Original Research Article

Lepidium Sativum seed mucilage: Development of a systematic Extraction and Isolation process with maximum yield

ABSTRACT

Objective

The present investigation helps to isolate the seed mucilage of *Lepidium Sativum* by using an economic extraction process with utmost yield. Diligence effort was given to get the maximum yield of the mucilage with good flow properties without affecting the intrinsic properties of the extracted mucilage. The yield of the seed mucilage of *Lepidium Sativum* was optimized by using a quality by design approach. The full factorial design was used using three variables (pH of extract, water to seed ratio, and drying temperature) at two levels (high and low). Overall, ten formulation trials were generated through statistical software Minitab placing 2 center points, 1 replicate, and 1 block. All trials were executed to evaluate the percentage yield.

Results

The extraction process was evaluated with different solvents and techniques to monitor the yield of the process. A factorial design was used to observe the responses like the yield of the isolated mucilage for different levels of all the variables. A polynomial equation was developed and model plots (contour plot and Pareto chart) were generated to study the impact of the critical variables on the response yield. The main and interaction effect of the critical variables were studied to identify the critical variable that is affecting the response yield. Finally, the optimized parameter was established based on statistical evaluation obtained from the polynomial equation as well as the recommendation obtained from the response optimizer. The study indicates that the obtained linear regression equation has a linear relation with a regression coefficient of 0.9869 % for response % yield. Finally, the seed mucilage of *Lepidium Sativum* was extracted by boiling the seeds in water, blending by hand blender, and using acetone as a solvent.

Conclusion

A cost-effective extraction and isolation process was established to extract the seed mucilage of *Lepidium Sativum* with maximum yield. The isolated mucilage was evaluated for flow properties and demonstrated that it is having good flow properties and can be used as an

excipient or adjuvant in different pharmaceuticals and food industries. The applicability of factorial design was well demonstrated to optimize the yield by optimizing the critical factors.

Keywords: Seed mucilage, Isolation, Extraction, Garden cress seeds, *Lepidium Sativum,* maximum yield.

INTRODUCTION

The plant-derived products are now in high demand because they are easily available, cost-effective, and compatible with a wide range of active medicinal ingredients. India is accomplished with ancient traditional plant-derived products for the treatment of various diseases. The different plant-derived products are highly enriched with different phytochemical constituents which can show different therapeutic activities against critical and chronic diseases[1].

Recent investigations on different plant-derived products demonstrated that these naturally obtained products can be used as an alternative to different excipients used in pharmaceuticals and food industries[2.] The majority of the plant-derived products are gums or mucilages in nature. Mucilages are polysaccharide-based compounds and significantly vary in composition [3,4]. *Lepidium Sativum* is a medicinal herb readily available in different parts of India. It is popularly known as halon and is categorized as an edible plant. In India, the *Lepidium Sativum* plant is mainly utilized for seeds, roots, and leaves. However, the plant is recognized for seeds [5]. The seeds of the halon plant are supplemented with different alkaloids which enable its use as an excipient and adjuvant in different pharmaceutical and food preparations [5].

The recent investigation and research on the *Lepidium Sativum* plant mainly focused on the use of the seed mucilage derived from this plant in various medicinal products because of their unique inherent functional related characteristics. The use of seed mucilage of *Lepidium Sativum* is gaining popularity and researchers already explored the use in pharmaceutical formulations with various distinctive roles in tablet dosage forms and liquid dosage forms [5-12]. However, designing innovative medicinal products containing mucilage is becoming extremely prevalent. Moreover, the mucilage of *Lepidium Sativum* is obtained from the seed as a natural source. However, diminutive information is available on how to get the maximum percentage of mucilage from the available seeds. Based on this assurance, the present study focused on the development of a cost-effective isolation and extraction process of mucilage from *Lepidium Sativum* seeds. The current research emphasized the critical attributes that are affecting the yield and utilizes the quality by design (QBD) approach to explore the study. Special attention and focus have been given to optimizing the critical factors and based on the study, recommendations were proposed. Extraordinary effort and consideration were given to discussing the flow properties of the obtained powdered mucilage to enable it to use as an excipient in different pharmaceutical, ayurvedic and food preparations, and formulations.

MATERIALS AND METHODS

Materials

Seeds of *Lepidium Sativum* were procured from the nearest retail store in Mumbai, India. All the chemicals and other reagents such as ethanol, acetone, chloroform, HCl, and NaOH utilized were of analytical quality and were procured from S.D. Fine chemicals, Mumbai, India. Distilled water was produced *in-house* in the laboratory and this water was employed in all the experiments.

Methods

Extraction and Isolation of mucilage from seeds of Lepidium Sativum

The mucilage extracted from the seeds of *Lepidium Sativum* is popularly used as an excipient in different pharmaceutical and food preparations. The mucilages are mainly present around the out layer of seeds of *Lepidium Sativum*. From the literature, it was identified that it is very difficult to extract the mucilage from the seeds as it swells around the seeds but does not separate easily. In literature, different methods were adopted to find out the suitable extraction process utilizing various organic solvents. In our study, we evaluated the literature database by performing the available prior art information by taking small quantities (50 g for each method) of seeds. The main purpose behind this is to understand the feasibility of the extraction process and ease of operation. The different techniques adopted for the extraction of seed mucilage from *Lepidium Sativum* were summarized below.

Table 1: Details of screening trials for extraction and isolation of mucilage from seeds of *Lepidium Sativum*

Sr. No.	Extraction Process	References
Method-1	Precipitation of mucilage in acetone after boiling in water and blending by using a hand blender	[3,12,13]

Method-2	Precipitation of mucilage in Ethanol by using soaked seed	[3,12,13,14,15]
Method-3	Extraction and isolation of mucilage by boiling seeds in water and using ethanol as a solvent	[3,18, 13,14]
Method-4	Extraction and isolation of mucilage by soaking crushed seeds in water and using ethanol as a solvent	[16,17]

From the screening methods, three methods were finally selected for the pilot-scale study. The details of observation and yield of the pilot-scale study were enumerated in the results section. Based on the results of the pilot study the quality by design approach was performed to identify the critical factors and finally a modified method was proposed.

Experimental design for the extraction process to get maximum yield by using full factorial design

A full factorial design was used for the development of a systematic extraction and isolation of Seed mucilage of *Lepidium Sativum* using software Minitab 173.0. Three factors having two levels (+1and -1) of the critical extraction process variables were used, designated as X1: pH of extract (5-10) and X2: Water to seed ratio (10-30), and X3: Drying temperature (40-60˚C) keeping all other extraction processes same. A full factorial design was used to evaluate the influence of critical factors on the response yield. The model was further examined through the obtained polynomial equation and ANOVA study. A total of 10 experimental runs were generated having 2 center points, 8 base designs, 1 replicate, and 1 block. The different levels of variables along with coded and un-coded factors of experimental design are presented in table 2 and table 3. For each trial, the same extraction process was adopted and only the critical factors are changed as per the full factorial design.

Table2: Levels of variables for optimization.

Code	Variables	Level of variables		
		-1	0	+1
X1	pH of extract	5	7.5	10
X2	Water to seed ratio	10	20	30
X3	Drying temperature	40	50	60

Table3: Full factorial design obtained by statistical software Minitab 17.3.0.

Run order	pH of extract	Water to seed ratio	Drying temperature
1	5.00	10	60
2	5.00	30	60
3	10.00	30	60
4	10.00	10	60
5	5.00	10	40
6	10.00	10	40
7	7.50	20	50
8	5.00	30	40
9	10.00	30	40
10	7.50	20	50

A modified method for extraction and isolation of mucilage from seeds of Lepidium Sativum

On the pilot scale, three methods were studied to evaluate the scalability effect as well as % the yield of dried mucilage. Finally, extraction and isolation of mucilage by boiling seeds in water using acetone as a solvent was selected as a final method. This method was selected due to its ease of operation and a higher % of yield. However, based on the design of experiment trials the modified extraction process was proposed. The recommended critical steps as compared to the existing process available in the literature domain are

- Adjustment of pH to 10 by using 0.1 M HCl or NaOH
- Maintaining the water to seed ratio of 30:1
- Inclusion of soaking of seeds before boiling step
- Keep the filtrate in the refrigerator for five hours before the removal of mucilage from the filtrate.
- Optimization of drying temperature

The details of the modified method were enumerated below

Extraction and isolation process (Modified method)

In this method, about 100 g of *Lepidium Sativum* seeds were soaked for overnight at least 12 hours in 3000 ml of distilled water. The pH was adjusted to 10. The soaked seeds were boiled in distilled water for 15 minutes at a temperature of 60°C and kept aside for half an hour to settle down the foam generated during heating. The seeds were then blended by using a hand blender which enables easy removal of the swollen mucilage from the sides of the seeds. Hand blending of seeds was performed for 15 minutes by using a 175-watt Philips HR 3700 hand blender. The mucilage from the seeds was separated by filtering

Comment [Ma1]: The extraction modified method should be in one form, paragraph or diagram not both through a muslin cloth. The retained blended seeds were again blended with additional 200 ml of hot water for 15 minutes by using a 175 watt Philips HR 3700 hand blender. To get the best yield, the blended seeds were re-filtered using a muslin cloth. The filtrate from both the steps was collected in a beaker and treated with an equivalent amount of acetone (1:1 ratio of liquid containing mucilage and acetone). After the addition of an equivalent amount of acetone, the beaker was kept in a deep refrigerator for 5 hours. This helps to get the maximum amount of mucilage from the treated liquid mixture. After 5 hours the liquid was removed and the white supernatant coagulant mass was separated above the beaker. The precipitated mucilage was collected in a petridish and drying was done at 45°C for 6 h in a hot air oven till it was completely dried. A mortar and pestle were used to grind the dried material. Finally, the triturated material was sieved using an 80 # mesh and properly weighed to determine the yield. The finalized powder is stored in a desiccator until it is further used.

Comment [Ma2]: Diagram of the procedures in Fig 1 not mentioned

Note: The following precautions were taken to maximize the yield-

- Time for soaking of seeds fixed for NLT 8 hours.
- After boiling the seeds, it was allowed for half an hour to settle down the foam.
- During hand blending crushing of seed was avoided.
- Storage of the filtrate in a deep freezer before precipitation or separation of mucilage

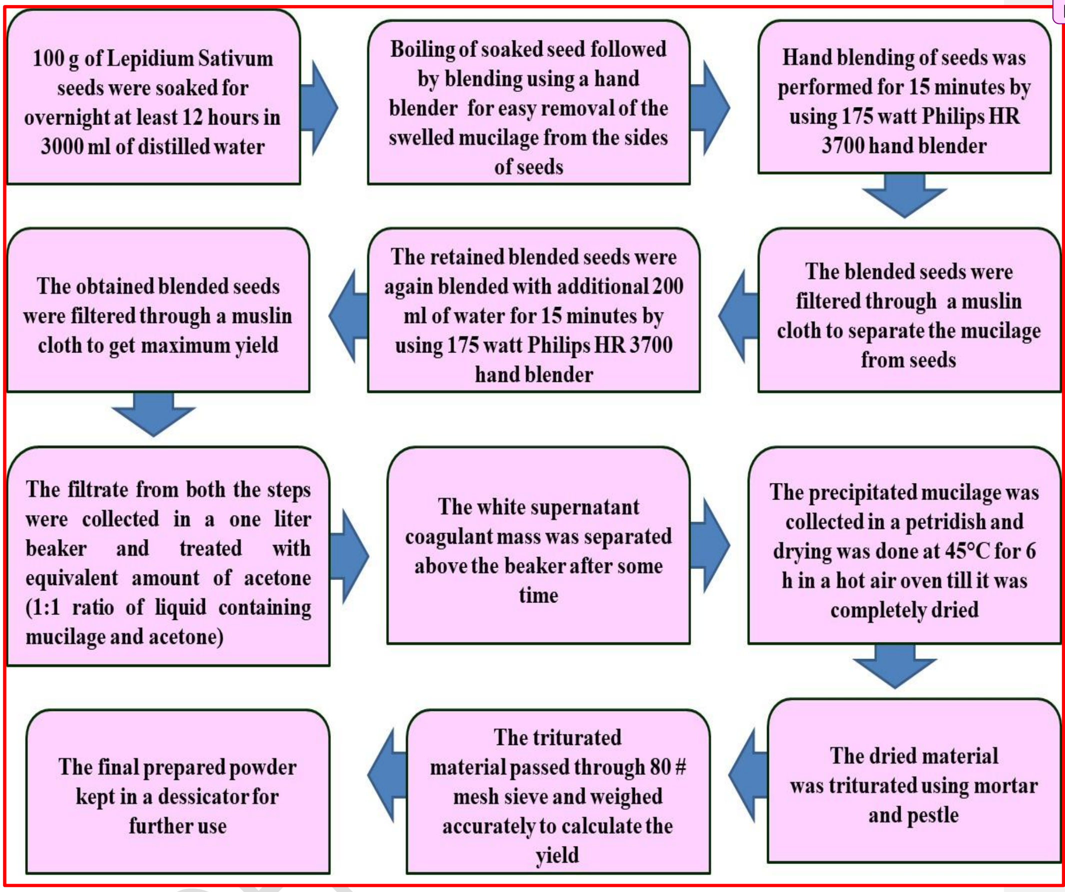

Figure 1**:** A modified method for extraction and isolation of mucilage from seeds of *Lepidium Sativum*

Evaluation of flow properties of extracted mucilage

The extracted mucilage was evaluated for flow characteristics and compressibility index. To evaluate these parameters, the sample was characterized for BD, TD, CI, HR, and angle of repose[18,19]. Approximately 1 g of sample was placed in the sample pan of a Sartorius moisture analyzer and the sample was heated at 105^{0}C temperature till constant weight was achieved. Then the final value was noted and considered as the percentage of moisture content present in the blend.

Comment [Ma3]: PH and boiling temperature and duration not mentioned in 2nd step. The 8th step should be rephrased as procedure

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In present study symbols used in method and full name in table?

RESULTS

In the present study, the extraction and isolation were carried out by using seeds of *Lepidium Sativum* as the base material with the help of suitable solvents, the extraction process, and by using suitable reagents and analytical instruments.

Extraction and isolation of mucilage from seeds of *Lepidium Sativum*

Different methods and techniques; have been reported for the isolation and extraction of seed mucilage from *Lepidium Sativum*. Thus, it is highly essential to screen the available methods to optimize the robust process. The observations of different methods used for screening the extraction of seed mucilage from *Lepidium Sativum* were summarized below in table 4.

Table 4: Observation and percentage yield of screening trials for extraction and isolation of mucilage from seeds of *Lepidium Sativum*

Sr No.	Extraction process	Observation	Observed Yield (g)	% yield
1.	**Extraction and isolation of mucilage by boiling seeds in water and using acetone as a solvent**			
	Method 1	Some of the seeds broken during hand blending	0.846	8.46
2.	**Extraction and isolation of mucilage by soaking seeds in water and using ethanol as a solvent**			
	Method 2	A very less amount of mucilage was precipitated.	0.544	5.44
3.	**Extraction and isolation of mucilage by boiling seeds in water and using ethanol as a solvent**			
	Method 3	Heating of seeds generated foam which was settled down after cooling.	0.711	7.11
4.	**Extraction and isolation of mucilage by soaking crushed seeds in water and using ethanol as a solvent**			
	Method 4	In the extracted mucilage some colored particles were observed which seem to be crushed parts of the seed.	0.628	6.28

Pilot-scale manufacturing of selected methods for extraction and isolation of mucilage from seeds of *Lepidium Sativum*

From the screening trials, three processes were selected for the further pilot-scale study. The observations of each process with their % yield were summarized below in table 5.

Table 5: Observation and percentage yield of screening trials of extraction and isolation of mucilage from seeds of *Lepidium Sativum*

Sr No.	Extraction process	Observation	Observed Yield (g)	% yield
1.	Precipitation of mucilage in acetone after boiling in water and blending by using a hand blender	The hand blender very effectively helped to remove the swelled mucilage around the seeds.	1.318	11.18
2.	Precipitation of mucilage in ethanol by using soaked seed	With the increase in batch size, the ethanol consumption was very high.	0.924	9.24
3.	Extraction and isolation of mucilage by boiling seeds in water and using ethanol as a solvent	Heating of seeds generated foam which was settled down after cooling.	1.048	10.48

Based on the results of the pilot study method (Table 5, Sr No. 1) was selected for optimization study by using an experimental design approach.

Experimental design

In the present study, the critical process attributes were optimized by using a full factorial design. The details of the factors with their studied level are presented in Tables 2 and 3. As per the factorial study design, 10 trials were planned and the % yield obtained was evaluated. Quadratic models were applied to study the relationships of factors pH of extract, water to seed ratio, and drying temperature on response % yield. A statistical model summary of the response variable (% yield) is presented in table 7.

Table 6: Summary of the response variable (% yield) obtained with different levels of studied factors i.e., pH of extract, water to seed ratio, and drying temperature

Run order	Factors (Independent variable)			Response (Dependent variable)
	pH of extract	**Water to seed ratio**	**Drying temperature**	**% Yield**
1	5.00	10	60	6.21
2	5.00	30	60	8.81
3	10.00	30	60	20.12
4	10.00	10	60	8.46
5	5.00	10	40	5.68
6	10.00	10	40	13.32
7	7.50	20	50	14.81
8	5.00	30	40	8.11
9	10.00	30	40	20.16
10	7.50	20	50	13.66

Table 7: Statistical model summary of pH of extract, water to seed ratio, and drying temperature on yield

Source of Variation	DF	Adj Sum of square	Adj Mean square	F value	P-value summary	
					Value	**Significant**
Model	7	252.813	36.116	21.47	0.045	Model significant
Linear	3	209.087	69.696	41.44	0.024	
pH of extract	1	130.195	138.195	82.16	0.012	
Water to seed ratio	1	69.208	69.208	41.15	0.023	
Drying temperature(˚C)	1	1.684	1.684	1.00	0.422	
2-Way Interaction	3	30.490	10.163	6.04	0.145	
pH of extract* Water to seed ratio	1	22.680	22.680	13.48	0.067	
pH of extract* Drying temperature(˚C)	1	4.697	4.697	2.79	0.237	
Water to seed ratio *Drying temperature(˚C)	1	3.113	3.113	1.85	0.307	
Curvature	1	13.237	13.237	7.87	0.107	
Lack-of-Fit	1	2.703	2.703	4.09	0.292	Lack-of-Fit not significant
Model Summary						
S	R-sq		R-sq(adj)			
1.29693	98.69%		94.09%			

Effect pH of extract, water to seed ratio, and drying temperature on yield

The result of the response variable yield is presented in table 6. The full quadratic polynomial equation for the measured response (yield) is given below:

Comment [Ma5]: How can we describe table 6 after table 7?

Hardness (N) = 0.15 + 1.85 pH of extract + 0.523 Water to seed ratio

+ 0.059 Drying temperature + 0.0673 pH of extract*Water to seed ratio

- 0.0307 pH of extract*Drying temperature

+ 0.00624 Water to seed ratio*Drying temperature + 2.88 Ct Pt

The above polynomial equation represents the quantitative effect of the critical attributes (factors) on the studied response yield. The above-studied equation showed a good fit with the studied factors and response due to the close concurrence of R^2 between R^2 and adjusted R^2. The result of multiple linear regression analysis for yield demonstrated a positive sign for all three studied variables. This suggested that with an increase in the level of all three studied variables, the response yield increases. The ANOVA analysis of the model indicated that the applied model is significant and all the studied factors had significantly affected ($p < 0.05$) in envisaging the response (yield). The relationship between the factors and the studied response can be easily exemplified from the Pareto chart and contour plot as shown in fig 2, fig 3, fig 4, and fig 5.

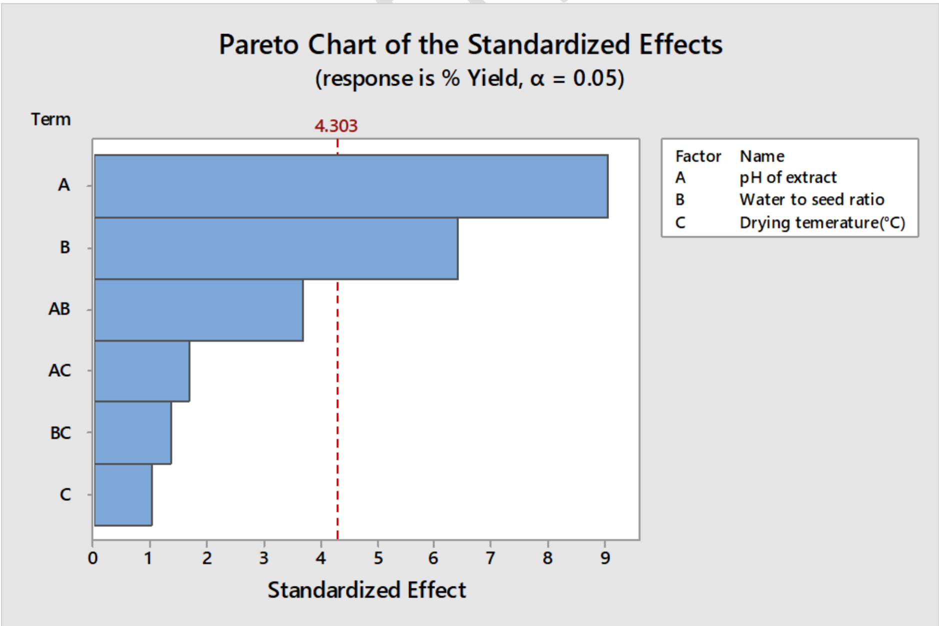

Figure 2: Contour plot of yield on the variables pH of extract, water to seed ratio, and drying temperature

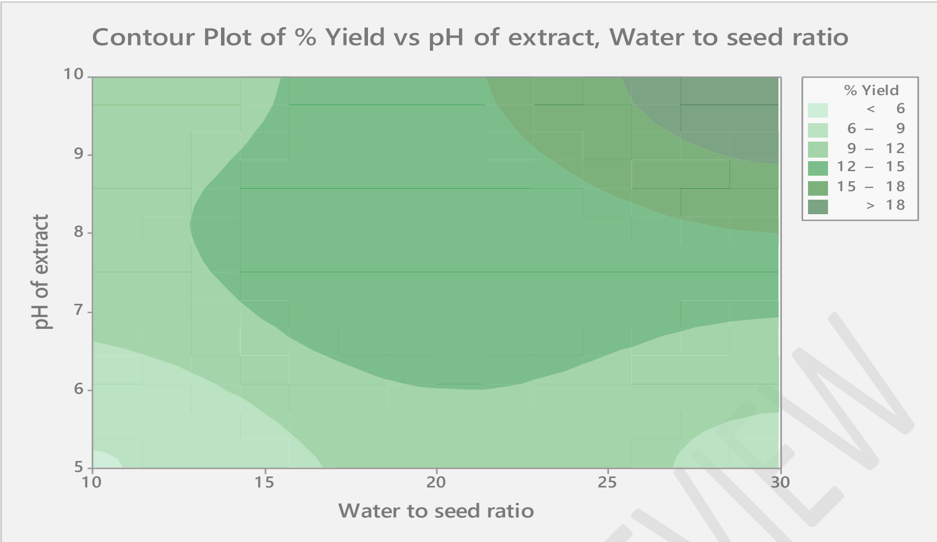

Figure 3: Contour plot of % yield on the variables pH of extract and water to seed ratio

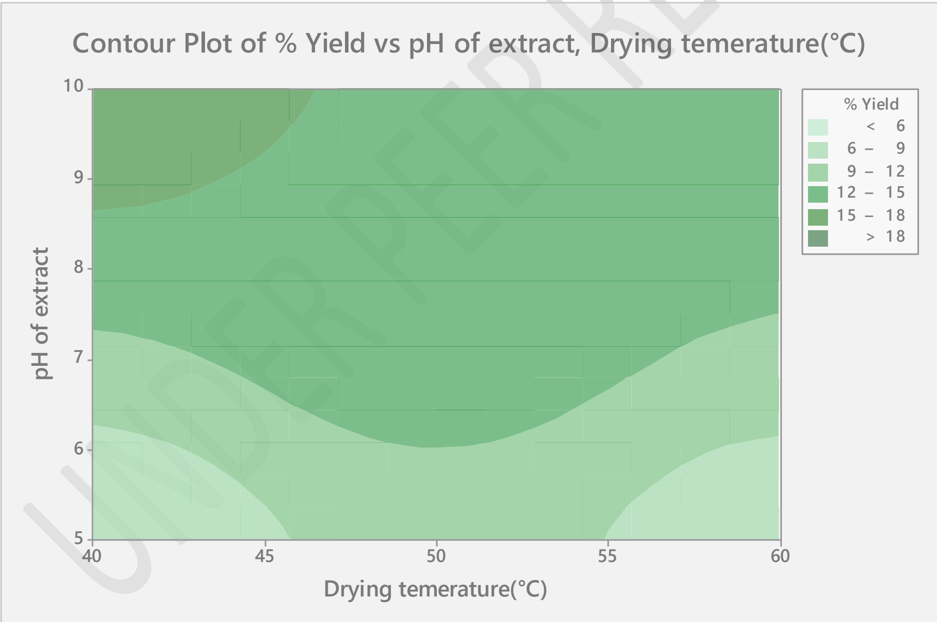

Figure 4: Contour plot of % yield on the variables pH of extract and drying temperature

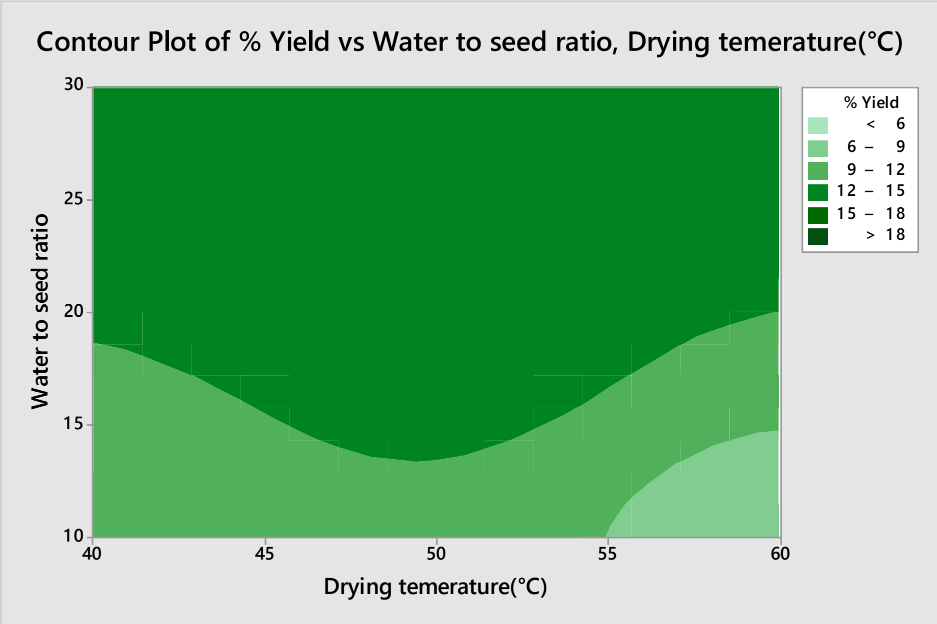

Figure 5: Contour plot of % yield on the variables pH of the water to seed ratio and drying temperature

Factorial plot for disintegration time

An interaction effect is used to study the synchronous effect when two or more independent variables have a combined influence on at least one dependent variable. In the present study, the factorial plot was used to study the main effects and interaction effects between the studied variables and the responses % yield. The main effect plot indicated that the factor pH of the extract and water to seed ratio had a significant effect on the response yield as compared to the factor drying temperature. Similarly, the interaction effect indicated that the interaction between the factors is minimum.

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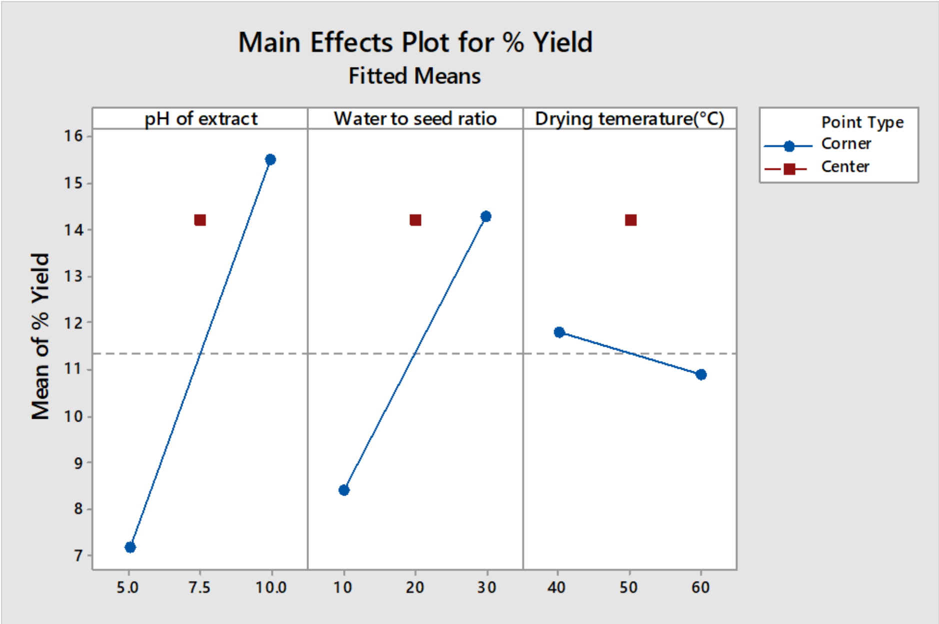

Figure 6: Main effects plot for % yield against studied variables pH of extract, water to seed ratio, and drying temperature on yield

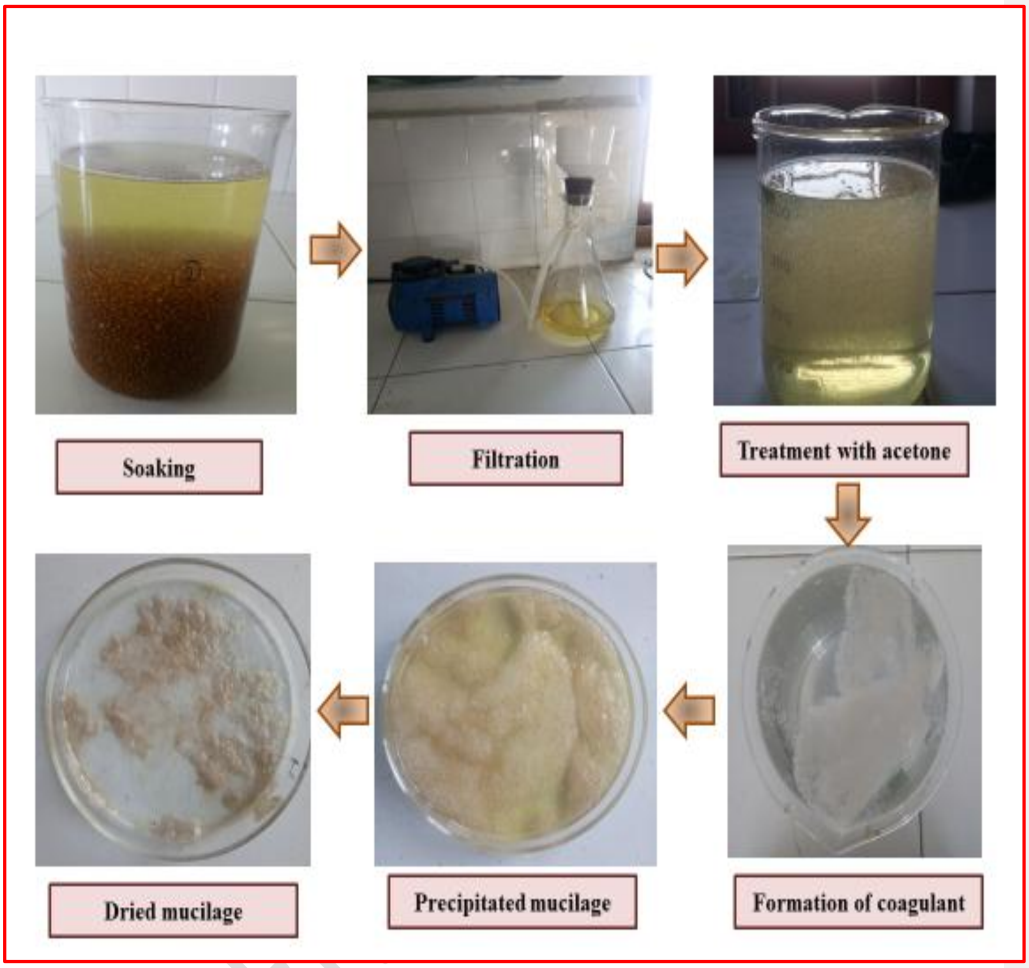

Figure 7: Pictorial diagram of final extraction process showing the dried mucilage

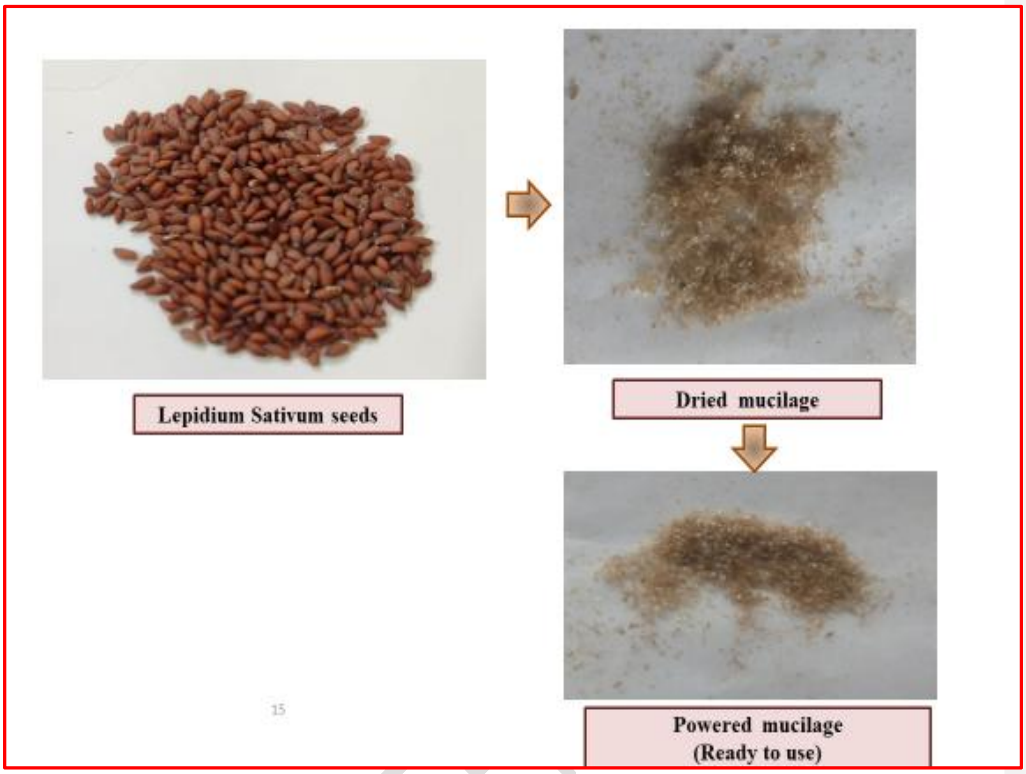

Figure 8: Pictorial presentation of raw seeds of *Lepidium Sativum*to powdered mucilage

Evaluation of flow properties of extracted mucilage

The micrometric parameters for the extracted seed mucilage were accomplished and the results are presented in Table 6. The bulk density and tapped density (0.36 and 0.45 g/cc), angle of repose (38.21°), Hausner's ratio (1.25), and Carr's index (20.00 %) were found to be satisfactory.

Comment [Ma7]: ????

Table 8: Evaluation of flow property of extracted seed mucilage of *Lepidium Sativum*

Evaluation of flow property of extracted seed mucilage of *Lepidium Sativum*	
Bulk Density	0.36±0.06 g/cc
Tapped Density	0.45±0.08 g/cc
Carr's Compressibility Index	20.00±1.1 %
Hausner ratio	1.25±1.4
Angle of repose	38.21±1.1

DISCUSSION

Preparation and optimization of extraction process of seed mucilage of *Lepidium Sativum*

In the current research, an attempt has been made to extract seed mucilage of *Lepidium Sativum*. A screening study was performed and the selected methods were evaluated. The first and second methods by using acetone and ethanol as a solvent are feasible and easy to use, thus further taken into pilot study. During execution, it was observed that the third extraction process was not user-friendly process due to difficulty in the adaptation of the extraction process. The heating process generated a large amount of foam which needs time to settle down and increases the time of the extraction process. Finally, in the fourth method, the seeds of *Lepidium Sativum* were crushed and the crushed seeds were soaked in water. The mucilage finally obtained was precipitated by using ethanol as a solvent. This method gave predictable results. However, during crushing some seed parts were crushed which was difficult to separate from the mucilage. Thus, this process was not selected for the further pilot study. Finally, from the studied methods, the first three methods (using acetone as a solvent) were selected for pilot study and optimization of the extraction process.

Four methods were evaluated in the preliminary stage and out of these four methods three methods were finalized based on steps involved, ease of processing, cost, and finally yield of the mucilage obtained. Out of three methods, finally, the first method was selected for the extraction process. The method selected was based on the feasibility of the extraction process as well as observed yield. The observed yield was 11.18 % by using acetone as a solvent and using a hand blender as compared to other methods. Based on this fact method 1, involved boiling of seeds in water and precipitation of mucilage using acetone as the final method.

It is always censorious and laborious to develop and optimize complex extraction processes for the isolation and extraction of any natural plant origin product. Thus, to arrive at a cost-effective process full factorial design was used for statistical process control. Based on the selected design, a total of 10 formulation combinations were made and evaluated for the response percentage yield. The above-discussed extraction method was used to perform the trials as per the suggested design.

The effect of each factor was studied with respect to the dependent variable percentage yield. From the study, it has been observed that the response varies significantly with the change in the independent variables. There was a clear indication from the results that the factor pH of extract and water to seed ratio imposes a great impact on the response variable yield. A trend of increase in the yield was observed with an increase in the pH of the extract from 5 to 10. It was observed that an alkaline environment helps to increase the yield. The alkali condition helps to hydrolyze the insoluble component which enables to increase in the extraction yield. The same type of pH-based extraction study concerning yield was proposed by Karazhiyan et al.[20,21] They extracted mucilage from cress seed (*Lepidium Sativum*) by optimizing pH, temperature, and water: seed ratio by using response surface methodology.

A similar trend was observed for the water to seed ratio where with increasing the water to seed ratio from 1:10 to 1:30, the yield drastically increases. The seed mucilage *of Lepidium Sativum* is a rich source of polysaccharides. The increase in the yield with an increase in the ratio of seed to water is attributed due to the hydrolysis capability of the solvent. The higher the amount of solvent, the higher the capacity to hydrolyze the polysaccharides which ultimately affect the yield. The same type of solvent effect was studied by Karazhiyan et al.[20]

A similar type of trend was observed in the main effect plot. The main effect was evaluated for % yield with three studied factors pH of the extract, water to seed ratio, and drying temperature. In our study, it has been observed that the line is horizontal for the pH of the extract, and the water to seed ratio which signifies that the main effect is present for these two factors only. In the main effect plot, the pH of the extract 10 appears to be associated with the highest % yield, and a decrease in the value from 10 to 5 decreases the % yield. The same trend was observed for the water to seed ratio. It followed a linear trend where with an increase in water to seed ratio the percentage yield increases.

The drying of mucilage is an important step in the extraction process. However, there was a clear indication from the results that there was no significant effect of the factor temperature on the response yield. It was anticipated that a higher temperature may show a charring effect. However, the efficiency of drying also depends on the type of solvent used. In our process, we are using acetone as a solvent for the extraction process. Acetone can be evaporated at a low temperature. Thus, a drying temperature of 45^0C was selected and a drying time of 6 hours was finalized. Maintaining the temperature during the extraction process is highly essential as high temperatures may lose the functional characteristics of the extracted mucilage, and may hydrolyze the polysaccharides which ultimately affect the yield. The neutral effect of temperature can also be confirmed from the main interaction plot. The main effect was evaluated for % yield with the factor temperature. In our study, it has been observed that the line is vertical which signifies that the main effect is not present for the studied factor.

Response optimization was used to find the best combination of variables studied for the response yield. This is useful to assess the best possible combinations of the variable to get the highest percentage yield. In our study, we used response optimization recommendations to arrive at a suitable extraction process parameter. The standard error of the regression equation (S) which represents the relation between actual and predicted response was found to be 1.29693 for the response percentage yield. It represents the average distance of the data points from the fitted line and was found to be 1.29 % for response yield. The adjusted regression value (R^2) was 94.09 % for the studied response yield. The lower S value and higher adjusted regression values (R^2) reflected the appropriateness or goodness of the model. Based on the factorial design study and the recommendation obtained by the response optimization run 3 and 9 are selected as an optimization process. However, the process was modified to ease the process of operation and improve the yield. The modified process implemented some modifications as compared to other reported methods such as maintaining pH to 10, optimizing the extraction temperature, soaking time of seeds to a minimum of 8 hours, time for settling down the foam, precaution to avoid crushing of seeds during hand blending, and storage of filtrate in the refrigeration before precipitation of mucilage.

The modified method based on the discussed recommendations produced a yield of 21.00 %. Many researchers worked on the extraction process of seed mucilage of *Lepidium Sativum*. However, such type of higher yield was never reported. Thus, our recommended process can be used for extraction and isolation of the *Lepidium Sativum* mucilages present around the seeds.

From the micromeritics study presented in table 8, it can be concluded that the good angle of repose (Fair- aid not needed) enables the dried mucilage to have good flow properties, and also the fair CI and HR showed that it is having good compressibility. The Bulk density and tapped density were observed as 0.36 and 0.45 g/cc respectively. Furthermore, Archana et al. [14] reported bulk and tapped densities of 0.2857 and 0.3389 (g/cc) respectively. Meanwhile, Bhatia et al.[3] reported 0.673 and 0.7 g/ml and Hassan et al. [22] reported 0.1396 and 0.2178g/ ml for bulk and tapped densities respectively. It is observed that the reported bulk densities and tapped densities vary in a wide range. The bulk density of material depends on the type of material and mainly the porosity of the studied material. Several methods and processes have been reported for the extraction of seed mucilage of *Lepidium Sativum*. It is well known that different extraction processes may change the nature of the final material and may affect the porosity of the material. The extraction process adopted by us is completely different and not reported in the studied domain. Thus, it is believed that the different bulk densities and tapped densities observed might be due to the extraction process adopted.

Comment [Ma8]: No one mentioned how come?

All these studied flow properties with good flow properties and compressibility index indicated that it can be recommended as an excipient for a direct compression formulation in the pharmaceutical and food industry manufacturing process.

CONCLUSION

The seed mucilages of *Lepidium Sativum*were extracted by using cost-effective and easily accessible solvents. The method selected was based on the feasibility of the extraction process as well as observed yield. A full factorial design was used to investigate the impact of formulation factors on response variables using a quadratic model. Statistical analysis revealed that the model was capable to measure the response with a small change in the independent variable. The statistical model represented a good relationship between the studied factors with response % yield as the actual and predicted results are in agreement with a 95% confidence interval. The study demonstrated that seed mucilage of *Lepidium Sativum* extracted by using an economic extraction process and flow property was characterized by a systematic approach.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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