



## Formulation, Characterization and Optimization of Peel-Off Gel of Soybean Extract as a Face Mask

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### ABSTRACT

A peel-off mask is a type of dosage form that is gently applied onto the skin and peeled off after a few minutes. It is one of the most popular skincare products used to treat facial-related conditions. This study is aimed at designing and optimizing a peel-off facial mask formulation containing 20% w/w of soybean extract by response surface methodology (RSM). The peel-off mask was formulated using different concentrations of polyvinyl alcohol (PVA), ethanol, and carbomer 940. The isoflavonoids content of the soybean extract was determined using spectrophotometric method. Physicochemical properties (homogeneity, colour, odour, and pH) of the peel-off mask formulation were examined. Other parameters investigated include drying time, applicability, film-forming performance, and loading efficiency. The stability of the formulation was also investigated. The results showed that the PVA and ethanol concentrations significantly affected the drying time of the formulation. The PVA concentration and its quadratic term significantly influenced applicability of the formulation. The amount of carbomer in the formulation had the most significant impact on how well the film formed. The optimized formulation is composed of 20% soybean extract, 12.03% PVA, 8.36% ethanol, and 0.52% carbomer. Stability test results show that low temperature ( $5 \pm 1^\circ\text{C}$ ) and room temperature, protected from sunlight ( $22 \pm 2^\circ\text{C}$ ) have no effect on the stability of the formulation, and that phenoxyethanol at 1.0% w/w could preserve the formulation from microbial degradation under various storage conditions. Therefore, RSM was a helpful way to figure out the best conditions for making soybean peel-off facial masks.

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**Keywords:** Extraction, Optimization, Peel-Off Gel, Face Mask, Soybean Extract.

### Introduction

Flavonoids are a large group of polyphenolic compounds with a ubiquitous presence in medicinal plants. The biosynthesis of flavonoids is through the phenylpropanoid pathway. Research evidence shows that phenolic-based secondary metabolites, such as flavonoids, contribute to different pharmacological functions.<sup>1</sup> Isoflavones, a significant group of phytoestrogens, can be defined as polyphenolic non-steroidal compounds with estrogen-like biological actions.<sup>2</sup> Polyphenols have been known for their potent antioxidant and anti-inflammatory properties. One of the plants rich in polyphenol antioxidants is soybean (*Glycine max* (L.) Merr.).<sup>3</sup> Studies have shown that soy extract strengthens the epidermal layer of the skin, it increases collagen production, reduces hyperpigmentation, and stops hair growth. Therapeutically, soy has mild anti-inflammatory, moisturizing, light protecting, brightening, and skin-lightening effects.<sup>4</sup> A peel-off mask is one of the types of dosage forms that is gently applied onto the skin and peeled off after a few minutes. The gel form peel-off mask is one of the most popular skincare products used as a therapy for facial-related conditions.

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This mask creates an elastic film on the face, easily removed from the skin after application, leaving nothing behind and makes the skin looks clean due to its firming and tensor properties. Due to its ability to increase the skin's contact time with the active substances, peel-off mask can improve the effectiveness of active substances on the epithelium and increase the skin's moisture due to the occlusive properties of the polymer.<sup>5-8</sup>

In this study, peel-off masks were designed by response surface methodology (RSM) based on a central composite design (CCD) which is aimed at analyzing various parameters related to the essential properties of the peel-off masks, thereby optimizing the final formula. CCD can simultaneously determine the interactive and quadratic effects of different variables that influence product responses.<sup>9</sup> Additionally, CCD has been successfully used in several studies for formulation development and optimization, as data obtained using CCD showed good and reliable predictions.<sup>10-12</sup>

With these considerations, this study proposes the design and testing for the first time, an optimized peel-off gel of soybean extract as a face mask aimed at improving the applicability of soybean extract and its therapeutic properties. Using this extract in a peel-off formulation creates an innovative and acceptable product with strong commercial appeal.

### Materials and Methods

#### Materials

All chemicals used were of analytical grade and were commercially available. These include: kaolin (Merck, Germany), ethanol 96% (EtOH) (Golriz, Iran), polyvinyl alcohol (PVA) 100%, fully hydrolyzed (Mw approx. 30000) (Merck, Germany), carbomer 940 100% (KBR, India), phenoxyethanol  $\geq 99\%$  (Merck, Germany), EDTA disodium 100% (Sigma – Aldrich, Germany), propylene glycol 100% (Merck,

Germany), sodium hydroxide 100% (Sigma – Aldrich, Germany), butylated hydroxytoluene (BHT) ≥ 99% (Merck, Germany), butylated hydroxyanisole (BHA) ≥ 98% (Sigma – Aldrich, Germany), Aluminum chloride 100% (Merck, Germany), sodium nitrate ≥ 99.5% (Sigma – Aldrich, Germany). The purified water (PW) used for the preparation of solutions was purified by reverse osmosis.

#### Methods

##### Collection and Identification of Plant material

The seeds of *Glycine max* (L.) Merr. (soybean) were collected in May 2021, and identified by the Department of Pharmacognosy, School of Pharmacy, Alborz University of Medical Sciences, Karaj, Iran. An herbarium specimen was deposited in the herbarium of Alborz Pharmacy School, and a voucher number 158 was issued.

##### Extraction of Plant Material

Dried soybeans powder was macerated with ethanol (80% v/v) at room temperature at a solvent to powder ratio of 4:1.<sup>13</sup> The maceration was done for three days in a tightly closed container protected from light with constant stirring. This process was repeated four times. The extracts were combined, and the solvent was removed by evaporation at 50°C under vacuum using a rotary evaporator (RE-5PRO Labfreez, China) to obtain the dried extract. The extract was kept in a freezer (Jal Tajhiz Labtech, Iran) at -20°C until ready for use.

##### Determination of the Extraction Yield

The extraction yield (EY) of the dry extract was obtained by the weight method (gram of dry extract in 100 g of dry sample) using the formula below. In this study, the EY was about 17%.

$$\% EY = \frac{C_1}{C} \times 100 \quad (1)$$

Where % EY is the extraction yield percent, C<sub>1</sub> represents the dry extract weight, and C is the initial dry sample weight.

##### Preparation of Peel-Off Masks

The composition of the formulations is shown in Table 1. PVA was dispersed by first heating in 80% of the total amount of water needed, and then carbomer 940 was dissolved in 10% of the water. EDTA disodium was added to propylene glycol and the remaining water (10%), followed by the soybean extract, and mixed with constant stirring. Finally, kaolin and ethanol were added, and the solution was mixed using an ultra-homogenizer (M TOPO, BL1010D, Lab stirrer) at 300 rpm for 20 min. After cooling, phenoxyethanol, BHT, and BHA were added to the mixture. Research has shown that the optimum viscosity for carbomer 940 occurs at pH~ 6-7, and the pH of the skin for most individuals is in the acidic range.<sup>14</sup> For this reason, the pH for the formulations was adjusted to 6.5 by the addition of triethanolamine. Each formulation was left to stand for about 24 h prior to measurement.

##### Determination of Isoflavonoids Content of Soybeans Extract

Quantitative analysis of the dried alcoholic extract of soybeans was done using a UV-VIS spectrophotometer (Hewlett Packard 8453, USA). Standardized soybean extract with isoflavonoids was used as the standard. For the preparation of calibration curve, the standard solutions were made at different concentrations (2 mg/mL, 1.2 mg/mL, 0.6 mg/mL, 0.3 mg/mL, and 0.24 mg/mL) in ethanol (96%). About 1 mL of each standard solution was added to a beaker containing 4 mL of water. Then 0.3 mL of 5% NaNO<sub>3</sub> was added, after 5 min, 0.3 mL of 10% AlCl<sub>3</sub> was added. Following a 5-minute interval, 2 mL of 1 M NaOH was added to the solution and diluted with purified water to 10 mL in a volumetric flask. The absorbance of the solution was measured at 261 nm.<sup>15</sup> The regression equation obtained from the calibration curve was used for the quantitative determination of isoflavonoids in the soybeans extract.

##### Determination of the Drying Time

For drying time, an *in vitro* technique was designed to calculate the time required in each formulation for complete drying.<sup>16</sup> About 1.0 g of peel-off gel mask for each formulation was spread on a 50 mm×50 mm area of the arm's skin, and a uniform mask layer with a nearly 0.12 mm

thickness appeared. The thickness was measured by a digital Vernier caliper (Mitutoyo 500-197-30, Japan). The time the gel formed a film layer, dried entirely and quickly peeled off was determined by using a stopwatch. Ideally, the peel-off face mask needs 15–30 minutes to dry completely.

##### Applicability Assessment

A sensorial score was considered to analyze the applicability of individual formulations. Scoring was done on the model designed by Beringsh and colleagues so that a thin layer of peel-off gel mask was put on see-through glass and how it felt on the skin was rated on a scale of 0 to 5.<sup>12</sup> The feelings assessed include; "Is it easy to spread?" "Does it feel nice?" "Can it be easily applied?" "Does it look good?" and "Does it stay on top?" The formulation is given a score of one point for every positive answer. Higher scores represent better formulation applicability. The scores allocated by the five volunteers were averaged as the total score of each formulation.

##### Determination of Film-Forming Performance

The film-forming analysis was done following the model designed by Beringsh and colleagues. Briefly, about 2.0 g of each formulation was placed on a 120 mm×120 mm glass plate, to make a uniform 13.8 mg/cm<sup>2</sup> and nearly 1.0 mm thick mask layer.<sup>12</sup> The glass plate was put in the oven at 35.0 ± 2.0°C to make it feel like a skin. The sensorial score ranged from 0 to 5 in the following form: "Does the film form?" "Does the film generated have homogeneity?" "Is half of the surface (50%) covered?" "Is most of the surface (80%) covered?" and "Is the whole surface (100%) covered?" The scores were added up, allowing for internal comparison throughout the experimental design. The analyzed formulation is given a score of one point for every positive response; hence, higher scores represented a better film-forming performance for the formulation. The overall value was calculated from the average scores of five observers.

##### Determination of Loading Efficiency

The practical loading efficiency was calculated using the equation obtained from isoflavonoids analysis. Samples were diluted in triplicate by dissolving 0.03 g of the formulation in the same solvent that was used for the extract analysis, and the volume was made up to 10 mL and then centrifuged (Rotina 380, Hettich, Germany) at 3,000 rpm for 15 min. The supernatant was examined using a UV-VIS spectrophotometer at a wavelength of 261 nm. The loading efficiency was calculated using the equation below.

$$\text{Drug Loading Efficiency (\%)} = \left( \frac{\text{Weight of the isoflavonoids inside the gel}}{\text{Total weigh of the gel}} \right) \times 100 \quad (2)$$

**Table 1:** Formula for Soybean Extract Peel-Off Facial Mask Formulation

Ingredient	Percent (w/w)
Soybean extract	20
Polyvinyl alcohol (PVA)	2.5–17.5
Ethanol 96%	0–12.0
Propylene glycol	6.0
Kaolin	2.5
Phenoxyethanol	0.1
EDTA disodium	0.1
Carbomer 940 (Carbopol)	0–2.4
BHT	0.25
BHA	0.25
Triethanolamine	pH 6.5
Purified Water	Up to 100

### Colour, Homogeneity and Odour Analysis

The color and homogeneity were checked visually. The odor was assessed by smelling the product.

### pH

A 1% solution was prepared, and the pH was tested using a pH meter.

### Physicochemical Stability Testing

Three identical samples were stored in non-shiny polyethylene tubes under specific conditions to see how they would stand over time at: (1) low temperatures ( $5.0 \pm 3.0^\circ\text{C}$ ), (2) oven ( $45.0 \pm 2.0^\circ\text{C}$ ), (3) room temperature while exposed to sunlight ( $22.0 \pm 5.0^\circ\text{C}$ ), and (4) room temperature with no exposure to sunlight ( $22.0 \pm 2.0^\circ\text{C}$ ).<sup>16</sup> Daily examinations of the individual formulation with respect to organoleptic parameters (odour, colour), applicability, film-forming performance, drying time, pH, and sedimentation were performed for 28 days.

### Microbiological Stability Testing

Triplicate samples of the optimized formulation containing 1.0% phenoxyethanol were prepared to estimate the microbiological stability. The microbial examination was performed using a direct plate count method.<sup>17</sup> The initial microbial load in the formulation was determined right after the mixture was made (time 0). Three dilutions ( $10^{-1}$ ,  $10^{-2}$ , and  $10^{-3}$ ) of each formulation were prepared. Then, the dilutions were incubated in Petri dishes (INCUBE-120 STD, Kavosh, Iran) with special culture growth media for bacteria and fungi. Dilution of every formulation was done using phosphate buffer at pH 7.2. After the initial microbial load was determined, each formulation was placed in polyethylene tubes and kept under the storage conditions below:

$5.0 \pm 1.0^\circ\text{C}$

$45.0 \pm 2.0^\circ\text{C}$

$22.0 \pm 5.0^\circ\text{C}$  while exposed to sunlight

$22.0 \pm 2.0^\circ\text{C}$  with no exposure to sunlight

As mentioned earlier, the direct plate count method was used to check the number of microbes in each formulation after storing them under different conditions for 28 days.<sup>17</sup>

All diluted samples were placed in pairs in 20 mL of the correct culture medium (casein-soya and Sabouraud-dextrose agar for bacteria and fungi, respectively) on Petri dishes, followed by incubation for 72 h (bacteria) or 7 days (fungi). The average number of colony-forming units per gram (CFU/g) was determined.

### Statistical Analysis

The response surface methodology (RSM) design was used to determine the most important factors affecting how well a soybean extract peel-off facial mask works.<sup>9</sup>

A central composite design, with extra points, was made to find the square terms. The loud noise was reduced by doing four middle interactions, giving the right amount of freedom to accurately assess the model using statistics. Screening was done to determine how much of PVA, carbomer 940, and EtOH are needed in the film formation. The design that was used include alpha;  $\alpha = \pm 1.5$  and the variables studied are listed in Table 2. The study also examined the responses (dependent variables) related to drying time, applicability, and film-forming performance. A statistical software program (Design-Expert® version 7.0.0, Stat Ease, Inc, MN, USA) was used to perform regression analysis of the data, assuming a quadratic model within which the variables interacted.

The analysis of variance (ANOVA) helped to understand the interactions between different variables, and how the squared terms affected the responses studied. The probability value ( $p < 0.05$ ) represented the level of significant difference between the variables. The non-significant terms were removed from the model unless required as critical terms for a significant interaction. The polynomial model concerning the coded variables is as follows:

$$\eta = \beta_0 + \beta_1x_1 + \beta_2x_2 + \beta_3x_3 + \beta_{12}x_1x_2 + \beta_{13}x_1x_3 + \beta_{23}x_2x_3 + \beta_{11}x_1^2 + \beta_{22}x_2^2 + \beta_{33}x_3^2 \quad (3)$$

Where  $\eta$  represents the dependent response related to a single independent variable level combination;  $\beta_0$  represents the mean value, and  $x_1$ ,  $x_2$ , and  $x_3$  denote the respective symbols for the independent variables EtOH, PVA, and carbomer 940;  $x_1x_2$ ,  $x_1x_3$ , and  $x_2x_3$  indicate the binary interactions within the variables;  $x_{12}$ ,  $x_{22}$ , and  $x_{32}$  represent the quadratic factors. The coefficients of the main, interaction, and quadratic factors are indicated as  $\beta_1$ ,  $\beta_2$ ,  $\beta_3$ ;  $\beta_{12}$ ,  $\beta_{13}$ ,  $\beta_{23}$ ; and  $\beta_{11}$ ,  $\beta_{22}$ ,  $\beta_{33}$ , respectively.

The math model was used to make the formulation with the highest applicability and film-forming performance and the lowest drying time. RSM was used to obtain the optimal parameters, while the desirability function and experiments confirmed the predicted model's validity in optimum conditions. The mentioned function performs as a multiple response technique in which the calculation of the objective function ranges between zero (out of the limits) and one (at the goal). Using the desirability function, one can combine an appropriate series of conditions and obtain the optimum experimental condition satisfying all goals. The geometric mean of the whole transformed response is represented in the simultaneous objective function:

$$D = (d_1 \times d_2 \times \dots \times d_n)^{1/n} = (\prod_{i=1}^n d_i)^{1/n} \quad (4)$$

Where  $D$  = desirability function,  $d_i$  = the desirable range concerning single responses, and  $n$  = number of responses within the estimations.<sup>9</sup>

**Table 2:** Variables Used in Central Composite Design

Levels	PVA (%)	EtOH (%)	Carbomer 940 (%)
Minus alpha ( $\alpha = -1.5$ )	2.5	0	0
Low	5	2	0.4
Central point	10	6	1.2
high	15	10	2
Plus alpha ( $\alpha = +1.5$ )	17.5	12	2.4

## Results and Discussion

### Extraction Yield

The extraction yield obtained in this study was about 17%.

### Soybeans Extract Isoflavonoids Content

The  $\lambda_{\text{max}}$  of isoflavonoids in the extract was at 261 nm, which was the same as the  $\lambda_{\text{max}}$  of standardized samples of soybean extract. The analyte showed good linearity in the 24–200  $\mu\text{g/mL}$  concentration range. The regression equation of the calibration curve is  $Y = 14488x - 0.0503$  with  $R^2$  value of 0.9983. The content of isoflavonoids in the extract was calculated as 2.85%.

### Results of the Central Composite Design and Statistical Outcomes of the Experimental Design

Table 3 shows the values for the response variables under various experimental conditions, while Table 4 shows the statistical outcomes of the response variable. A, B, AB, A, A<sup>2</sup>, B<sup>2</sup>, and C<sup>2</sup> are significant model terms for drying time. A, C, A<sup>2</sup>, and C<sup>2</sup> are significant model terms for applicability, while C and A<sup>2</sup> are significant model terms for film-forming. The best model using the adjusted R<sup>2</sup> and predicted R<sup>2</sup> coefficients was the quadratic model for all the results. Acceptable precisions of 6.665 (applicability), 24.064 (drying time), and 10.378 (film-forming performance) indicated a good balance between the signal and the background noise (adequate precision >4.0).

#### Loading Efficiency

The value of  $\geq 81\%$  was obtained for the soybean extract loading efficiency in all the formulations. No formulation parameter had significant effects on the loading efficiency.

#### Influence of the Variables on the Drying Time

The variables that considerably impacted the drying time were the PVA and ethanol. PVA had a negative effect (coefficient = - 5.64) on the drying time, indicating that an increasing PVA concentration (A) decreases the drying time (Table 4). This could be because, with increased polymer concentration, the water content in the formulation decreases. On the other hand, low concentration of the polymer did not influence the diffusion coefficient of the water from the film. Subsequently, the time required for drying will be reduced.<sup>18</sup> This is in agreement with previous reports where increasing the PVA concentration shortened the film's drying time.<sup>19</sup> Besides, the PVA quadratic term ( $A^2$ ) affected the drying time positively ( $p = 0.0005$ ) (Table 4), indicating a nonlinear association, coming to a plateau at nearly 12% concentration (Figure 1A) indicating that an excess of over

12% of the PVA will not be recommended. This effect could be due to the diffusion coefficient sharply decreasing at high concentrations of polymers, leading to the trapping of water molecules between polymer chains so that the water will not evaporate quickly.<sup>18</sup> This observation supports the research of Apriani and colleagues, where PVA was shown to interact with water, absorbs the liquid and forms a thick mass that causes the liquid to be trapped and difficult to evaporate, leading to a prolonged or increased drying time of the formula.<sup>20</sup>

The ethanol variable also negatively influenced (coefficient = - 6.84) the drying time (Table 4). This is because ethanol helps things dry faster, and it evaporates more quickly than water.<sup>21</sup> Like PVA, the quadratic term for ethanol ( $B^2$ ) had a significant effect on the drying time of the film ( $p < 0.0001$ ) (Table 4) and indicated a nonlinear association that reached a plateau at nearly 8% concentration (Figure 1A). Consequently, adding more than 8% (w/w) of ethanol cannot be recommended as the drying time reduction is no longer proportional. This agrees with previous reports where adding purified water/EtOH in amount more than that recommended for drying time in the formulation was no longer proportional.<sup>22</sup>

**Table 3: Results of Central Composite Design**

Experiment (Run)	Factors			Responses		
	A: PVA (% w/w)	B: EtOH (% w/w)	C: carbomer (% w/w)	Drying time (min)	Applicability	Film forming
1	2.50	6.00	1.20	40	1	0
2	5.00	10.00	0.40	35	2	5
3	10.00	6.00	2.40	30	1	2
4	17.50	6.00	1.20	25	4	3
5	15.00	10.00	2.00	20	2	2
6	15.00	10.00	0.40	21	5	4
7	5.00	2.00	2.00	49	1	2
8	10.00	6.00	1.20	25	5	4
9	10.00	6.00	1.20	24	5	4
10	15.00	2.00	0.40	35	5	5
11	10.00	6.00	1.20	28	5	4
12	10.00	6.00	1.20	35	5	4
13	10.00	6.00	0.00	30	4	5
14	10.00	6.00	1.20	35	5	4
15	5.00	2.00	0.40	43	4	4
16	5.00	10.00	2.00	36	4	4
17	15.00	2.00	2.00	39	3	2
18	10.00	0.00	1.20	51	3	3
19	10.00	12.00	1.20	30	5	4

#### Influence of the Variables on the Film-Forming Performance

Carbomer concentrations were the most influential on the peel-off facial mask film-forming performance ( $p = 0.0005$ ) (Figure 1B). It had a linear relationship since increasing the carbomer concentration decreased the film-forming performance. The PVA quadratic term ( $A^2$ ) affected film-forming performance ( $p = - 0.81$ ) (Table 4), indicating a nonlinear association and showing that PVA of a specific concentration is suggested for film-forming performance, which is in agreement with previous reports.<sup>22</sup>

#### Influence of the Variables on the Applicability of the Formulation

One of the significant formulation characteristics associated with applicability is viscosity. The PVA concentration exerted a positive influence (coefficient = 0.68) (Table 4) on viscosity of the formulation, indicating an enhanced applicability by increasing the PVA concentration. The PVA quadratic term ( $A^2$ ) affected the applicability

( $p = - 0.86$ ) (Table 4), indicating a nonlinear association, which reached a plateau at nearly 12% concentration (Figure 1C) which indicated that the addition of over 12% of the PVA would not be recommended. PVA will also reduce the responsiveness of spreadability. The higher the concentration of PVA used, the more the viscosity will increase due to hydrogen bonding occurring between the OH group of PVA, and the water used in the composition.<sup>20</sup> This finding supports the research of Asthana and colleagues that shows that increasing or decreasing the PVA concentration is not beneficial for the uniformity and thickness of the PVA film.<sup>22</sup>

How fast the material spreads depend on how much force is applied, for how long, and how thick the material is. But, when the topical preparation is thicker, it does not spread quickly.<sup>19</sup> From the results of the applicability test of the peel-off mask gel, it was also noted that an increase in carbomer concentration affected the gel applicability. The carbomer concentration (C) and the quadratic term for it ( $C^2$ ) were

significant (a negative influence) (Table 4), indicating a nonlinear association (Figure 1D). these observations suggest that adding over 0.5% of carbomer may lead to a decrease in applicability.

#### Optimized Peel-off Mask Formulation

Combining the adjusted and predicted  $R^2$  values (Table 4; difference < 0.2) from the mathematical models derived from RSM allows for the prediction of the best formulation for a soybean extract peel-off mask according to predetermined measurements. Numerical optimizations resulted in the point maximizing the best function. Adjustment of the weight and significance may change the properties of a goal. Combining all goals into one desirability function for several responses and factors is possible. The desirability function approach is a popular way to optimize multiple responses simultaneously. The desired level ranges from 0 to 1 and represents how close the response is to its ideal value. Creating these standards maximizes the applicability and film formation while minimizing drying time. The experimental conditions of the study enabled the design of the optimized formulation with 12.03% (w/w) of PVA, 8.36% (w/w) of EtOH, and 0.52% (w/w) of carbomer 940 (Figure 2; Table 5). The

formulation's combined desirability function equaled 0.976. At the same time, the values of 21.3 min, 5, and 4.7 were reported for the drying time, applicability, and performance of film-forming predicted for the developed models, respectively. Each factor had a desirability value of 1.0000 for EtOH, PVA, and carbomer. This means that the model is good at meeting all the needs for optimization. For every response, the values of desirability for the drying time, applicability, and film-forming performance were 0.9569, 0.9999, and 0.9349, respectively, indicating the model's capability to optimize all the responses promptly until reaching an acceptable stage considering the requirements (suggesting individual desirability of > 0.9).

The confirmation runs (n = 3) were prepared to exert the obtained mathematical conditions experimentally, leading to outcome of the confirmation experiments within 95% prediction (PI) and 95% confidence (CI; Table 5) intervals, confirming the model's good predictability for all responses and the developed model's significant robustness for the responses found in the RSM design.

The optimized formula looked even and consistent before and after it dried. The soybean extract that could be loaded into the new mixture was found to be at least 81%.

**Table 4:** Significant Model Terms, Regression Coefficient Values, and ANOVA (P Values) for the Experimental Design Responses

Polynomial term	Drying time (quadratic model)		Applicability (quadratic model)		Film forming (quadratic model)	
	Coefficient	p-value	Coefficient	p-value	Coefficient	p-value
Model		<0.0001		0.0012		0.0002
Intercept	24.86		4.77		3.96	
A: PVA	- 5.64	<0.0001	0.68	0.0285		
B: EtOH	- 6.84	<0.0001				
C: Carbomer			- 0.84	0.0080	- 1.0	0.0005
AB*	- 1.50	0.0430				
AC*						
BC*						
A <sup>2</sup> *	2.75	0.0005	- 0.86	0.0126	- 0.81	0.0058
B <sup>2</sup> *	6.31	<0.0001				
C <sup>2</sup> *	1.64	0.0157	- 0.86	0.0126		
R <sup>2</sup>	0.9733		0.6794		0.6236	
Adjusted R <sup>2</sup>	0.9577		0.5939		0.5794	
Predicted R <sup>2</sup>	0.8911		0.4168		0.3629	
Adequate precision	24.064		6.655		10.378	

\*AB: Interaction between PVA and EtOH; AC: Interaction between PVA and carbomer; BC: Interaction between EtOH and carbomer; A<sup>2</sup>: Quadratic term of PVA; B<sup>2</sup>: Quadratic term of EtOH; C<sup>2</sup>: Quadratic term of carbomer.

#### Physicochemical Stability of the Soybean Peel-off Mask Formulation

Physicochemical parameters were monitored weekly to confirm formulation stability in various conditions. Table 6 shows the results of the physicochemical stability testing of the formulation. The optimized formulations kept at low temperature ( $5 \pm 1^\circ\text{C}$ ) and room temperatures, with no exposure to sunlight ( $22 \pm 2^\circ\text{C}$ ) and room temperature with exposed to sunlight ( $22 \pm 5^\circ\text{C}$ ) were not unstable with respect to the evaluated parameters. An alteration in the odour occurred in the formulation exposed to sunlight in the fourth week, probably indicating oxidation of the oily part of the soybean extract. The formulations were unstable under extreme temperatures (oven;  $45 \pm 2^\circ\text{C}$ ). There was a considerable decrease in the drying time after 28 days, probably due to the increase in the temperature of the formulation and the faster evaporation of ethanol after opening the lid of the tubes and using it. Following storage, the density of the formulations increased for facial applications, indicated by its reduced applicability from 5 to 4 (Table 6). After 28 days, there was evidence of significant suspended particle sedimentation since the increase in the temperature resulted in a short-term reduction of the viscosity of the PVA-based formulation while accelerating the insoluble particles' normal sedimentation process,

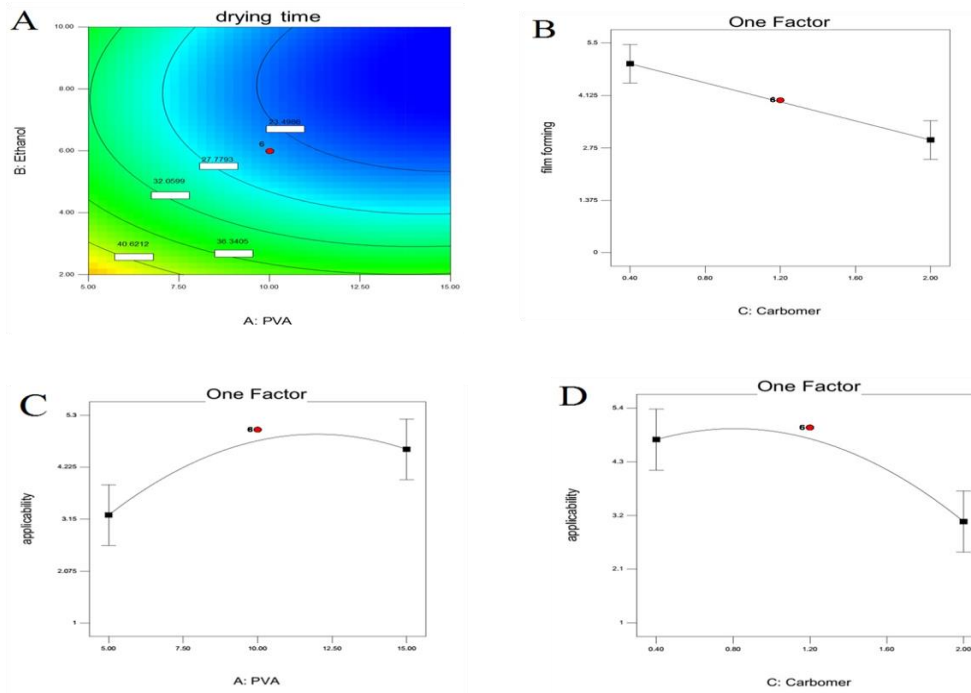
including Kaolin. These formulations get darker when the oxidation process occurs.

Storage conditions should be carefully chosen to avoid stability problems. The results indicated that polyethylene tubes with tight sealing are recommended for this type of formulation.

#### Microbiological Stability

From the result of the microbiological tests, all the samples, including the ones stored for 28 days, showed no bacterial or fungal growth. The number of microbes in the samples was less than  $1 \times 10^1$  CFU/g.

This study evaluated the formulations' eventual microbial load following the production and 28 days storage under extreme conditions. As shown by the results, the concentration of the phenoxyethanol 1.0% w/w was able to preserve the optimized formulation from microbial degradation under various storage conditions. According to international applied standards, the formulations' eventual microbial load was satisfactory for facial applications.



**Figure 1:** Contour graphics of (A) drying time, (B) film-forming performance, (C) and (D) applicability

**Table 5:** The Optimized Formulation's Point Prediction Information

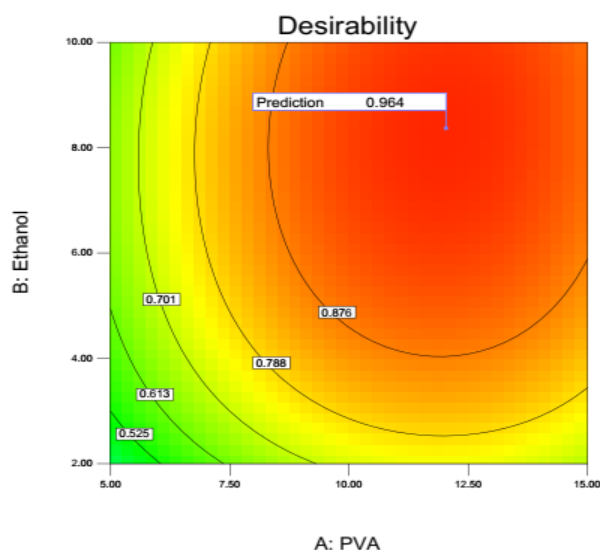
	Factors			Responses		
	A: PVA (% w/w)	B: EtOH (% w/w)	C: Carbomer (% w/w)	Drying time (min)	Applicability	Film-forming
Predicted formulation	12.03	8.36	0.52	21.3	5	4.7
Confirmation runs (n=3)	12.03	8.36	0.52	23	5	4
				23	5	4
				24	5	4
Individual desirability	1	1	1	0.957	0.999	0.935
Combined desirability	0.964					
95% confident interval (CI)				19.58- 23.09	4.22- 5.78	4.05- 5.30
95% Prediction interval (PI)				16.89-25.78	2.79- 7.21	2.82- 6.53

**Table 6:** Physicochemical Stability Results of the Optimized Formulation

Parameter	Time (week)				1st	2nd	3rd	4th
	1st	2nd	3rd	4th				
	<b>22 ± 2°C - Protected from sunlight</b>				<b>22 ± 5°C - Exposed to sunlight</b>			
Colour	N*	N	N	N	N	N	N	N
Odour	N	N	N	N	N	N	N	↑
Applicability	5 ± 0	5 ± 0	5 ± 0	5 ± 0	5 ± 0	5 ± 0	5 ± 0	5 ± 0
Drying time	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58
Film-forming	4 ± 0	4 ± 0	4 ± 0	4 ± 0	4 ± 0	4 ± 0	4 ± 0	4 ± 0
pH	6.15 ± 0.08	6.14 ± 0.07	6.12 ± 0.08	6.02 ± 0.03	6.13 ± 0.08	6.11 ± 0.09	6.00±0.00	6.00 ± 0.00
Sedimentation	NS*	NS	NS	NS	NS	NS	NS	NS
	<b>5 ± 3°C - Refrigerator</b>				<b>45 ± 2°C - Oven</b>			
Colour	N	N	N	N	N	N	↑	↑↑
Odour	N	N	N	N	N	N	↑	↑↑
Applicability	5 ± 0	5 ± 0	5 ± 0	5 ± 0	5 ± 0	4 ± 0	4 ± 0	4 ± 0
Drying time	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58	23.3 ± 0.58	22.3 ± 1.15	20.3±0.58	18.3 ± 1.53	17.0 ± 1.00

Film forming	4 ± 0	4 ± 0	4 ± 0	4 ± 0	4 ± 0	5 ± 0	5 ± 0	5 ± 0
pH	6.15 ± 0.08	6.15 ± 0.08	6.14 ± 0.08	6.10 ± 0.09	6.12 ± 0.11	6.00 ± 0.00	5.83 ± 0.06	5.46 ± 0.06
Sedimentation	NS	NS	NS	NS	NS	MIS*	MIS	MAS*

\*N: normal, NS: no sedimentation, MIS: minor sedimentation, MAS: major sedimentation



**Figure 2:** Contour graphic of desirability of the optimized peel-off mask

## Conclusion

This study investigated the parameters that have the most significant effect on the desirable characteristics of soybean peel-off mask. It was observed that the drying time was significantly affected by the concentration of ethanol. The amount of carbomer and PVA in the mask can influence the applicability of the peel-off mask because they can make it thicker or thinner. Carbomer concentration only affected the film formation performance. Suitable concentrations to achieve an optimal formula that had the highest levels of applicability and performance of film forming and the shortest drying time was calculated as follows: 12.03% (w/w) of PVA, 8.36% (w/w) of EtOH, and 0.52% (w/w) of carbomer 940. The initial stability study showed that the optimized formula was stable under regular storage conditions. The preservative used in this formula worked well in preventing germs from growing under the tested storage conditions. Microbial load was less than  $1 \times 10^1$  CFU/g for fungi and bacteria. It was discovered that the RSM based on CCD is a helpful way to figure out how different chemicals work together and their amounts based on the parameter being studied. This helped find the best conditions for making soy extract peel-off masks, but one of the limitations of this method is the number of experiments. It is recommended for future studies that data analysis and optimization of operating parameters be performed using a tool with minimal testing and less computerization, and as a result, the chart is easy to read and understand.

## Conflict of Interest

The authors declare no conflict of interest.

## Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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## References

- Innocent E. A New Isoflavone Glycoside from *Dalbergia vacciniifolia* (Fabaceae). *Sci Pharm*. 2012; 80(2):469-474.
- Křížová L, Dadáková K, Kašparovská J, Kašparovský T. Isoflavones. *Molecules*. 2019; 24(6):1076.
- Ganesan K and Xu B. A Critical Review on Polyphenols and Health Benefits of Black Soybeans. *Nutrients*. 2017; 9(5):455.
- Natarelli N, Gahoonia N, Maloh J, Sivamani RK. Clinical Efficacy of Topical or Oral Soy Supplementation in Dermatology: A Systematic Review. *J Clin Med*. 2023; 12(12):4171.
- Hawaisa S, Rosita N, Soeratri W. Effects of Polyvinyl Alcohol and Hydroxypropyl Methylcellulose Combination on Physical Stability and Irritability of Glutathione Peel-Off Masks. *Trop J Nat Prod Res*. 2023; 7(11):5081–5086.
- Hariyadi DM, Isnaeni I, Sudarma S, Shandra NMK, Rosita N. Formulation of Peel-Off Mask Containing Natural Antibacterial: Study on Poly Vinyl Alcohol (PVA) and Virgin Coconut Oil (VCO) Contents. *TJNPR*. 2021; 5(3):514–518.
- Mohammadzadeh M, Hashemi S, Salmannejad F, Ghari T. Identification of Key Success Factors in the Marketing of Cosmetics Based on Knowledge, Attitude and Practice (KAP) analysis Using Topsis Technique (The Case of Iran). *J Pharm Sci*. 2017; 23(3): 222-230.
- Baby AR, Zague V, Maciel CPM, Salgado-Santos IMN, Kawano Y, Arêas EPG, Kaneko TM, Consiglieri VO, Velasco MVR. Development of Cosmetic Mask Formulations. *Semana Farmacêutica de Ciência e Tecnologia da FCF-USP*. 2004; 40:159-61.
- Myers RH, Montgomery DC, Anderson-Cook CM. *Response Surface Methodology: Process and Product Optimization Using Designed Experiments*. (4rd ed.). New Jersey: John Wiley & Sons; 2016; 856 p.
- Savic Gajic IM, Savic IM, Gajic DG, Dasic A. Ultrasound-Assisted Extraction of Carotenoids from Orange Peel Using Olive Oil and Its Encapsulation in Ca-Alginate Beads. *Biomol*. 2021; 11(2):225.
- Savic Gajic I, Savic I, Boskov I, Žerajić S, Markovic I, Gajic D. Optimization of Ultrasound-Assisted Extraction of Phenolic Compounds from Black Locust (*Robinia Pseudoacaciae*) Flowers and Comparison with Conventional Methods. *Antioxidants (Basel)*. 2019; 8(8):248.
- O'Reilly Beringsh A, Rosa JM, Stulzer HK, Budal RM, Sonaglio D. Green Clay and Aloe Vera Peel-Off Facial Masks: Response Surface Methodology Applied to the Formulation Design. *AAPS PharmSci Tech*. 2013; 14(1):445-455.
- Tzanova M, Atanasov V, Yaneva Z, Ivanova D, Dinev T. Selectivity of Current Extraction Techniques for Flavonoids from Plant Materials. *Proc*. 2020; 8(10):1222.

14. Kolman M, Smith C, Chakrabarty D, Amin S. Rheological Stability of Carbomer in Hydroalcoholic Gels: Influence of Alcohol Type. *Int J Cosmet Sci.* 2021; 43(6):748-63.
15. Guemari F, Laouini SE, Rebiai A, Bouafia A, Meneceur S, Tliba A, Majrashi KA, Alshareef SA, Mena F, Barhoum A. UV-Visible Spectroscopic Technique-Data Mining Tool as a Reliable, Fast, and Cost-Effective Method for the Prediction of Total Polyphenol Contents: Validation in a Bunch of Medicinal Plant Extracts. *Appl Sci.* 2022; 12:9430.
16. Vieira RP, Fernandes AR, Kaneko TM, Consiglieri VO, Pinto CASDO, Pereira CSC, Baby AR, Velasco MVR. Physical and Physicochemical Stability Evaluation of Cosmetic Formulations Containing Soybean Extract Fermented by *Bifidobacterium animalis*. *Braz J Pharm Sci.* 2009; 45(3):515-525.
17. The United States Pharmacopeia 41, the National Formulary 36. In: USP/NF, editor. Baltimore, MD: United Book Press; 2018.
18. Talini L and Lequeux F. Formation of Glassy Skins in Drying Polymer Solutions: Approximate Analytical Solutions. *Soft Matter.* 2023; 30(19):5835-5845.
19. Rahmasari D, Ermawati D, Nugraheni RW, Putri DR, Pratiwi IN. Design and Development of Peel-Off Mask Gel Formulation of Citronella Oil for Acne Vulgaris. *HSIC.* 2019; 157-163.
20. Apriani EF, Miksusanti M, Fransiska, N. Formulation and Optimization Peel-Off Gel Mask with Polyvinyl Alcohol and Gelatin Based Using Factorial Design from Banana Peel Flour (*Musa paradisiaca* L) As Antioxidant . *Indones J Pharm.* 2022; 33(2):261–268.
21. Yagishita M, Kölling R, Einfalt D. Introducing a Simple Method to Investigate Relative Volatilities of Flavour Compounds in Fruit Brandies. *Beverages.* 2023; 9(2):32.
22. Asthana N, Pal K, Aljabali AAA, Tambuvala MM, Souza FGD, Pandey K. Polyvinyl Alcohol (PVA) Mixed Green-Clay and *Aloe vera* Based Polymeric Membrane Optimization: Peel-Off Mask Formulation for Skin Care Cosmeceuticals in Green Nanotechnology. *J Mol Struct.* 2021; 1229:1-7.