Studii şi Cercetări Ştiinţifice
Chimie şi Inginerie Chimică, Biotehnologii, Industrie Alimentară

2024, **25** (4), pp. 353 – 363

https://doi.org/10.29081/ChIBA.2024.603

Scientific Study & Research
Chemistry & Chemical Engineering, Biotechnology, Food Industry

ISSN 1582-540X

ORIGINAL RESEARCH PAPER

OPTIMIZATION OF TOTAL SOLUBLE PROTEIN OF YOGHURT WITH ADDITION OF PSYLLIUM HUSK POWDER AND FULL CREAM MILK POWDER

Safri Ishmayana[1*], Muhammad Fadhlillah[1], Fajriana S. Nurrusyda[2], Agus Safari[1], Riza Apriani[3], Puspitasari R. Devi[1], Debora T. Horasio[1], Ukun M.S. Soedjanaatmadja[1]

[1]Universitas Padjadjaran, Faculty of Mathematics and Natural Sciences, Department of Chemistry, Jl. Raya Bandung-Sumedang km. 21 Jatinangor, Sumedang, 45363, West Java, Indonesia
[2]Universitas Padjadjaran, Industrial Chemistry Technology Study Program, Vocational School, Jln. Raya Bandung-Sumedang km. 21 Jatinangor, Sumedang, 45363, West Java, Indonesia
[3]Universitas Garut, Faculty of Mathematics and Natural Sciences, Department of Chemistry, Jl. Jati No.42, Tarogong Kaler, Garut, 44151, West Java, Indonesia

*Corresponding author: ishmayana@unpad.ac.id

Received: September, 09, 2024
Accepted: November, 25, 2024

Abstract: Yoghurt is a fermented dairy product with many beneficial properties. During fermentation process using lactic acid bacteria, the physicochemical properties of yoghurt change including the soluble protein (SP) content due to the formation of insoluble protein as a result of decreasing *p*H. The present study was directed to determine the best conditions to obtain the highest SP content, even though some of proteins become insoluble in yoghurt. The optimal conditions were determined by response surface method-central composite design (RSM-CCD). Yoghurt was prepared by fermenting pasteurized milk with *Lactobacillus bulgaricus* with varying concentrations of psyllium husk powder (PHP) and full cream milk powder (FCMP). The SP content was determined using Lowry method. The results of the present study indicate that the highest SP content can be achieved when 0.162 % w/v PHP and 5.621 % w/v FCMP are used in the yoghurt preparation process. The SP concentration at these conditions can reach up to 2.704 ± 0.102 mg·mL^{-1}. The organoleptic tests of the yoghurt indicate that it has better acceptance by semi-trained panelists compared to yoghurt prepared without the addition of PHP and FCMP.

Keywords: *dairy product, full cream milk powder, protein, psyllium husk, yoghurt*

INTRODUCTION

Yoghurt is recognized globally not only for its rich nutritional value, but also for its enhanced taste and health-promoting properties [1, 2], which are related to the beneficial components produced during the fermentation process [3]. During this process, casein, the main protein component in milk, becomes unstable and coagulates due to milk acidification [4]. Protein solubility is an important factor in digestibility, as more soluble proteins are easier to digest, especially during gastric digestion [5], which may lead to better bioavailability. Therefore, increasing the amount of soluble protein in yoghurt is crucial for enhancing protein bioavailability. The addition of basil seed gum as a stabilizer has been shown to increase whey protein isolate solubility [6]; thus, we propose that adding psyllium (*Plantago ovata*) husk powder (PHP), which contains natural gum, may also impact the soluble protein content in yoghurt.

Following fermentation, yoghurt thickens and is susceptible to the separation of solid and liquid components. Yoghurt might experience consistency concerns due to syneresis during storage. Syneresis, the release of whey into the liquid fraction, may increase the concentration of soluble protein in the released liquid [7]. During the process of syneresis, the hydrogen bonds between water and proteins weaken, reducing the interactions between casein molecules. This results in the formation of larger pores within the gel structure, allowing water to escape more freely [8]. Syneresis negatively impacts yoghurt quality, manifesting as undesirable textural changes [7]. These alterations in texture lead to inconsistencies in mouthfeel, compromising the sensory experience. The addition of stabilizers, such as psyllium husk, plays a critical role in reducing syneresis by improving water retention and enhancing the gel matrix, thereby improving textural properties and overall product quality [9, 10].

Adding stabilizers can prevent textural change. They improve the thickness of the mixture, stabilize proteins to avoid separation, keep flavor particles suspended, form a consistent matrix that is easier to cut and maintain in the freezer for packaging, minimize moisture movement, and prevent the product from shrinking during storage. When exposed to water, several of these chemicals can also interact with proteins and lipids in the mixture [7]. Bhat *et al*. [10] reported that adding psyllium husk to yoghurt at a concentration of 0.7 % can increase the water content of yoghurt to 82.08 %. This indicates that higher amounts of psyllium husk can absorb more water to reduce syneresis. Another finding by Fu *et al*. [11], claimed that adding psyllium husk up to 0.6 % to ice cream will nearly eliminate the separation of liquid that happens at the bottom of ice cream, even after being stored in the refrigerator for 7 days.

Moreover, the sensory attributes of dairy products are largely determined by the distribution of taste components derived from carbohydrates, proteins, and fats in the milk [12]. The taste of yoghurt significantly influences consumers' perception of these sensory qualities. The amount of fat in yoghurt determines how creamy it is. Enhancing yoghurt with full-cream milk can improve its taste and appeal. According to Vasilean *et al*. [13], the fat content has a significantly positive impact on the sensory properties of products. The color of the samples with higher fat contents (3.5 % and 1.5 %) was better appreciated by the assessors probably because of their higher luminosity. Therefore, the present study aims to investigate the use of PHP as a stabilizer to enhance the soluble protein content of yoghurt, thereby increasing protein bioavailability. Additionally, PHP is expected to improve consumer acceptance by enhancing the texture and reducing syneresis [9] other than improving soluble fiber content in the yoghurt product [10].

MATERIALS AND METHODS

Materials

Fresh milk was purchased from a local farmer, while full cream milk powder (Dancow, Nestle Indonesia) and skim milk powder (NZMP Skim Milk Powder, PT Culina Gumilang Indonesia) were purchased from a local store. The nutritional value of the materials is presented in Table 1. Soluble protein, total protein, lactose, fat and water content of fresh milk were determined using Kjeldahl [14], Lowry [15], alkaline ferricyanide [16], Soxhlet [14], and gravimetric [14] methods, respectively. While nutrition value of full cream milk powder and skim milk powder were reported based on nutrition fact provided by the manufacturer. Psyllium husk was purchased from a local store, ground until fine and sifted through an 80-mesh sieve. The sifted psyllium husk was then collected and used in the following experiment.

Table 1. *Nutritional value of fresh milk, full cream milk powder, and skim milk used in the present study*

Parameter	Fresh Milk	Full cream milk powder	Skim milk powder
Soluble protein content [%]	0.12	n.a.	n.a.
Total protein [%]	3.7	24	32.9
Lactose content [%]	0.09	36	54.5
Fat content [%]	2.57	28	0.9
Water content [%]	87.8	n.a.	3.8

Note: n.a. = not available

Lactobacilus bulgaricus was isolated from commercial yoghurt and maintained on an agar slant. Bovine serum albumin (BSA), Folin-Ciocalteu solution, sodium carbonate, sodium hydroxide, copper sulfate pentahydrate, and sodium tartrate were purchased from Sigma Aldrich and are of analytical grade.

Preparation of Starter Culture

10 % (w/v) skim milk powder was mixed with hot distilled water and transferred to an Erlenmeyer flask, then pasteurized at 75 °C for 30 minutes. The *L. bulgaricus* culture, isolated by Safari *et al.* [17], was introduced into the milk solution after cooling and then incubated for 24 hours at 37 °C.

Yoghurt Fermentation

Yoghurt fermentation was carried out based on the method proposed by Abdelazez *et al.* [18] with some modification. Briefly, a total of 250 mL of milk was pasteurized at 75 °C for 30 minutes. When the temperature reached 50 °C, FCMP and PHP were added as described in Table 2. The concentrations of FCMP and PHP were determined using response surface method central composite design (RSM-CCD), a statistical approach commonly employed to optimize processes by evaluating the effects of multiple variables and their interactions. This method was used to determine the optimal conditions for maximizing soluble protein content. The pasteurized milk was cooled until the temperature reached 37 °C. Then 5 % (v/v) of *L. bulgaricus* starter was added and incubated at 37 °C for 20 hours. Samples were stored in a refrigerator for about 24 hours until assays were performed, and each sample was measured twice.

Table 2. *Experimental design for Response Surface Method (RSM-CCD): concentrations of psyllium husk powder (PHP) and full cream milk powder (FCMP) in yoghurt samples*

No.	Concentration [% w/v]	
	PHP	FCMP
1.	0.05	2
2.	0.2	2
3.	0.05	5
4.	0.2	5
5.	0.0189	3.5
6.	0.2311	3.5
7.	0.125	1.379
8.	0.125	5.621
9.	0.125	3.5
10.	0.125	3.5
11.	0.125	3.5
12.	0.125	3.5
13.	0.125	3.5

Soluble Protein Content

Soluble protein (SP) was measured using the method described by Lowry *et al.* [15]. Samples for measurement were prepared by centrifuging the sample at 10000 rpm for 10 minutes to remove insoluble fraction. For measurement, a total of 0.5 mL of BSA solution (0.1; 0.5; 1; 2; 3; and 4 $mg \cdot mL^{-1}$) as standards or samples was put into a test tube. Then 2.5 mL of Lowry C reagent (50 mL of 2 % sodium carbonate in 0.1 N sodium hydroxide and 1 mL of 0.5 % copper sulfate pentahydrate in 1 % sodium tartrate) was added and allowed to stand for 10 minutes. A total of 0.25 mL of Folin-Ciocalteu solution was then added, shaken, and allowed to stand for 30 minutes. The absorbance was recorded at 750 nm using a spectrophotometer. The sample absorbance was plotted on the standard curve to calculate the concentration of soluble protein contained in the sample.

Sensory Acceptability

The method used to determine the sensory characteristics of yoghurt, in accordance with Tavakolipour *et al.* [19], is the hedonic test. Two types of yoghurt samples (control and test) were prepared for the hedonic test. Control yoghurt was prepared without addition of PHP and FCMP, while test yoghurt was prepared according to the obtained optimal conditions. Both types of yoghurt samples used in the hedonic test were added with sucrose to achieve a final concentration of 10 % (w/v). The addition of sucrose was intended to reduce sourness and enhance the sweetness of the sample. The hedonic test was based on panelist preference, which is assessed based on color, aroma, texture, taste, and overall appearance. Hedonic evaluation was performed by 15 semi-trained panelists using a five-point hedonic scale (1 = dislike, 2 = somewhat dislike, 3 = neutral, 4 = somewhat like, and 5 = like).

Statistical Analysis

All collected data was tabulated on Minitab 20 Statistical Software and analyzed using ANOVA. Factors were considered to have a significant effect when the p-value was lower than 0.05. All graphs reported in this study were also generated using Minitab 20.

RESULTS AND DISCUSSION

The prediction of protein in the present study is important, as the goal of the study was to improve the bioavailability of protein, primarily related to the amount of soluble protein in yoghurt. Using the obtained equation, one can calculate the optimal conditions (i.e., psyllium husk and full cream milk powder in this study) to achieve the highest soluble protein content. The experimental range for the concentration of psyllium husk and full cream milk powder refers to Safari *et al.* [17]. Table 3 shows the experimental SP content in comparison to the predicted values. The experimental protein levels closely match the predicted SP levels as calculated by equation (1), suggesting that the experiment is in a good agreement with the mathematical model obtained.

Table 3. *Result of soluble protein content in yoghurt samples*

No.	Concentration [% w/v]		Experimental SP [$mg \cdot mL^{-1}$]	Predicted SP [$mg \cdot mL^{-1}$]
	PHP	FCMP		
1.	0.05	2	2.201	2.05
2.	0.2	2	2.384	2.52
3.	0.05	5	2.869	2.70
4.	0.2	5	2.339	2.45
5.	0.019	3.5	2.097	2.32
6.	0.231	3.5	2.673	2.48
7.	0.125	1.379	2.247	2.25
8.	0.125	5.621	2.632	2.66
9.	0.125	3.5	1.971	1.95
10.	0.125	3.5	1.828	1.95
11.	0.125	3.5	2.320	1.95
12.	0.125	3.5	1.923	1.95
13.	0.125	3.5	1.707	1.95

Note: Experimental SP data were obtained from laboratory experiments with condition stated on the table, while predicted SP were calculated using equation (1)

Table 4 shows the results of the analysis of the variance of each factor on SP content. The p-value for the linear interaction effect of PHP is 0.503, while for FCMP is 0.121.

Since both p-values exceed 0.05, this indicates that neither individual component has a significant impact on SP content. However, the quadratic effects of both PHP and FCMP significantly affect the SP concentration, with p-values of 0.038 and 0.024, respectively. Furthermore, there is no significant interaction effect between PHP and FCMP (p-value = 0.172).

Table 4. *ANOVA table of the effect of full cream milk powder (FCMP) and psyllium husk powder (PHP) on concentrations of soluble protein (SP) in yoghurt*

Source	**DF**	**Adj SS**	**Adj MS**	***F*-Value**	***p*-Value**
Model	5	1.03802	0.20760	3.78	0.056
Linear	2	0.19770	0.09885	1.80	0.234
Psyllium Husk, [% w/v]	1	0.02733	0.02733	0.50	0.503
Full cream milk powder, [% w/v]	1	0.17037	0.17037	3.11	0.121
Square	2	0.71322	0.35661	6.50	0.025
Psyllium Husk (% w/v)*Psyllium Husk, [% w/v]	1	0.35720	0.35720	6.51	0.038
Full cream milk powder (%w/v)*Full cream milk powder, [% w/v]	1	0.44828	0.44828	8.17	0.024
2-Way Interaction	1	0.12709	0.12709	2.32	0.172
Psyllium Husk (% w/v)*Full cream milk powder, [% w/v]	1	0.12709	0.12709	2.32	0.172
Error	7	0.38403	0.05486		
Lack-of-Fit	3	0.17202	0.05734	1.08	0.452
Pure Error	4	0.21200	0.05300		
Total	12	1.42205			

Note: DF = Degree of freedom; Adj SS = Adjusted sum of squares; Adj MS = Adjusted mean of squares

Equation (1) illustrates the mathematical model for calculating the experiment's SP in relation to concentrations of PHP and FCMP. Equation (1) shows that the variables PHP and FCMP have a negative coefficient, indicating that increasing the amount of PHP and FCMP decreases the SP level in yoghurt. The regression equation in this study demonstrated that an increase in PHP and FCMP led to a decrease in soluble protein content. This similar with research from Bhat *et al.* [10] that found increasing in the fiber content from 0.1 % to 0.7 %, the protein content of yoghurt samples was found to decrease, which could be attributed to the dilution of protein content as psyllium husk has an almost negligible amount of protein content. Other than that, a decrease in soluble protein can occur because psyllium husk can increase the viscosity of yoghurt [20], so that the soluble protein component forms solids.

$$\begin{aligned} \mathit{Soluble\ protein\ content}\ [\mathrm{mg \cdot mL^{-1}}] = 2.830 - 3.75\mathrm{PHP} - 0.494\mathrm{FCMP} + \\ +40.3\mathrm{PHP \cdot PHP} + 0.1128\ \mathrm{FCMP \cdot FCMP} - 1.58\mathrm{PHP \cdot FCMP} \end{aligned} \tag{1}$$

Figure 1 shows the individual effect of PHP and FCMP on soluble protein content. Both graphs show a parabolic pattern where the SP content decreases at the beginning of the addition until it reaches its lowest point. Then, after reaching the lowest point, the addition of PHP and FCMP can increase the SP content in yoghurt. This happens because the lowest point is the optimum point, where each variable interacts to provide the smallest SP content. When the PHP level approaches 0.12 %, the soluble protein level decreases, but when the PHP level is greater than 0.12 %, the SP level will increase again. The same thing happens with FCMP, but the soluble protein level will reach its lowest point when FCMP is added as much as 3 % to the yoghurt.

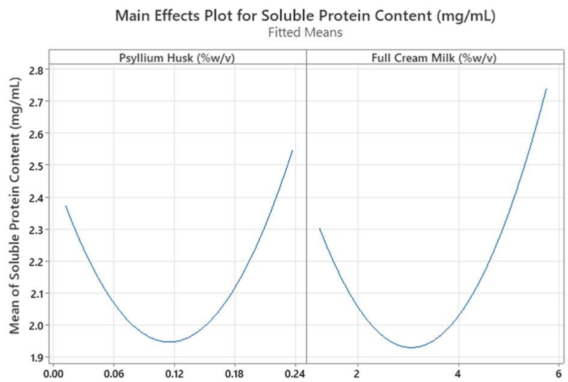

Figure 1. *Main effect plot of psyllium husk powder (PHP) and full cream milk powder (FCMP) for soluble protein content*

Figure 2 shows a contour plot illustrating the impact of PHP and FCMP on SP concentration. The minimum SP content can be obtained by adjusting the psyllium husk and full cream milk powder concentrations to approximately 0.06 - 0.15 % w/v and 2 - 3.7 % w/v, respectively, as shown in Figure 2. To enhance soluble protein (SP) content, the ratio of psyllium husk powder (PHP) to full cream milk powder (FCMP) can be adjusted by either decreasing PHP while increasing FCMP or increasing PHP while reducing FCMP. Using a low amount of PHP will result in excessive syneresis of the yoghurt. To prevent this, adjustments should be made to ensure the protein content is high while minimizing syneresis.

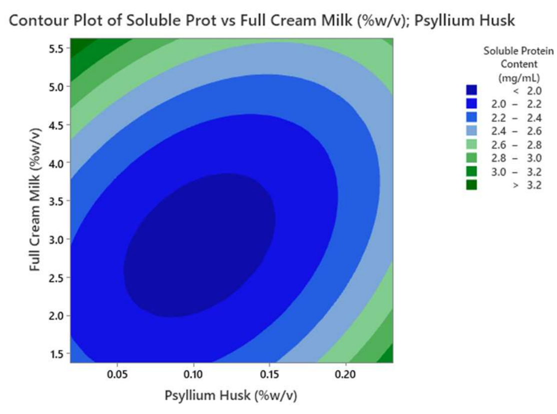

Figure 2. *Contour plot of psyllium husk (PHP) and full cream milk powder (FCMP) effect on soluble protein content in yoghurt*

Figure 3 shows the optimized combination of PHP and FCMP to reach the highest SP concentration. Figure 3 illustrates that SP levels are optimized to achieve excellent nutritious content in yoghurt production. High SP levels were achieved by optimizing each variable, resulting in a PHP of 0.0189 % w/v and FCMP of 5.6213 %. The optimal condition suggested by the software has a low PHP content, which can lead to high syneresis and eventually may reduce consumer preference due to unattractive texture and overall appearance [17]. Thus, the PHP content was elevated at the expense of the SP level. The ideal concentration of PHP to minimize syneresis is around 0.16 % w/v, as stated by Safari *et al.* [17]. Therefore, in this investigation, the concentration of PHP was adjusted to 0.162 % w/v and the optimization results are presented in Figure 4. Under these conditions, it is expected that the yoghurt will exhibit low syneresis, a rich flavor attributed to a high full cream milk powder content, and a reasonably high soluble protein level.

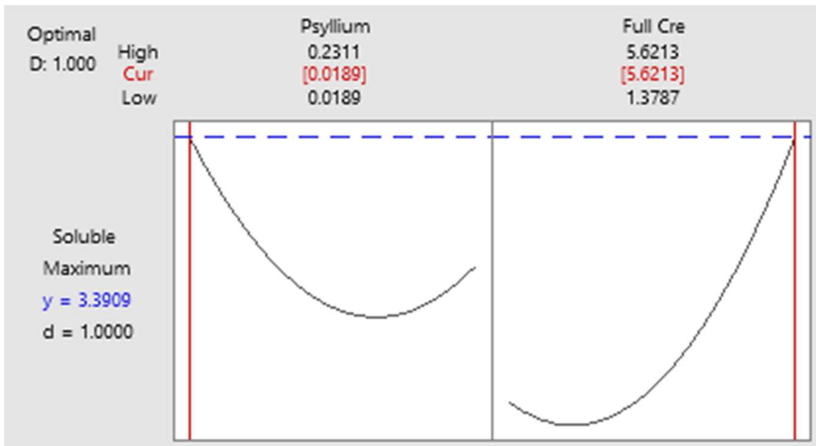

Figure 3. *Graph of optimal concentrations of psyllium husk powder (PHP) and full-cream milk powder (FCMP) for maximum soluble protein content in yoghurt*

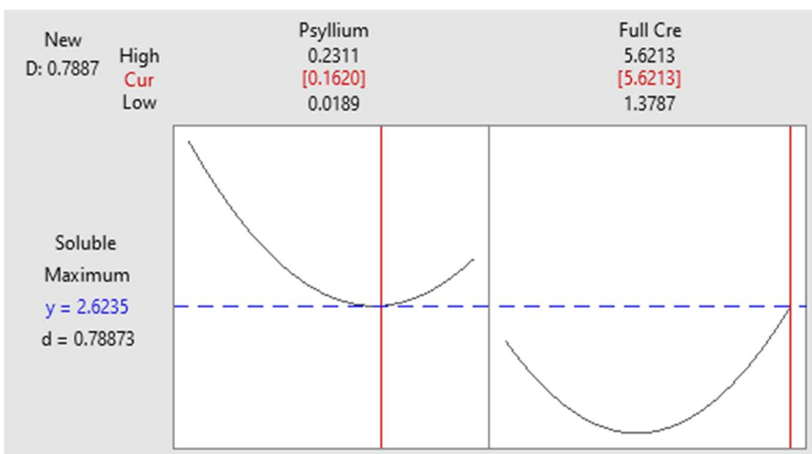

Figure 4. *Adjusted optimal concentrations of psyllium husk powder (PHP) and full-cream milk powder (FCMP) for high soluble protein content in yoghurt*

Table 5 shows the validation results of the optimal combination of PHP and FCMP. The experimental results were found to be in an agreement with the calculated optimum value. This suggests that the mathematical model obtained in this work can be utilized to vary the soluble protein content of yoghurt by adjusting PHP and FCMP. To the authors' knowledge, no reports have specifically mentioned the soluble protein content in yoghurt products. Most studies report only total protein content typically ranging from 2 - 8 % w/v [21 – 23]. In the present study, the soluble protein content at the center point of the experiment was 1.95 $mg \cdot mL^{-1}$, while under the suggested optimal condition it increased to 2.704 $mg \cdot mL^{-1}$, indicating approximately a 1.4-fold increase compared to the center point condition.

Table 5. *Validation results of optimal conditions of psyllium husk powder (PHP) and full-cream milk powder (FCMP) to get a high soluble protein (SP) content in yoghurt*

Condition	**Experimental Variable**		**Response**
	PHP [% w/v]	**FCMP [% w/v]**	**SP [$mg \cdot mL^{-1}$]**
Center point	0.125	3.5	1.95
Optimum (calculated)	0.162	5.621	2.869
Validation 1	0.162	5.621	2.588
Validation 2			2.782
Validation 3			2.741
Average			**2.704 ± 0.102**

Figure 5 shows the result of the organoleptic test. The test was carried out to evaluate the panelists' preference for the product. The control in the organoleptic test consisted of yoghurt without PHP and FCMP. The test sample included 0.162 % w/v PHP and 5.621 % w/v FCMP, as verified in the validation result.

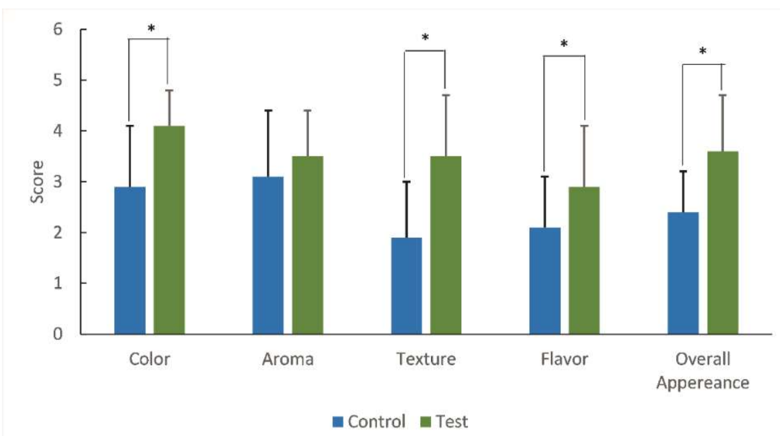

Figure 5. *Sensory acceptability results of test yoghurt samples with optimized concentrations of psyllium husk powder (PHP, 0.162 % w/v) and full-cream milk powder (FCMP, 5.621 % w/v) compared to control samples without PHP or FCMP The yoghurts were assessed by 15 semi-trained panelists.*
Data shown are mean values and error bars represents the standard deviation.
**Indicate significant differences ($p < 0.05$) between samples based on t-test.*

According to Figure 5, the panelists' preference for color, texture, flavor, and overall appearance of the test yoghurt is significantly higher than that of the control yoghurt. Only the aromas were not significantly different between the test and control yoghurts. The results are in agreement with a recent study by Vasilean *et al.* [13], which found that panelists preferred yoghurt with the highest fat content. Additionally, psyllium husk can decrease syneresis, enhancing the texture of yoghurt, potentially leading to a higher acceptance among panelists, as noted by Safari *et al.* [17]. The flavor of yoghurt can also be enhanced by adding full cream milk powder, as indicated by the higher sensory scores of the test yoghurt compared to the control. This finding is consistent with the reports by Lucey *et al.* [24] and Rashidi *et al.* [25]. Overall appearance scores of test yoghurt also were significantly higher than control, and this result is in agreement with findings by Rashidi *et al.* [25].

CONCLUSIONS

The aim of this study was to prepare yoghurt with a high soluble protein (SP) content, while maintaining a low syneresis and creamy taste by optimizing the concentrations of added psyllium husk powder (PHP) and full-cream milk powder (FCMP). The findings of this study suggest that it is optimal to add PHP and FCMP as much as 0.162 % w/v and 5.621 % w/v, respectively. Yoghurt prepared under these conditions has a projected soluble protein value of 2.869 $mg \cdot mL^{-1}$, while the validation findings indicated a protein content of 2.704 ± 0.102 $mg \cdot mL^{-1}$, which show a good agreement between the model and experimental data. The sensory acceptability results indicated that the color, texture, taste, and overall acceptance of the test yoghurt were significantly more acceptable than those of the control yoghurt. Nevertheless, the aroma of both yoghurts did not show a substantial variance.

ACKNOWLEDGEMENT

The present study was supported by The Ministry of Education, Culture, Research and Technology Republic of Indonesia through Regular Fundamental Research scheme with contract number 3018/UN6.3.1/PT.00/2023 and Directorate of Research and Community Service Universitas Padjadjaran through Academic Leadership Grant with contract number 1549/UN6.3.1/PT.00/2023. Therefore, we would like to acknowledge the funding support.

REFERENCES

1. Dabija, A., Codină, G.G., Gâtlan, A.-M., Sănduleac, E.T., Rusu, L.: Effects of Some Vegetable Proteins Addition on Yogurt Quality, *Scientific Study & Research. Chemistry & Chemical Engineering, Biotechnology, Food Industry*, **2018**, **19** (2), 181-192;
2. Taneva, I., Zlatev, Z.: Total Phenolic Content and Antioxidant Activity of Yoghurt with Goji Berries (*Lycium Barbarum*), *Scientific Study & Research. Chemistry & Chemical Engineering, Biotechnology, Food Industry*, **2020**, **21** (1), 125-131;
3. Hadjimbei, E., Botsaris, G., Chrysostomou S.: Beneficial Effects of Yoghurts and Probiotic Fermented Milks and Their Functional Food Potential, *Foods*, **2022**, **11** (17), 2691;
4. Mahomud, M.S., Katsuno, N., Nishizu T.: Role of Whey Protein-Casein Complexes on Yoghurt Texture, *Reviews in Agricultural Science*, **2017**, **5**, 1-12;
5. Zhang, J., Wang, J., Li, M., Guo, S., Lv, Y.: Effects of Heat Treatment on Protein Molecular Structure and In Vitro Digestion in Whole Soybeans with Different Moisture Content, *Food Research International*, **2022**, **155**, 111115;
6. Behrouzain, F., Razavi, S.M., Joyner, H.: Mechanisms of Whey Protein Isolate Interaction with Basil Seed Gum: Influence of *p*H and Protein-Polysaccharide Ratio, *Carbohydrate Polymers*, **2020**, **232**, 115775;

7. Sebayang, F., Bulan, R., Wahyuni W.: The Utilization of Carboxymethyl Cellulose (CMC) from Groundnut (*Arachis hypogaea* L) Cellulose as Stabilizer for Cow Milk Yogurt, *Journal of Chemical Natural Resources*, **2019**, **1** (2), 38-51;
8. Fennema, O.R.: *Food Chemistry*, Marcel Dekker, New York, **1996**;
9. Ladjevardi, Z.S., Gharibzahedi, S.M.T., Mousavi M.: Development of a Stable Low-Fat Yogurt Gel Using Functionality of Psyllium (*Plantago ovata* Forsk) Husk Gum, *Carbohydrate Polymers*, **2015**, **125**, 272–280;
10. Bhat, S.V., Deva, A.M., Amin T.: Physicochemical and Textural Properties of Yogurt Fortified with Psyllium (*Plantago Ovata*) Husk, *Journal of Food Processing and Preservation*, **2018**, **42** (2), e13425;
11. Fu, Q.-Q., Liu, R., Zhou, L., Zhang, J.-W., Zhang, W.-G., Wang, R.-R.: Effects of Psyllium Husk Powder on the Emulsifying Stability, Rheological Properties, Microstructure, and Oxidative Stability of Oil-in-Water Emulsions, *Food Control*, **2022**, **134**, 108716;
12. Chen, C., Zhao, S., Hao, G., Yu, H., Tian, H., Zhao, G.: Role of Lactic Acid Bacteria on the Yogurt Flavour: A Review, *International journal of food properties*, **2017**, **20** (sup1), S316-S330;
13. Vasilean, I., Aprodu, I., Patrascu L.: Fat Content in Yoghurts versus Non-Fat Fortifying–A Rheological and Sensorial Approach, *Studia Universitatis Babes-Bolyai, Chemia*, **2015**, **60** (2), 259-269;
14. Rima, R., Ishmayana, S., Malini, D.M., Soedjanaatmadja, U.M.S.: Nutritional Content and the Activities of L-Dopa (L-3, 4-dihydoxyphenyalanine) from *Mucuna pruriens* L. DC Seeds of Central Java Accession, *Arabian Journal of Chemistry*, **2023**, **16** (1), 104390;
15. Lowry, O.H., Rosebrough, N.J., Farr, A.L., Randall, R.J.: Protein Measurement with the Folin Phenol Reagent, *Journal of Biological Chemistry*, **1951**, **193** (1), 265-275;
16. Walker, J., Harmon, D.: A Simple, Rapid Assay for A-Amylase in Bovine Pancreatic Juice, *Journal of Animal Science*, **1996**, **74** (3), 658-662;
17. Safari, A., Devi, P.R., Horasio, D.T., Apandi, S.D., Nurrusyda, F.S., Fadhlillah, M., Soedjanaatmadja, U.M., Ishmayana S.: Improvement of Syneresis and Viscosity of Yoghurt with the Addition of Psyllium Husk (*Plantago Ovata*) and Full Cream Milk with Response Surface Method Approach, *International Journal of Chemical and Biochemical Sciences*, **2023**, **24** (9), 132-139;
18. Abdelazez, A., Muhammad, Z., Zhang, Q.-X., Zhu, Z.-T., Abdelmotaal, H., Sami, R., Meng, X.-C.: Production of a Functional Frozen Yogurt Fortified with *Bifidobacterium* spp, *BioMed research international*, **2017**, **2017** (1), 6438528;
19. Tavakolipour, H., Vahid-moghadam, F., Jamdar, F.: Textural and Sensory Properties of Low-Fat Concentrated Flavored Yogurt by Using Modified Waxy Corn Starch and Gelatin as a Fat Replacer, *International Journal of Biosciences*, **2014**, **5** (6), 61-67;
20. Sodini, I., Montella, J., Tong, P.S.: Physical Properties of Yogurt Fortified with Various Commercial Whey Protein Concentrates, *Journal of the Science of Food and Agriculture*, **2005**, **85** (5), 853-859;
21. Matela, K.S., Pillai, M.K., Matebesi-Ranthimo, P., Ntakatsane, M.: Analysis of Proximate Compositions and Physiochemical Properties of Some Yoghurt Samples from Maseru, Lesotho, *Journal of Food Science and Nutrition Research*, **2019**, **2** (3), 245-252;
22. Purwantiningsih, T.I., Bria, M.A.B., Kia, K.W.: Levels Protein and Fat of Yoghurt Made of Different Types snd Number of Cultures, *Journal of Tropical Animal Science and Technology*, **2022**, **4** (1), 66-73;
23. Tamime, A.Y., Hickey, M., Muir, D.D.: Strained Fermented Milks–A Review of Existing Legislative Provisions, Survey of Nutritional Labelling of Commercial Products in Selected Markets and Terminology of Products in Some Selected Countries, *International Journal of Dairy Technology*, **2014**, **67** (3), 305-333;
24. Lucey, J., Munro, P., Singh, H.: Rheological Properties and Microstructure of Acid Milk Gels as Affected by Fat Content and Heat Treatment, *Journal of Food Science*, **1998**, **63** (4), 660-664;
25. Rashidi, H., Kashaninejad, M., Bolandy, M., Ghaffary, S.: Investigation of the Effect of Starter Type and Formulation on Physicochemical and Sensory Properties of Concentrated Yoghurt Using Mixture Design and Two-Stage Clustering Method, *Research and Innovation in Food Science and Technology*, **2021**, **10** (1), 43-58.