

Study of Effect of Different Parameters on The Extraction Yield of Mucilage From *Salvia Hispanica* Seeds

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Abstract

Optimizing the parameters for maximizing the yield of mucilage from *Salvia hispanica* seeds is the principal aim of this research. There are four parameters that were evaluated in this research, namely; pH (ranges from 4-10), temperature (25-80°C), seed to water ratio (1:10-1:70 w/v), and seed to water contact time (3-24 h). Results showed that the yield of mucilage increased to an optimum pH of 7 and temperature of 60°C and then decreased thereafter. The yields increased to 1:40 (w/v) seed to water ratio and 12 h contact time but then leveled off. The mucilage yields that were attained under the optimal parameters were: pH 7 (9.57%), temperature 60°C (9.6%), seed to water ratio of 1:40 (9.61%), and contact time of 12 h (9.58%).

Keywords: *Salvia Hispanica*, Yield, Temperature, Seed-To-Water Ratio, Seed-To-Water Time

Introduction

Salvia hispanica (chia) seeds has remarkable recognition in both industry and research for their nutritional value and for being a source of biopolymers, in particular, soluble mucilage that aerates and expands to a gelatinous mass. This mucilage water-binding, emulsifying, and film-forming properties and is a composite of heteropolysaccharides such as xylose, glucose, arabinose, and various uronic acids. These properties are desirable and can be used in foods, medicine, biodegradable products and in many other industries (Knez et al, 2019; Khalid et al 2022). The mucilage from *S. hispanica* is pH responsive and chemically modifiable in nature (Khatoun et al., 2024 and 2025).

The ratio of mucilage to water, temperature, pH, hydration time, and extraction time are, as in many other extractions, the major factors influencing the yield of mucilage (Hussain et al., 2025; Hussain et al., 2023; Ali et al., 2022; Bukhari et al., 2022). New extraction methods based on

ultrasound and microwaving have shown promise to enhance the extraction process (Mensah et al 2024). These and other factors not only affect the yield of mucilage, but also its viscosity and the functional properties of the mucilage (Tavares et al, 2017). The use of response surface methodology, especially Box-Behnken or Central Composite designs, is dominant in the research literature to optimize extraction conditions of interest. Moderate ratios of seeds to water (medium hydration) coupled with extraction temperatures in the range of 60 to 80°C, and adequate hydration times, yield significant amounts of mucilage (varies with the polymer and procedure used). Muñoz et al., 2012 and Santana et al., 2022 reported 4 to 8% yield, with higher amounts in literature when the extraction conditions are optimized or the process is assist (Pérez-Orozco et al., 2019).

As the mucilage/hydrogels from plant seed are pH-responsive, biocompatible, non-toxic, and chemically modifiable in nature, therefore they have been successfully applied for the development of drug delivery systems and as adsorbents for water purification (Koetting et al., 2015; Sood et al., 2016; Hussain et al., 2021; Hussain et al., 2022; Hussain et al., 2023; Ali et al., 2025; Amjad et al., 2025; Hussain et al., 2025a & b; Shehzad et al., 2025; Iqbal et al., 2025; Sadraei et al., 2025; Zhao et al., 2025). The state of the literature indicates the need for systematic research into the extraction conditions to assess the yield extracted mucilage in order to optimize the yield of mucilage (gel) from the seeds of *S. hispanica*. Seed origin, the extraction process, and the analytical methods all provide complexity that needs a structured experimental approach. The present research work is aimed at understanding the composition generated from plant polymeric material to assess the individual influence of a number of factors (pH, temperature, seed to water ratio, and extraction time) on the yield of mucilage.

Methodology

Materials

S. hispanica seeds were obtained from the local market. Riedel-de Haën, Germany, provided analytical-grade chemicals and reagents. Throughout the experiment, distilled water (DW) was used.

Mucilage Extraction

S. hispanica seeds were manually cleaned and soaked in DW, 1:40 (w/v) for a period of 12 h at a temperature of 70°C to allow proper swelling and mucilage release. After this period, the soaked seeds were mucilage-sieved using a spatula to rub the pretentious seeds against a nylon sieve. Collected mucilage was purified using a two-step solvent extraction. First, *n*-hexane was used to remove any non-polar impurities, and DW was used afterward to remove any polar impurities. The purified material was dried in a hot vacuum oven at 60°C for 48 h. The dried material was ground and passed through a No. 60 sieve, and the fine mucilage was stored in sealed containers for later use. The extraction procedure was slightly modified from a method previously described (Keaton et al. 2024).

Determination of Experimental Yield

Equation given below was used to evaluate the percentage yield of mucilage:

$$\text{Experimental yield of mucilage (\%)} = \frac{\text{Dry weight of mucilage}}{\text{Weight of } S. \text{ hispanica seeds taken for mucilage extraction}} \times 100$$

Effects of Different Parameters on Mucilage Yield

Effects of pH

This study was conducted to determine the effect of pH on the amount of mucilage which can be extracted from *S. hispanica* seeds. 500 mL of DW was added into separate beakers and then the pH was set to the values of 4-10 using HCl or NaOH. The other extraction conditions, *i.e.*, temperature, seed to water ratio, and contact time, were kept constant at their optimized values. The seeds were then soaked under the conditions set, and the obtained mucilage was collected and the yield was determined.

Effects of Temperature

To determine the effect of temperature on mucilage yield, temperature was set to 25 to 70°C while keeping pH, seed to water ratio, and contact time at their optimal levels. Each beaker was filled with 500 mL of DW and the desired temperature was set before the optimized amount of seeds was added. Mucilage extraction was then done as per the procedure described above, and the yield was measured afterwards.

Effect of Seed-to-Water Ratio

In order to understand the impacts of varying the seed-to-water ratio in the mucilage yield of the *S. hispanica* seeds, the DW was taken into the beakers, 500 mL at a time, and the pH of each water sample was adjusted to the desired and optimized level with the use of HCl or NaOH, as done previously. All remaining extraction parameters such as temperature, pH and, contact time were held constant to ensure that the seed to water ratio was the only variable present. The seeds were then added to all of the prepared media with the specified ratios and allowed to hydrate under controlled settings. After the soaking period, the mucilage was extracted using the standard method, the seed residues were separated, and the dried and weighed end products were used in order to determine the yields. This methodology, therefore, offered a systematic assessment of the degree of water availability on the efficiency of mucilage extraction that was obtainable for each ratio.

Effect of Seed-to-Water Contact Time

In order to determine the effect of contact time on mucilage yield released from the seeds of *S. hispanica*, 500 mL of DW were transferred into different beakers and the pH of each sample was adjusted to the optimized value using dilute HCl or NaOH. The remaining variables for the extraction conditions, temperature, and seed-to-water ratio, were kept at their previously determined optimum conditions in order for contact time to be the only variable that can be changed without restriction. Various contact durations were established by incorporating precisely measured quantities of *S. hispanica* seeds to pH-adjusted DW. The combinations were allowed to hydrate for the specified period of time under controlled temperature and atmospheric conditions that facilitates the release of the mucilage. After the hydration period, the mucilage was recovered by applying the pH-adjusted DW samples to the developed standard procedure without the seed residues, whereupon the mucilage samples were oven-dried to constant weight and weighed to determine the extraction yield for each contact time. This repetitive sequential process made it possible to determine and analyze the influence of contact time on the seeds' swelling, release of mucilage, and efficiency of the complete extraction process.

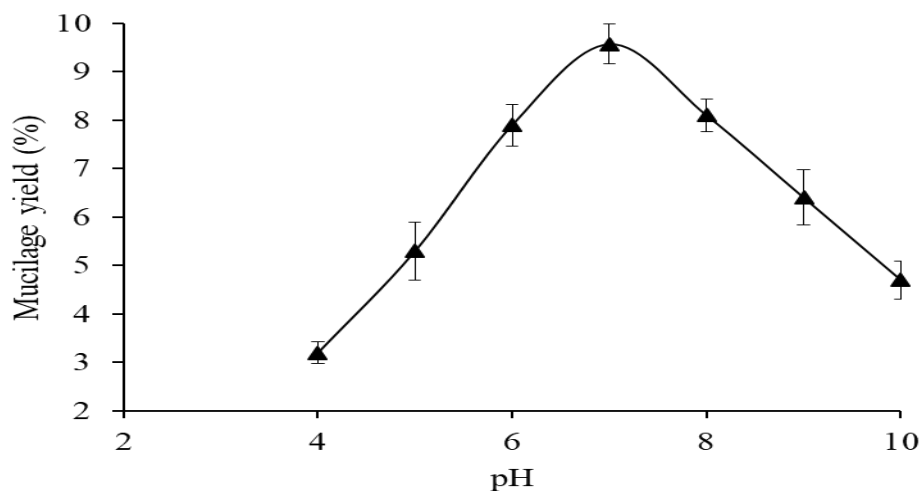
Table 1. Parameters Affecting Conditions for The Extraction of Mucilage

pH	Temperature (°C)	Seed-to-water ratio (w/v)	Seed-to-water contact time (h)
4	25	10	3
5	30	20	6
6	40	30	9
7	50	40	12
8	60	50	15
9	70	60	18
10	80	70	24

Results and Discussion

Effect of pH

The role of pH on mucilage extraction at a temperature of 60°C was determined by keeping a seed-to-water ratio of 1:40 (w/v), and a contact time of 12 h unchanged. At strongly acidic pH conditions (0-4), extraction yields were very low (< 3.2 %), and therefore, further experiments were performed only on pH 4-10 (Figure 1). With the pH increasing from 4 to 6, mucilage yields increased steadily up to 7.9% and subsequently to the maximum yield of 9.57% at pH 7. Then, beyond that pH, the yields began to decline to about 4.7 at pH 10. Greatest yield at neutral pH was likely due to greater mucilage swelling, corroborating previous observations that neutral conditions are most favorable for the hydration of plant mucilage, including that from *Artemisia vulgaris*. The decline in yields noted in alkaline pH conditions might be due to greater mucilage component solubility at higher pH, leading to a net loss of mucilage in the extracted fraction as a result of partial dissolution. In addition, pH 10 would favor the hydrolysis of polysaccharides to smaller more soluble fractions that are more easily lost during extraction. Just as in other studies on pH-responsive plant polysaccharides, we have similar results here. For example, *A. vulgaris* hydrogels swell best in neutral to mildly basic pH (6.8–7.4) and collapsed in strongly acidic (high) conditions due to the protonation of the carboxyl group (Bukhari et al., 2022). Such has also been reported on the seed mucilage of Psyllium (*Plantago ovate*) where maximum extraction was at pH 7.5, and lower yields was at either side of the pH (Assi et al., 2023). These results suggest that pH is one of the most important factors during extraction with neutral pH providing the most favorable conditions for optimum swelling, greatest stability, and recovery efficiencies of the mucilage.

**Fig. 1:** Effect of pH on The Mucilage Yield

The Effect of Temperature on Mucilage Yield

To study the effect of temperature on the mucilage extractable yield, other conditions were held constant (pH 7, seed-to-water ratio of 1:40 w/v, and 12 h contact time) during the temperature range study (25 to 80°C). At 25°C, the yield was low (2.9%) but with increasing temperature, the yield increased to the maximum of 9.6% at 60°C, where the temperature was further increased 80°C, and the yield was 5.9% (Figure 2). During the first rise in temperature, the yields increase the first time as a result of improved mass transfer. Higher temperatures reduce the viscosity of the surrounding medium as well as the enhanced solvent diffusion into the seed matrix, which in turn promotes the release of the mucilage. Higher temperatures of above 60°C, however, will result in the thermal degradation of polysaccharides, thus, decreasing the mucilage that could be recovered. This pattern has also been sighted in relevant studies. For example, Nazir et al. (2017), studies, stated that the mucilage from basil seed mucilage extraction was also temperature dependent as the yields increased up to the optimum range of 50–65°C and then decreasing due to polysaccharide degradation. Other studies that focused on basil seeds also stated that the optimum extraction was around 50°C and at a pH of 7 with a water to seed ratio of 30 to 1, and then the degradation was due to thermal de-polymerization (Salehi et al., 2025). Overall, the data point to the conclusion that while moderate heating improves the extraction of the mucilage, excessive heating on the other hand improves the breakdown of the mucilage and also lowers the potential yield.

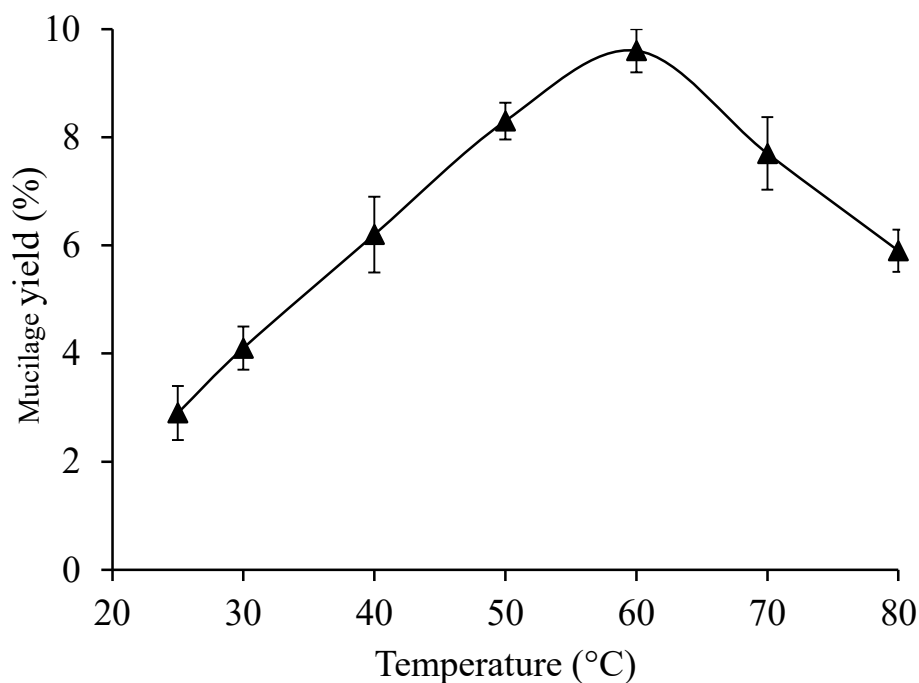


Fig. 1: Effect of Temperature on The Mucilage Yield

The Influence of the Seed-to-Water-Ratio

With the extraction conditions held constant at pH 7, 60°C, and 12 h of seed-to-water contact time, the mucilage yield when using different seed-to-water ratios was explored. As the ratio changed from 1:10 to 1:40, there was an increase in the mucilage yield from 3.57 to 9.61% at an increasing rate. This could be the result of the increased volume of water acting as a greater force in mass transfer than in the mucilage being held at the seed surface. Furthermore, the water in the slurry would decrease the viscosity and stickiness to increase extraction efficiency. For ratios above 1:40 (w/v), the yield plateaued, indicating that a solvent–solute equilibrium was reached, and so further

dilution did not enhance extraction (Figure 3). Other authors describe similar behavior for other mucilage-secreting seeds. For instance, mucilage extraction from *Mimosa pudica* was reported to show yield improvement with increasing seed-to-water ratios up to 1:20 (w/v), after which no further gains could be expected due to the limitation of diffusion at high water volume (Bukhari et al., 2022). Also, the water-to-seed ratio for the seeds of *O. basilicum* (basil) was reported as a significant factor, with a central composite design suggesting optimal yield at around 66.8:1 (Nazir et al., 2017). Overall, these findings suggest a precise optimization of the seed-to-water ratio, as retention of water will limit the amount of mucilage released, whereas too much water will yield no further gains past equilibrium. For *S. hispanica*, a ratio of about 1:40 (w/v) may represent the best trade-off between extraction efficiency and solvent consumption.

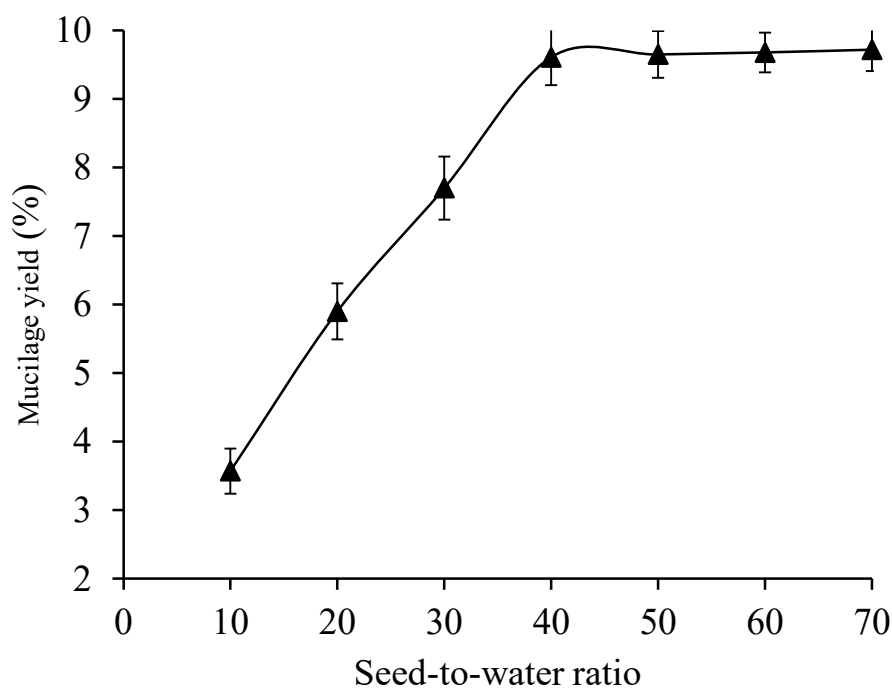


Fig. 1: Effect of Seed-to-Water Ratio on The Mucilage Yield

Effect of Seed-to-Water Contact Duration

The study aimed to measure the yield of mucilage by changing the length of time. The seeds were allowed to rest in water, in which the temperature and pH were controlled at 60°C and 7, respectively. The seed to water ratio of 1:40 was used, as previously discussed. Between 3 and 12 h of contact time, there was a marked increase in the mucilage yield, increasing from 3.2 to 9.58%. This can be explained by the increased water infiltration into the seed coat, creating hydrostatic pressure which causes increased mucilage release. However, at 12 h, the yield plateaued, suggesting equilibrium between the seeds and the extraction medium had been reached. Other documents describing seed mucilage extraction have also reported an initial rise in yield followed by yield stagnation or decline. This trend was also seen in the *M. pudica* example where Box-Behnken optimization revealed extraction yield was significantly higher at 6 h extraction times, and any extraction time longer than 6 h extraction reduced yield (Bukhari et al., 2022). Similarly, in *Ximenia Americana*, extraction during extended times enhanced yield due to better mass transfer as water penetration increased to a point, yet excessive extraction duration achieved no additional yield increments due to equilibrium limits (Bazezew et al., 2022). In *O. basilicum*, however,

extraction time sensitivity was lower and extraction yields increased between about 1.6 and 3 h, moderate yields, also with prolonged times potentially reducing extraction yield due to the undesirable degradation or dissolution of other materials (Nazir et al., 2017). These accounts suggest that 12 h is sufficient to obtain a predominant yield for the factors optimized in this study. Also, 12 h extraction time not only lacks yield-material improvements but extends time loss in large scale operations in the form of energy expenditures that could promote destruction of the mucilage including degradation.

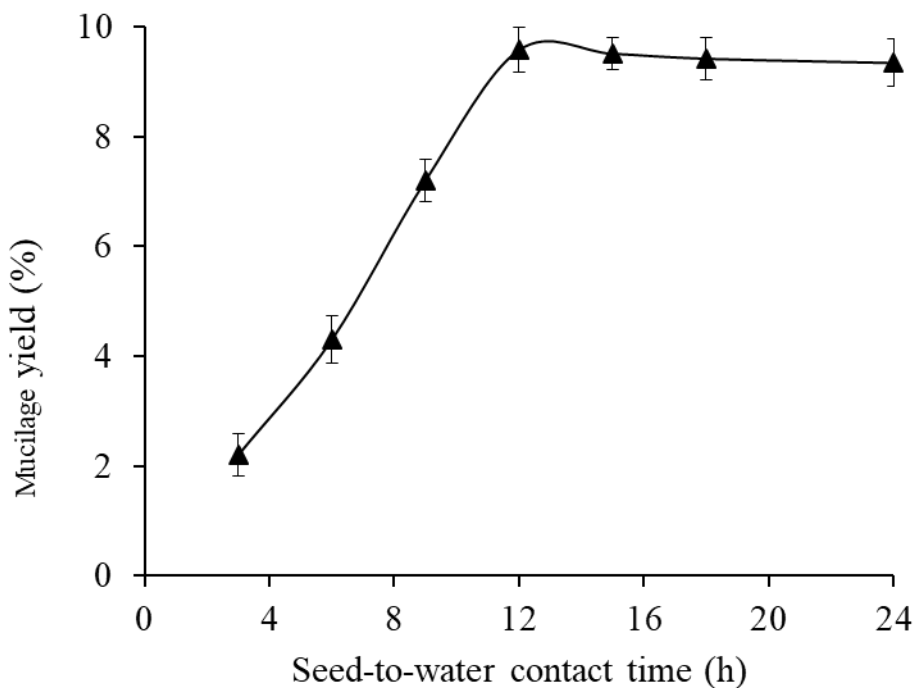


Fig. 4: Effect of Seed-To-Water Contact Time on The Mucilage Yield

Conclusion

The present study investigated the influence of various factors on the yield mucilage from *S. hispanica* seeds, such as pH, temperature, seed-to-water ratio, and seed-to-water contact time. The evidence suggests that mucilage yield depends on the operational parameters. The highest yield was observed at pH 7, since the polysaccharides are likely better swelling in the near physiological state. The temperature positively influence the extraction rate; however, at temperatures exceeding 60°C, the yield decreased, indicating that there was thermal degradation of polysaccharides. Extracting mucilage from the seeds was more effective the higher the seed to water ratio, up to a 1:40 (w/v) ratio, at which point equilibrium was reached. The same saturation equilibrium was evident with contact time, with recovery increasing up to 12 h, after which it stabilized. The greatest extraction efficacy was achieved with the parameters that had been previously established: pH 7, 60°C, a seed-to-water ratio of 1:40 (w/v), and a 12 h contact time. These results signify the development of a systematic procedure that allows for the procurement of substantial mucilage from *S. hispanica* seeds while also demonstrating the need for a systematic approach to parameter optimization in the food, pharmaceutical and biomaterial industries. The results obtained are suggestive of ample opportunity to for practical application of mucilage extraction and indicate that the extraction will result in a product of consistent quality and a process of high efficiency.

Statements & Declarations

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Competing Interests

The authors have no relevant financial or non-financial interests to disclose.

Availability of Data and Material

Corresponding authors will provide data on request.

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