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Optimization of Propolis and Vegetable Oils-based Soap Formulation to Enhance Product Quality and Antioxidant Properties

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Abstract

Consumers demanded the quality of solid soap for the bath were having a solid texture, high foam stability, and fulfillment of the national quality standard in SNI 3522:2021. Soap characteristics and quality are affected by soap bases and the existence of natural additive matter with antioxidant properties such as propolis extracts. The study aims to determine an optimum formula for soap production using Response Surface Methodology (RSM) with Box-Behnken experimental design. The research conducted in several stages, its consist to propolis extraction, soap formulation and production, RSM analysis towards predetermined responses, model validation, and products characterization (in optimum formulation). Optimized factors consist of the content of propolis extracts, coconut oils, palm oils, and olive oils, while the response was moisture content, foam stability, and antioxidant inhibition. The results showed each response has a significant model to get an optimum formula for propolis extracts soap production. The optimum formula for soap production requires 1.545 g propolis extracts, 13.097 g coconut oils, 29.629 g palm oils, and 29.809 g olive oils with the model validation for each response were 95.5% moisture content, 96.5% foam stability, and 97.5% antioxidant inhibition. Soap characterization in the optimum formula fulfilled the quality standard requirement in SNI 3522:2021 its consists of 20.24% moisture content, 2.22% insoluble ingredients in ethanol, and 0.07% free alkali content.

Keywords: Box-Behnken Design, Formula Optimization, Propolis Extracts, RSM, Solid Soap.

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1. Introduction

One of the cosmetic products were widely used and mostly distributed in the community is solid soap for baths. Soap is a cosmetic product with a treat, protection, and hygiene function for skin from any dirt material [1]. The demand for solid or bath soap increased continuously linear with the increase in population, especially in Indonesia. The high demand for soap impacted Indonesia to import any soap products from others. Central Agency on Statistics reported in February 2022 that Indonesia imported more than 18.9 thousand tons of soaps and similar products [2]. The report showed soap production in Indonesia has been not able to supply the domestic demand independently.

Solid soap production deployed the saponification reaction mechanism between fatty acids and alkalis which is carried through a cold process, hot process, or melt and pour methods [3]. Fatty acids were widely used for soap production was obtained from coconut oils, palm oils, and olive oils as the source, while the alkali was sodium hydroxide (NaOH) for solid soap and potassium hydroxide (KOH) for liquid soap [4]. Olive oils could be provided soft and gentle characteristics to soap caused of their high oleic acid content which has a moisturizing effect on the skin [5]. It made the soap has a soft texture and was not demanded by consumers in the solid soap category. Different from palm oils, high palmitic acid content in oils could produce a very hard or solid soap but the product would have a low foam-making capability [6]. High foam-making in soap production could be provided by coconut oils with high lauric acid content as foam-making agents. Therefore, although coconut oils-based soap has a good capability of making foam but produced soap would be having a semi-solid texture [3]. Physical characteristics like texture and foam-making capability are important attributes in solid soap for baths and could be affecting the interest of consumers.

Consumers assumed the soap that produces a lot of foam can clean dirt well [7]. Solid soap was chosen by communities for economic reasons, easy to use, and more efficient in skin cleaning than liquid soap [8]. The combination of three types of vegetable oils (coconut oils, palm oils, and olive oils) aimed to produce a soap with high foam capability, a solid texture and could be moisturized skin.

Provided soaps in the market mostly use synthetic antioxidants as an additive and could interfere with consumer skins [9]. The use of synthetic additives could be replaced by natural matters for skin safety and pointed to the green technology concept. One natural matter that could be deployed as an alternative antioxidant source for soap production is propolis extracts. Propolis is a natural matter collected by stingless bees or honeybees such as Trigona sp. and was used as a nest, a component of defense, external immune systems, and antimicrobials. Propolis was one of the antioxidant sources its rich in bioactive compounds such as phenolics and flavonoids [10]. The existence of bioactive compounds in propolis has pharmacological activities such as antibacterial, anti-inflammatory, antiseptic, antimutagenic, and antihepatotoxic [11,12,13]. Propolis could be used as a proper antioxidant source to be applied to soap formulation and enhance product performances. Recent studies of the uses of propolis extracts in soap production have been studied by Astuti et al., but this study has limitations in product characteristics [14].

Optimization in soap formulation, especially in vegetable oils-based soaps consisting of coconut oils, palm oils, and olive oils required to be conducted. Emerged demand of optimum formulation for solid soap was needed in industry, this prompted a study on optimization formula for solid soap which then carried out in this study. Optimum formulation that resulted by this study could be considerate to conduct further research on scale-up production, especially for pilot-scale and industrial-scale. The optimization could be deployed through Response Surface Methodology (RSM) with a Box-Behnken experimental design that is considered efficient because requires fewer experiments and reduced analysis costs [15]. This study aimed to optimize the content of propolis extracts, coconut oils, palm oils, and olive oils to obtain the optimum formula. The expected optimum formula was soap production which had minimum moisture content, maximum foam stability, and maximum antioxidant activity.

2. Methods

Materials

The materials deployed in this study consist of coconut oils, palm oils, olive oils, propolis extracts, and honey fragrances bought from local market in Bandung. Chemicals obtained from Merck (Germany) and consist of sodium hydroxide (NaOH), 2,2-diphenyl-1-picrylhydrazyl (DPPH), methanol absolute, 0.1 N potassium hydroxide (KOH), 0.1 N hydrochloride acids (HCl), 1% phenolphthalein, and distilled water.

Propolis Extracts Preparation

Propolis extraction was carried out by maceration technique following Yarlina et al. [16] with slight modifications. Crude propolis was soaked in 70% ethanol with a 1:3 ratio (w/v). The mixture was macerated for 24 hours, filtrated, and evaporated to obtain concentrated extracts. The number of extracts was measured and then determined by the yields.

Solid Soap Formulation

The standard formulation for soap production followed Sukawati et al. [17] with slight modification and shown in Table 1.

Table 1. The standard formulation for propolis solid soap

Ingredients	Quantity (gram)	Role of Ingredients
Propolis Extracts	1-3	Bioactive Matter
Coconut Oils	10-20	Soap Base
Palm Oils	10-30	Soap Base
Olive Oils	10-30	Soap Base
NaOH	8.9	Alkali
Distilled Water	20	Vehicle
Honey Fragrance	1.5	Fragrance/ Perfume

Production of Solid Soap

Solid soap production followed the Garden [18] procedure. The vegetable oils consisting of coconut oils, palm oils, and olive oils were blended with NaOH solution (under 40°C) and then agitate in the mixture continuously until the foam was traced. Propolis extracts were added and agitate the mixture until homogenous. The homogenous mixture was ready to mold and allowed to solidify for 24 hours, then the solid soaps were stored in a dry place for 2 weeks before product evaluation was conducted.

Antioxidant Activity Assay

Antioxidant activity was analyzed by 2,2-diphenyl-1-picrylhydrazyl methods to scavenge the free radical followed by Hayati et al. [19] with slight modification. The amount of 2 mL of soap extracts in methanol was added to 2 mL of 0.2 mM DPPH, shaken, and placed in a dark room for 30 minutes. The mixtures were quantified using a spectrophotometer UV-Vis at 517 nm and methanol was used as a blank. The percentage of the free radical scavenging effect is determined by dividing the difference between blank and sample absorbance with the absorbance of the blank. The results are then used as a response in statistical analysis.

Solid Soap Characterization

Some characteristics were analyzed to determine the quality of products. Moisture content, pH-value, insoluble ingredients in ethanol, and free alkali content followed standard procedure and parameters in SNI 3532:2021 [20]. Foam stability was analyzed by procedure following Fatimah & Jamilah [21]. Two characteristics then determined as responses in statistical analysis consist of moisture content and foam stability.

Experimental Design

Response Surface Methodology (RSM) was deployed in this study and applied Box-Behnken experimental design. Box-Behnken design was deployed in this study its cause of efficient to use and required fewer experimental samples that accordance with costs for analysis [15]. Efficient and costs were required aspects for products development in industry and its has been main consideration aspects in this study. There were 4 factors shown in Table 2 consist of propolis extracts (A), coconut oils (B), palm oils (C), and olive oils (D) was evaluated. Analysis and data processing was deploying Design Expert 13.0.5.0 to obtain 27 different formulations and predicted optimum formulation. The optimum formulation was determined by adjusted goal setting with some criteria such as minimization of moisture content, maximization of foam stability, and antioxidant inhibition. Validation and comparing between predicted and actual optimum formulas were then conducted to obtain the best solid soap formulation. The best formulation then analyzed towards another characteristic pointed to SNI 3522:2021 (except the characteristics were used as a response).

Table 2. Experimental design parameters

Symbols	Factors	Level of Variables (gram)	
		Low	High
A	Propolis Extracts	1	3
B	Coconut Oils	10	20
C	Pal Oils	10	30
D	Olive Oils	10	30

3. Result and Discussion

Propolis Extraction Yields

Propolis extraction yields in this study were measured as an amount of 24.45% (w/w). Propolis extract has been had brown color after evaporating until no solvent droplets were evaporating. Moderate results were obtained because of the effect of ethanol concentration that affected solvent polarity. A high concentration of ethanol provided propolis extracts in high yield as in the research of Puspita & Pramono [22] was deploying 96% ethanol (v/v) which obtained 51.67% of yield (w/w). According to Riwanti et al. [23], ethanol concentration affected their polarity which had nonlinear effects and caused the solvent difficulty in binding bioactive compounds as an antioxidant. Therefore, to obtain high antioxidant extracts were recommended to deploy a low concentration of ethanol instead of a high concentration to bind bioactive compounds such as phenols and flavonoids in high content [16].

Response Surface Model

Box-Behnken's experimental design provided 27 combinations of formulas based on the limitation of input variables. The results of the analysis of responses consisting of moisture content, foam stability, and antioxidant inhibition were shown in Table 3. The data were then analyzed using Design Expert 13.0.5.0 to determine the factors toward responses and to obtain the best formulation for solid soap

Table 3. Data analysis results for optimization of solid soap formulation

Formula(s)	Propolis Extracts (gram)	Coconut Oils (gram)	Palm Olis (gram)	Olive Oils (gram)	Moisture Content (%)	Foam Stability (%)	Antioxidant Inhibition (%)
1	2	20	30	20	20.68	88	49.13
2	2	20	10	20	21.94	89	50.75
3	2	15	20	20	23.37	89	50.69
4	1	15	10	20	27.24	62	51.05
5	1	15	20	10	24.82	62	50.26
6	1	10	20	20	25.38	85	49.42
7	3	20	20	20	21.38	89	50.99
8	2	15	30	10	21.95	85	51.77
9	2	10	20	10	29.82	71	49.90
10	3	15	10	20	33.51	61	48.69
11	2	20	20	30	19.50	90	51.97
12	1	15	30	20	19.41	88	51.79
13	3	15	20	10	26.43	72	50.38
14	2	15	30	30	19.64	88	51.95
15	3	10	20	20	22.96	85	49.92
16	2	10	20	30	21.59	91	51.87
17	2	15	20	20	20.58	85	51.79
18	1	20	20	20	19.59	88	51.53
19	2	15	10	10	27.44	64	48.79
20	1	15	20	30	20.00	91	51.29
21	2	15	20	20	22.43	84	50.26
22	3	15	20	30	22.59	92	50.14
23	3	15	30	20	20.48	87	50.95
24	2	10	30	20	22.20	91	51.85
25	2	20	20	10	24.89	84	49.19
26	2	10	10	20	29.55	61	47.90
27	2	15	10	30	21.58	86	51.15

Analysis of variances (ANOVA) was deployed in this study on the response of moisture content, foam stability, and antioxidant inhibition to determine a fit model to show the factors' effect on responses. The quadratic model has a greater determinant coefficient (R2) than the other models, chosen as fit models shown in Table 4. This model was significantly based on p-value criteria which are smaller than 5% and have a not significant on the Lack of Fit criteria (the value was greater than 5%). According to [Keshani et al. in Nurmiah et al. [24], no significant effect on the Lack of Fit value is a requirement for a good model, especially toward foam stability response which fits with the model.

Table 4. Results of response model analysis

Response	Moisture Content	Foam Stability	Antioxidant Inhibition
Models	Quadratic	Quadratic	Quadratic
Model significant (p<0,05)	0.0119	0.0017	0.0389
Factors significant (p<0,05)	A : 0.1910 B : 0.0113 C : 0.0005 D : 0.0022	A : 0.6143 B : 0.0420 C : 0.0002 D : 0.0002	A : 0.1567 B : 0.3581 C : 0.0073 D : 0.0144

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Response	Moisture Content	Foam Stability	Antioxidant Inhibition
Lack of fit (p<0,05)	0.2855	0.1738	0.5717
R^2	0.8193	0.8758	0.7679
Equation	$Moisture\ Content = 69.37 - 3.685A - 2.1235B - 1.12792C - 0.870917D + 0.2105AB - 0.13AC + 0.0245AD + 0.03045BC + 0.0142BD + 0.008875CD + 0.886667A^2 + 0.0139167B^2 + 0.0111542C^2 + 0.00444167D^2$	$Foam\ Stability = 86 + 0.83A + 3.67B + 8.67C + 8.33D + 0.25AB + 3.60689e16AC - 2.25AD - 7.75BC - 3.5BD - 4.75CD - 4A^2 + 2.25B^2 - 5.5C^2 - 2.25D^2$	$Antioxidant\ Inhibition = 50.9133 - 0.3558A + 0.225B + 0.7592C + 0.6733D - 0.26AB + 0.38AC - 0.3175AD - 1.3925BC + 0.2025BD - 0.545CD - 0.1817A^2 - 0.4304B^2 - 0.2617C^2 + 0.0996D^2$

Effects on Moisture Content

The moisture content equation in Table 4 showed the moisture content in solid soap declined although propolis extracts, coconut oils, palm oils, and olive oils were increased. This is indicated by negative constants for each factor. The increase in moisture content was not affected by the addition of propolis extracts cause of containing saponins. Saponins in propolis extracts are complex glycosides when hydrolyzed would be divided into sugars and non-sugar components especially sugar components that have hygroscopic properties that could absorb water. Widyasanti et al. [25] conducted a study of saponins in white tea extracts caused a declined effect on moisture content in transparent solid soap. This shows about more extracts are added, it would be a high amount of water absorption by sugars in saponins and cause the moisture content in solid soap to decline.

Coconut oils, palm oils, and olive oils showed a significant effect on moisture content in soap and indicated by the p-value of this response being less than 5%. High moisture content in soap would be caused the soap to shrink faster, but low content could be increasing its shelf life. Besides that, the moisture content in soap was related to hardness level, which means high content produces a softer soap and vice versa [8]. Hardian et al. explained saturated fatty acids affected the hardness level of soap, such as a high amount of palmitic acids, stearic acids, myristic acids, and lauric acids made the harder soap [26]. Palmitic acids and stearic acids can harden and or solidify soap [27,28]. Coconut oils, palm oils, and olive oils have a high amount of saturated fatty acids, and increasing the amount of these would be to increase the hardness level or decline the moisture content in soap.

The three-dimensional curve obtained from Design Expert 13.0.5.0 showed the relationship between factors and responses. Each color in curves represented the moisture content of each response. Red colors indicated high moisture content in soap, meanwhile blue indicated low moisture content. The lowest surface showed minimum moisture content in soap and it indicated then the higher content of coconut oils, palm oils, and olive oils provided low moisture content in soaps.

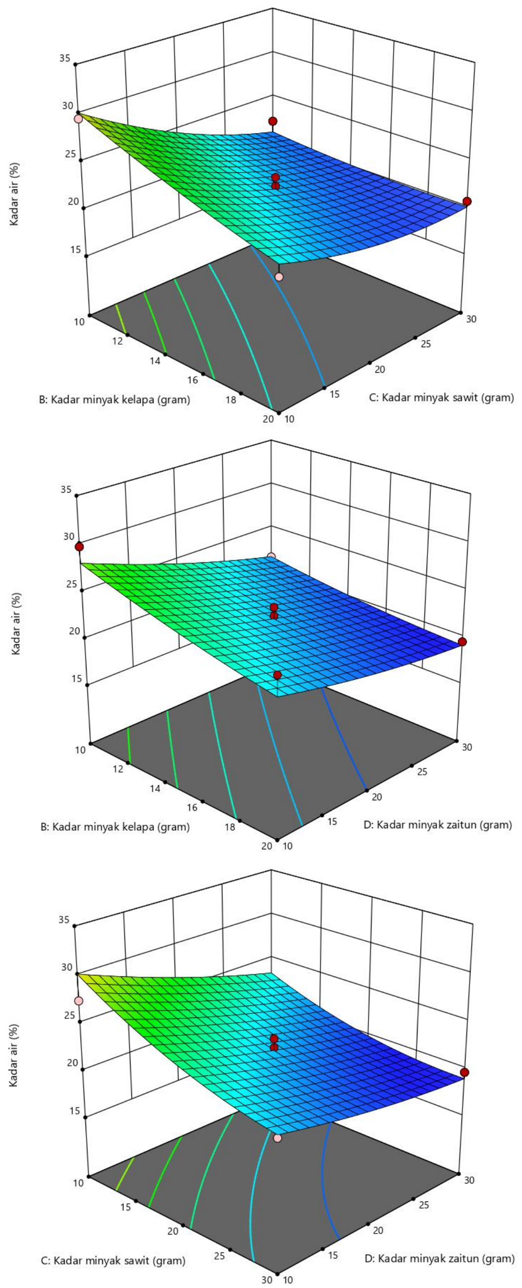

Figure 1. The three-dimensional response curve of moisture content

Effects on Foam Stability

The response surface model equation showed that all factors (propolis extracts, coconut oils, palm oils, and olive oils) affected linearly foam stability response. The positive constant in the equation could be interpreted by the increasing content of each factor improving foam stability in soaps produced. However, the addition of propolis extracts had no significant effect on foam stability caused by a *p-value* of more than 5%. Pangestika et al. [29] stated the addition of natural extracts in excess content caused soap formulation to have a mismatched composition to produce foams and affected to declined of foam stability. The declined foam stability of soap occurred in recent research in which excess addition of Avicennia marina leaves extracts to solid soap formulation [29].

Significant factors toward foam stability in soap were the content of coconut oils, palm oils, and olive oils. The increase in foam stability is related to high fatty acid content in the vegetable oils deployed in this study. Coconut oil contains lauric acids as the abundant component in oil which is recovered to 52% [30]. Lauric acids produced foam in coconut oil-bases soap to provide a high foaming ability [31]. Palmitic acid contained in coconut oil, palm oil, and olive oil also can produce a stable foam [28]. According to Oktari et al. [27], stearic and myristic acids in three types of vegetable oils in this study could be maintained foam stability. Therefore, the high content of coconut oils, palm oils, and olive oils increased foam stability in produced solid soaps. In a study conducted by Mopangga et al. [32], the results of produced soap deployed a combination of three-types vegetable oils same as in this study, and it obtained soap with good foam stability and required the standard.

The three-dimensional graph of the foam stability response shown in Figure 2, interpreted the influence of significant factors on responses. The parabolic-shaped curve showed the maximum foam stability at the top surface before the decline occurred. The position of maximum foam stability was indicated on the highest surface compared to others.

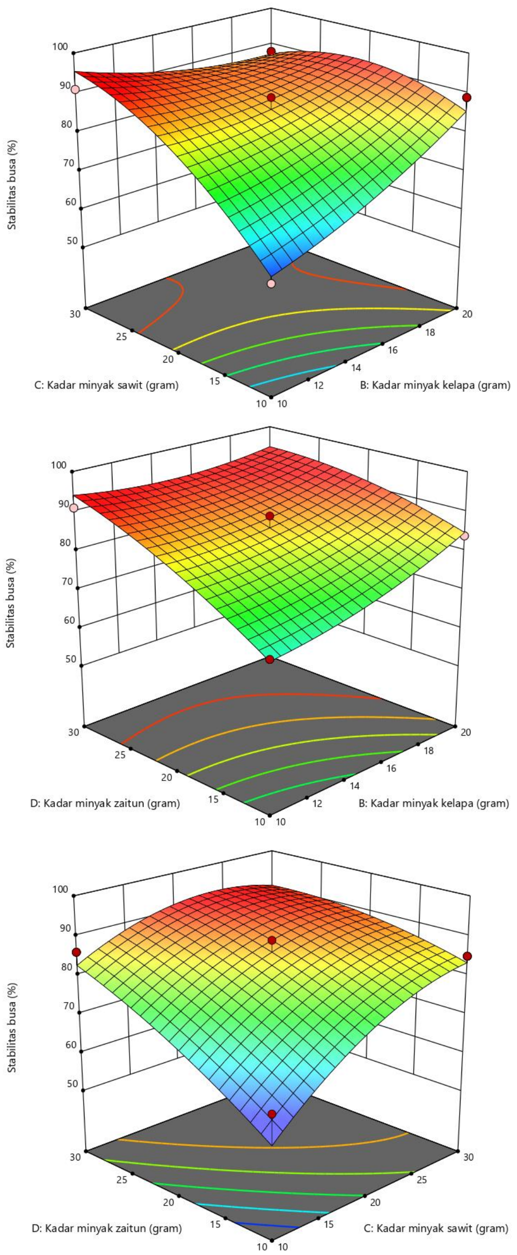

Figure 2. The three-dimensional response curve of foam stability

Effects on Antioxidant Inhibition

The results of ANOVA in antioxidant inhibition response, the significant factors consist of palm oils and olive oils (*p-value* less than 5%), it related to antioxidant content in oils as to increase the inhibition linearly with the addition of palm oils and olive oils. Sinaga & Siahaan [33] stated palm oils contained carotenoids and vitamin E as tocopherol and tocotrienol which have antioxidant properties. Yuniwarti et al. [34] explained olive oils have antioxidant compounds such as phenols. Vitamin E as tocopherol in olive oils has antioxidant properties and can protect skin from free radicals [5]. Antioxidant activity was discovered in coconut oils cause of the existence of polyphenols and vitamin E [35]. However, coconut oils have no significant effect on antioxidant inhibition based on the results of ANOVA. It is caused by during soap-making processes, coconut oils must be heated firstly to melt. The heating process could be harmful to thermolabile compounds such as flavonoids [36].

The increased content of propolis extracts had no significant effect on antioxidant inhibition response. It was caused by the blending process of propolis extracts with various non-polar compounds of oils in the soap-making process. That process inhibited flavonoids in propolis extracts to bind free radicals of DPPH and remain bound to its glycoside groups. Flavonoids were unable to donate their hydrogen ions to scavenging free radicals due to steric block [37]. Sari et al. [38] discovered similar mechanisms in their study about liquid soap-making with the addition of guava leaf extracts. Their results showed the addition of extracts decreased the antioxidant inhibition in soap and possibly occurred in saturated extracts.

The three-dimensional curve of antioxidant inhibition response was shown in Figure 3 and indicated the maximum antioxidant inhibition on the surface which is the highest compared to others. The higher content of coconut oils and palm oils showed a color change from blue to red. It showed the increase in these two factors affects increasing in antioxidant inhibition.

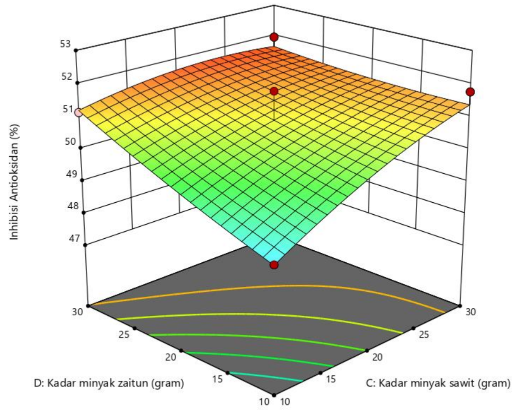

Figure 3. The three-dimensional response curve of antioxidant inhibition.

Optimization and Validation

Optimization assisted by Design Expert 13.0.5.0 provided the optimum formula of soap consisting of 1.545 grams of propolis extracts (A), 13.097 grams of coconut oils (B), 29.629 grams of palm oils (C), and 29,809 grams of olive oils. The predicted response generated by models were 19.36% of moisture content, 93.67% of foam stability, and 51.99% of antioxidant inhibition. The predicted solution then proceeded to validation models to determine the actual level of solution. Validation was conducted by deploying the optimum formula obtained from the program actually with two replications, and the results of the calculation showed in Table 5.

Model validation was carried out to determine model accuracy by comparing the data predicted by a model with the actual data from the analysis. The results shown in Table 5 indicated the deviation of moisture content, foam stability, and antioxidant inhibition respectively amount of 4.55%, 3.48%, and 2.54%. According to Sugiono et al. in Nurika et al. [39], the value of deviation aimed to measure accuracy between predicted and actual data and should be less than or equal to 5%. The validation results for all responses in this study were below 5% and were considered to meet the requirements for computational prediction calculations. The results of the validation of the optimal formula according to the predicted efficiency indicated the model was adequate for the optimization of factors or variables, Khoshnamvand et al. in Nurika et al. [39].

Table 5. Comparison of optimization and validation results

	Moisture Content (%)	Foam Stability (%)	Antioxidant Inhibition (%)
Predicts*	19.36	93.67	51.99
Validation**	20.24	90.41	50.67
Difference	0.88	3.26	1.32
Deviation (%)	4.55	3.48	2.54
Model Validity (%)	95.45	96.52	97.54

Further Analysis of Optimum Formula

Further analysis of the optimum formula was carried out to analysis toward several criteria following SNI 3522:2021 such as pH value, insoluble material in ethanol, free fatty acids, and free alkali. The results were then compared with the standard in SNI 3522:2021 and shown in Table 6.

Table 6. Results analysis for optimum formula

Parameters of Analysis	Quantity	
	Optimum Formula	SNI 3522:2021
Moisture Content (%)	20.24	Max. 23%
pH	9.96	9-11
Insoluble materials in Ethanol (%)	2.22	Max. 10%
Free Fatty Acids (%)	0	Max. 2.5%
Free Alkali (%)	0.068	Max 0.1%

The pH value is related to the degree of acidity in produced soap [40]. Produced soap with propolis extracts addition in this study was deployed optimum formula had a pH of 9.96 and accordance to the standard. Alkaloids were discovered in propolis extracts and provided alkaline properties then causing the increase of pH in produced soap [25]. It indicated to more addition of extracts would be a higher pH value in soap obtained. Mopangga et al. [32] proved similar results in their study about solid soap formulation with gedi leaves extracts has a higher pH which is linearly affected by the addition of extracts.

Hernani et al. in Murniati et al. [41] explained that insoluble materials in ethanol of produced soap were alkaline salt consisting of carbonates, silicates, phosphates, sulfates, and starch. The analysis results of the optimum formula indicated by the standard, where the maximum insoluble materials are 10%. The number of insoluble materials in ethanol of produced soap was 2.22% and produced soap had no appearance of wisps that would interfere with the soap [42].

Free alkali was no-bound alkali with free fatty acids in produced soap caused by excessive alkali deployed in the soap-making process. Free fatty acids were no-bound fatty acids with alkali during the soap-making process caused by a small amount of alkali addition and or excessive oils deployed [43]. In this study, produced soap had a negative content of free fatty acids and was interpreted as produced soap having excess alkali. However, the results showed that produced soap with propolis extracts addition was still safe to use for the skin which was proven by standard and requirement in SNI 3522:2021 were a maximum amount of 0.1%. The free alkali content in produced soap of this study was 0.068%. Similar results were discovered in another study conducted by Mopangga et al. [32], their formulation deployed gedi leaves extracts for solid soap-making with free alkali content in the range of 0.063% to 0.077%.

Conclusion

The optimum formula for solid soap with propolis extracts addition was the composition consist 1.545 grams of propolis extracts, 13.097 grams of coconut oils, 29.629 grams of palm oils, and 19.0809 grams of olive oils. This formula provided soap with 20.24% of moisture content, 90.41% of foam stability, and 50.67% of antioxidant inhibition. The results indicated the obtained models were quite good because model validation had a value above 95%. Produced soap from optimum formula had several characteristics that accordance with the standard in SNI 3522:2021 consisting of 9.96 of pH, 2.22% of insoluble materials in ethanol, and 0.068% of free alkali content.

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