7, yogurt may often display a weak body and/or stringy texture. Therefore, utilizing a pH meter is crucial for identifying the break point during the fermentation process, as highlighted by (Tribby, 2009).

The acidity of sample F was slightly higher than that of the control sample, with values of 0.82% as lactic acid and 0.77% respectively. A significant difference ($p < 0.05$) was observed between them. This increase in acidity could be attributed to the fermentation of lactose into lactic acid, resulting in a decrease in lactose content and pH, as commonly observed during fermentation processes. (Zourari et al., 1992).

The protein of sample F was found to be slightly lower than control sample i.e., 2.8 and 4.18 respectively. This is because raw material of whey yoghurt has low protein content reported by Darade and Ghodake (2012) compared to raw material of whole milk yoghurt (Tamime & Robinson, 2007). Protein content of control sample found in our study was similar to that reported by (Mohammad & El-Zubeir, 2011).

Fat content of control sample found in our study was similar to that reported by Hofi et al. (1978). The fat content increases during fermentation. This may be due to the presence of microbial cells in fermented products they are rich in protein and fat. The fat of sample F was found to be significantly lower than control sample i.e., 0.61 and 3.61. Whey based yoghurt has low fat and could be benificial heart patient as compared to milk yoghrt (Darade & Ghodake, 2012).

The ash content of sample F was found to be significantly higher than control sample i.e., 1.3% and 0.75% respectively. The ash content of yoghurt from whole milk was similar to that reported by (Khadka, 2018). The high amount of ash shows high minerals content in whey-based yoghurt. As compared to normal yoghurt which could be important from nutritional point of view.

The lactose content of sample F was slightly higher than that of the control sample, with values of 5.061% and 3.85% respectively. However, there was no significant difference ($p > 0.05$) between them. This increase in lactose content during fermentation aligns with the process of lactose conversion into lactic acid, as

(Zourari *et al.*, 1992). Furthermore, the lactose content of yogurt from whole milk was found to be similar to that reported by (Bhagiel, Mustafa, Tabidi, & Ahmed, 2015).

Table 8. Physicochemical composition of whey based yoghurt and milk yoghurt (control)

Parameter	**Best product (Sample F)**	**Control**
TS (%)	$14.26^{a} \pm 0.02$	$14.79^{b} \pm 0.76$
pH	$4.4^{a} \pm 0.016$	$4.50^{b} \pm 0.015$
Acidity (% as lactic acid)	$0.82^{b} \pm 0.023$	$0.77^{a} \pm 0.045$
Protein (%)	$2.82^{a} \pm 0.16$	$4.18^{a} \pm 0.59$
Fat (%)	$0.61^{a} \pm 0.064$	$3.61^{b} \pm 0.18$
Ash (%)	$1.3^{b} \pm 0.056$	$0.75^{a} \pm 0.047$
Lactose (%)	$5.06^{b} \pm 0.013$	$3.85^{a} \pm 0.09$

Storage stability of whey based yoghurt

The whey based yoghurt was packed in low density polyethylene cup covered with aluminium foil. The product was kept at refrigerated temperature for 10 days (day 0, 2, 4, 6, 8, 10). The storage stability of whey yoghurt was studied in terms of pH, acidity and syneresis.

pH of sample F was found to be decreased at refrigerated temperature from 4.42 to 3.40 and that of control sample at refrigerated temperature was found to decrease from 4.52 to 3.49 during the storage period of 10 days. In case of whey based yoghurt the rate of decreasing pH was low than that of control sample. The graph for pH is shown in Figure 6. Salji and Ismail (1983) reported the higher decreasing rate in pH as increase in storage time. This was similar to the result obtained in our study. The decrease in pH observed during fermentation may be attributed to the formation of lactic acid through the fermentation of lactose present in milk, as indicated by (Zourari et al., 1992). However, it's noteworthy that the rate of pH decrease was almost similar to that of the control sample. This similarity could potentially be explained by the composition of acid whey, as suggested by (Darade & Ghodake, 2012).

The acidity of whey based yoghurt was found to be increased at refrigerated temperature from 0.81-1.33% as lactic acid and that of control sample was found to increase from 0.76- 1.05% as lactic acid throughout the day 10 of the storage period. The increasing rate of acidity was low for control sample than that of sample F. The graph for acidity is shown in Figure 7.

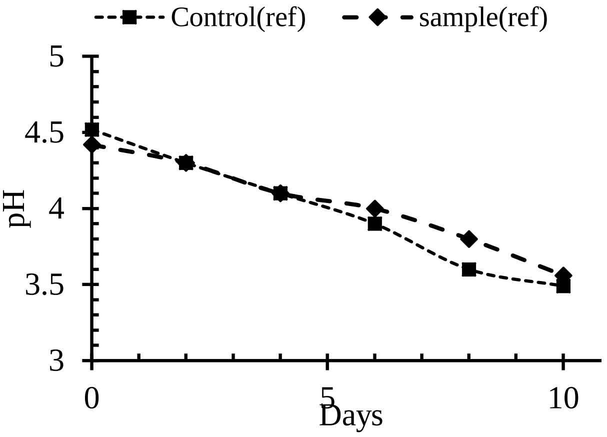

Figure 6. pH changes during the storage period of yoghurt.

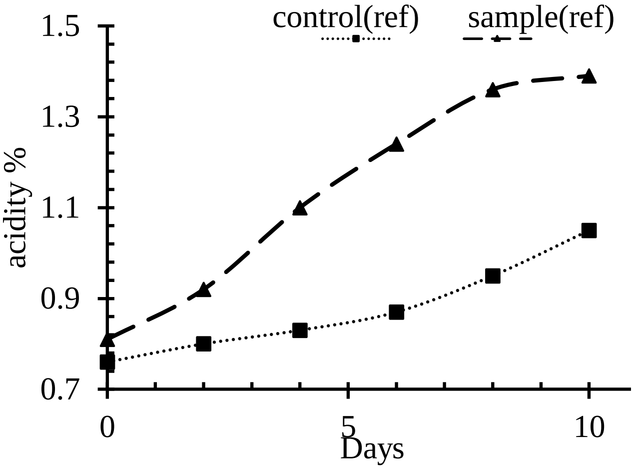

Figure 7. Acidity changes during the storage period of yoghurt

Salji and Ismail (1983) reported the higher increasing rate in acidity as increase in temperature. This was similar to the result obtained in our study The rise in acidity could stem from lactic acid formation via lactose fermentation (Zourari *et al.*, 1992). However, the rate of increase surpassed that of the control, possibly due to acid whey composition (Darade & Ghodake, 2012).

The syneresis of whey yoghurt at refrigerated temperature was found to be increased from 22.8-32.67% and that of control sample at refrigerated temperature was found to increase from 18.62-29.99 % during 10 days of storage period. The increasing rate of syneresis was higher for sample F than that of control sample. The graph for syneresis is shown in Figure 8.

Bhattarai et al. (2015) demonstrated a correlation between pH and syneresis, indicating that as storage time increased, pH decreased while syneresis increased. This trend was attributed to lactic acid formation over time. Interestingly, whey-based yogurt consistently exhibited significantly lower pH values each day compared to milk-based yogurt, while syneresis was significantly higher. The addition of stabilizers appeared to slow down both acid development and syneresis in yogurt.

On overall, the total changes in pH between milk yoghurt and whey based yoghurt during 10 days of refrigerated storage is almost similar and with respect to total increase in syneresis during 10 days refrigerated storage, whey based yoghurt showed better result as compared to milk yoghurt. But regarding total increase in acidity whey based yoghurt comparatively very high increment (0.59%) as compared to increment in acidity milk yoghurt (0.29 %) during 10 days of storage (Table 9). This high change in acidity might be due to initial adjustment of whey pH in preparation of whey based yoghurt.

Cost evaluation

From the cost calculation given in Table 10, the cost of yogurt per 100 g was calculated at NRs. 5.16, excluding labor, packaging, and tax. Mass production is anticipated to lower this cost further. Effective utilization of by-products has already reduced the yogurt's cost, with further potential reduction by decreasing the proportion of SMP.

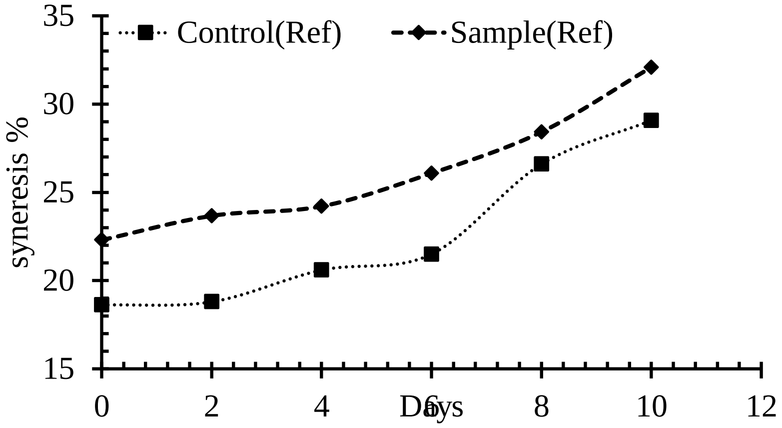

Figure 8. Syneresis changes during the storage period of yoghurt
* Vertical error bars represent the standard deviation.

Table 9. Total changes in parameter on 10 days storage period

Products	Total changes in parameter (at 10 days refrigerated storage)		
	Decrease in pH	Increase in % Syneresis	Increase in % Acidity
Whey based yoghurt	1.02	9.87	0.52
Milk yoghurt	1.03	11.37	0.29

Table 10: Cost calculation of the product

Ingridents	Quantity	Amount (Rs)	Quantity used	Amount(Rs)
Whey	1000 g	0	90 g	0
SMP	1000 g	500	8 g	4
Sugar	1000g	85	2g	0.16
Stabiizer & acidulents	100	20	0.06	1

CONCLUSION AND PERSPECTIVES

The aim of this study was to explore the optimization of whey utilization in yogurt production, focusing on minimizing water separation (syneresis) while preserving sensory attributes. Through careful formulation and analysis, several key findings have been elucidated, filling important gaps in understanding and application:

- Sensory analysis revealed that yogurt formulated with 8% skim milk powder and 0.06% stabilizer emerged as the superior product among the six prepared samples. This optimal formulation showcases the potential for maximizing whey utilization while maintaining product quality.
- Physicochemical analysis unveiled specific content percentages for total solid, fat, acidity, protein, total ash, lactose, and pH in the yogurt. These findings provide valuable insights into the composition and characteristics of whey-based yogurt.
- Investigation into total plate count (TPC) demonstrated that whey yogurt exhibited lower microbial growth at refrigerated conditions compared to room temperature. While overall TPC was higher in whey yogurt than in the control sample, refrigeration proved effective in extending the shelf life of the product.

Overall, this study lays the groundwork for continued research and innovation in whey utilization and yogurt production, contributing to advancements in dairy industry sustainability and product development.

Perspectives for Future Research

Moving forward, there are several avenues for future research in this domain:

- Exploration of flavored yogurt variants by incorporating different types of flavorings to

enhance consumer appeal and diversity of product offerings.
- Investigation into alternative stabilizers to further improve the quality and stability of yogurt formulations, expanding the range of options available for formulation optimization.
- Exploration of sugar addition to adjust sweetness levels in yogurt, catering to diverse consumer preferences and market demands.

CONFLICTS OF INTERESTS

None

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