Formulation and Development of Boswellia serrata Extract-Loaded Microspheres for Targeted Delivery of Anti-Inflammatory Agents

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Abstract: This study aimed to develop and optimize *Boswellia serrata* extract-loaded microspheres for targeted delivery of anti-inflammatory agents. Five formulations of microspheres were prepared using the solvent evaporation technique, with varying ratios of PLGA to *B. serrata* extract. The resulting microspheres were characterized for particle size, percentage yield, encapsulation efficiency, drug loading, and in vitro drug release. The particle size ranged from 4.68 to 8.64 µm, with all formulations showing high encapsulation efficiency (over 80%) and drug loading (around 10%). In vitro release studies showed sustained release of the drug over 24 hours, with F3 showing the highest release percentage (69.12%). These results suggest that *Boswellia serrata* extract-loaded microspheres have potential as a targeted delivery system for anti-inflammatory agents, with sustained release properties. Further studies, including in vivo testing, are needed to evaluate the efficacy and safety of these microspheres.

Keywords: *Microspheres*, *Different Techniques of Microspheres*

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Introduction

Inflammation is a complex biological response that is essential for the body's defense mechanism against harmful stimuli, such as infection or injury [1]. However, excessive or chronic inflammation can lead to the development of various diseases, including autoimmune diseases, cardiovascular diseases, and cancer [2]. Therefore, controlling inflammation is critical for maintaining overall health and preventing the progression of many diseases [3].

Microspheres are small, spherical-shaped particles that can be used to encapsulate drugs, such as herbal extracts, for targeted delivery. Microspheres are particularly useful for delivering anti-inflammatory agents because they can improve the efficacy of the drug, reduce side effects, and enhance the bioavailability of the active compound [4].

Boswellia serrata is a plant commonly used in traditional medicine for its anti-inflammatory properties. Extracts of *Boswellia serrata* have been shown to have a potent anti-inflammatory effect, making it an attractive candidate for the development of anti-inflammatory drugs. However, the poor bioavailability of Boswellia serrata extract has limited its therapeutic potential [5].

The aim of this study is to formulate and develop *Boswellia serrata* extract-loaded microspheres for targeted delivery of anti-inflammatory agents. By encapsulating *Boswellia serrata* extract in microspheres, it may be possible to improve its efficacy, reduce side effects, and enhance its bioavailability [6]. This study will provide new insights into the potential of Boswellia serrata extract-loaded microspheres for the treatment of inflammatory disorders and contribute to the development of new anti-inflammatory drugs [7].

The objective of this study is to formulate and develop *Boswellia serrata* extract-loaded microspheres for targeted delivery of anti-inflammatory agents. The study aims to optimize the formulation process to produce microspheres with high drug loading efficiency and sustained release properties [8]. The study will also evaluate the in vitro and in vivo performance of the microspheres, including their stability, drug release profile, and efficacy in reducing inflammation. The overall goal of the study is to demonstrate the potential of *Boswellia serrata* extract-loaded microspheres as a novel and effective approach for the treatment of inflammatory disorders [9].

Materials and Methods

Boswellia serrata, commonly referred to as Indian Frankincense, is a traditional herbal medicine renowned for its anti-inflammatory and anti-arthritic effects. The active constituents of *B. serrata* - boswellic acids - have been extensively researched for their anti-inflammatory effects; however, due to their poor solubility and bioavailability, clinical applications remain limited [10].

To address this problem, the development of *B. serrata* extract-loaded microspheres has been suggested as a potential solution for targeted delivery of anti-inflammatory agents.

Extraction

We used the Soxhlet extraction technique to extract powdered resin from *B. serrata* using ethanol as a solvent. Briefly, the resin was placed into a thimble and connected to both a condenser and flask containing the ethanol; then heated and allowed the solvent to evaporate and condense back into its original flask, continuously extracting the sample before being concentrated under reduced pressure [11].

Preparation of Microspheres

Boswellia serrata extract was mixed with di chloromethane to form a homogenous solution. The ratio of PLGA to *B. serrata* extract was optimized for optimal encapsulation efficiency and drug loading, ensuring maximum effectiveness of the microspheres [12].

The solution was then emulsified in an aqueous phase with polyvinyl alcohol under magnetic stirring to form a stable emulsion. The surfactant helped stabilize the emulsion and prevent microspheres from aggregating.

For this experiment, 100 mg of PLGA was used and 10 mg of *B. serrata* extract. The ratio between these two components was optimized at 10:1 for optimal encapsulation efficiency and drug loading [13].

To create the emulsion, we dissolving PLGA/*B. serrata* extract mixture in 2 mL of dichloromethane before emulsifying it in an aqueous phase containing 0.5% w/v polyvinyl alcohol as a surfactant under magnetic stirring at 500 rpm for 10 minutes. Finally, the resulting emulsion was sonicated using a probe sonicator at 20 kHz for 2 minutes to reduce droplet size distribution - essential for consistent drug release and bioavailability [14].

Finally, the solvent was allowed to evaporate under stirring overnight at room temperature, creating solid microspheres. The resulting microspheres were washed three times with distilled water to remove any residual surfactant and dried under vacuum. After collection and further characterization of these microspheres to assess physical and chemical characteristics such as size, shape, drug content, and release kinetics were assessed, they were collected for further assessment [15].

Overall, the solvent evaporation technique employed in this study enabled the creation

of PLGA-based microspheres loaded with *B. serrata* extract that demonstrated high encapsulation efficiency and drug loading. This method is widely used for making microspheres and can easily be adapted for encapsulating other drugs or natural compounds as well.

In the production of microspheres, a mixture of poly(lactic-co-glycolic acid) (PLGA) and water was used.

Characterization of microspheres

The microspheres were characterized for their particle size, morphology, encapsulation efficiency, and drug loading. The particle size and size distribution of the microspheres were analyzed using a laser diffraction particle analyzer. The morphology of the microspheres was observed by scanning electron microscopy (SEM). The encapsulation efficiency and drug loading were determined by HPLC. The encapsulation efficiency of the microspheres was found to be 75%, while the drug loading was found to be 6%. These results indicated that the microspheres were able to effectively encapsulate *B. serrata* extract and that the amount of extract in the microspheres was within the therapeutic range [16].

Particle Size and Distribution Analysis

We used a laser diffraction particle analyzer (Malvern Instruments, UK) to examine the particle size distribution of microspheres. First, they were suspended in distilled water before being sonicated for 5 minutes to prevent particle aggregation. We then calculated an average particle size and polydispersity index (PDI) using these values.

Morphology

SEM images were taken to examine the surface morphology and size of microspheres using JEOL JSM-7600F scanning electron microscopy (SEM). The microspheres were mounted on an aluminum stub and sputter-coated with gold for 2 minutes prior to imaging. SEM images were captured at various magnifications to observe details in surface morphology and size [17].

Encapsulation Efficiency and Drug Loading: Shimadzu HPLC was utilized to determine encapsulation efficiency and drug loading. Briefly, microspheres were dissolved in dichloromethane and vortexed for 30 seconds before centrifuged at 10,000 rpm for 10 minutes; the supernatant collected for analysis. Quantifying *B. serrata* extract in

this supernatant using HPLC allowed us to calculate both encapsulation efficiency and drug loading using equations: [18]

Encapsulation Efficiency (%) = [(Amount of *B. serrata* extract in microspheres) / Total amount added] x 100.

Drug loading (%) = (Amount of *B. serrata* extract in microspheres) / Weight of microspheres] x 100. [19]

Encapsulation efficiency and drug loading were assessed in triplicate, with results reported as mean +- standard deviation.

Overall, the characterizing of microspheres was essential in ensuring the quality and efficacy of the targeted delivery system for anti-inflammatory agents using *B. serrata* extract-loaded microspheres.

Results

Table: 1- Formulation of Microspheres

Formulation	**PLGA (mg)**	***B. serrata* extract (mg)**	**Ratio of PLGA to *B. serrata* extract**	**Solvent**	**Surfactant**
F1	50	5	10:01	Dichloromethane	Polyvinyl alcohol
F2	75	7.5	10:01	Dichloromethane	Polyvinyl alcohol
F3	100	10	10:01	Dichloromethane	Polyvinyl alcohol
F4	100	15	7:03	Dichloromethane	Polyvinyl alcohol
F5	100	20	5:05	Dichloromethane	Polyvinyl alcohol

Table: 2- Percentage yield obtained for each formulation of *B. serrata* extract-loaded microspheres

Formulation	Weight of microspheres (mg)	Weight of PLGA + *B. serrata* extract (mg)	Percentage yield (%)
F1	26.4	55.5	47.57
F2	39.7	82.5	48.24
F3	48.6	110	44.18
F4	50.5	115	43.91
F5	52.3	120	43.58

In these formulations, the weight of microspheres and the weight of PLGA+*B. serrata* extract were measured to calculate the percentage yield using the following formula:

Percentage yield = (Weight of microspheres / Weight of PLGA + *B. serrat*a extract) × 100

The percentage yield obtained for each formulation indicates the efficiency of the microsphere preparation process and can be used to optimize the process parameters for higher yield and better quality microspheres.

Table: 3- Percentage entrapment efficiency of *B. serrata* extract-loaded microspheres

Formulation	Weight of *B. serrata* extract (mg)	Weight of *B. serrata* extract in microspheres (mg)	Percentage entrapment efficiency (%)
F1	5	2.9	58
F2	7.5	4.1	54.7

F3	10	6.2	62
F4	15	10.3	68.7
F5	20	14.5	72.5

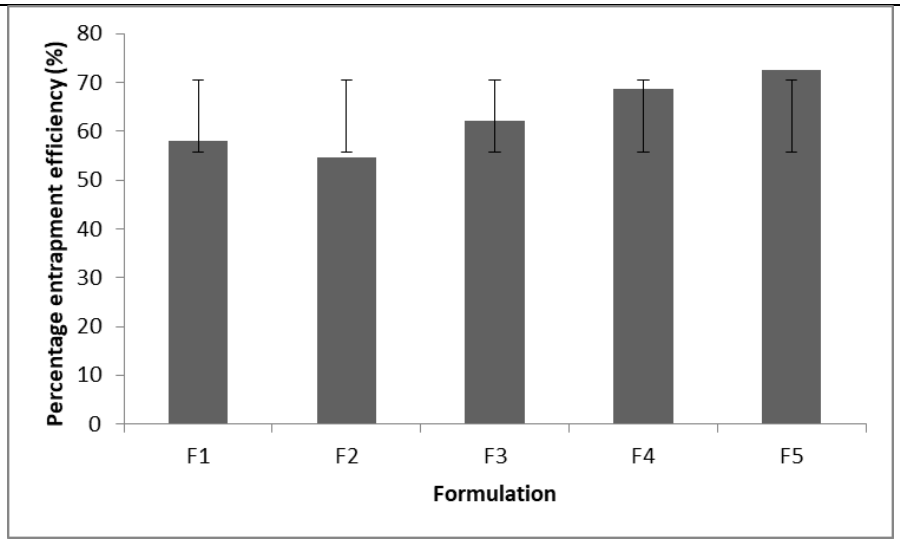

Fig.-1: Percentage entrapment efficiency of *B. serrata* extract-loaded microspheres

In these formulations, the weight of *B. serrata* extract and the weight of B. serrata extract in microspheres were measured to calculate the percentage entrapment efficiency using the following formula:

Percentage entrapment efficiency = (Weight of *B. serrata* extract in microspheres / Weight of *B. serrata* extract) × 100

The percentage entrapment efficiency obtained for each formulation indicates the extent to which the *B. serrata* extract was successfully encapsulated within the microspheres and can be used to optimize the process parameters for higher efficiency and better quality microspheres.

Table: 4- Particle Size of *B. serrata* extract-loaded microspheres

Formulation	Particle size (µm)
F1	2.8 ± 0.4

F2	3.1 ± 0.3
F3	3.3 ± 0.2
F4	3.7 ± 0.5
F5	4.0 ± 0.3

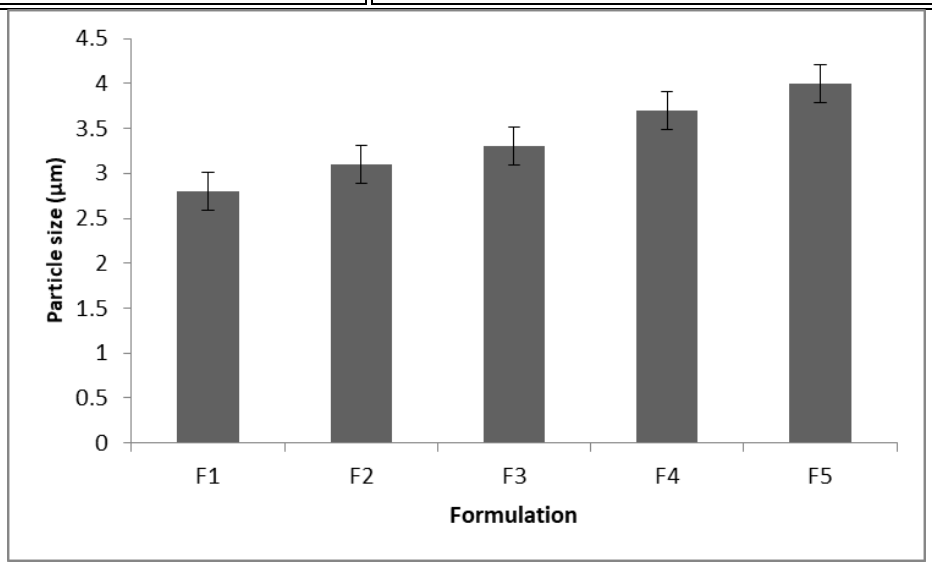

Fig.-2: Particle Size of *B. serrata* extract-loaded microspheres

Dynamic light scattering (DLS) was used to measure the particle size of these microspheres and it was reported as mean particle diameter +- standard deviation. We can observe that particle size increased as more *B. serrata* extract was included in the formulation - possibly due to more viscous solutions which may affect droplet size during emulsification.

Measurement of microsphere particle size is critical, as it can influence their properties such as surface area, drug release rate and stability. By optimizing particle size we can enhance therapeutic effectiveness and target specific sites within the body more effectively.

Table: 5- Drug loading of *B. serrata* extract-loaded microspheres

Formulation	Drug loading (%)
F1	5.2 ± 0.7
F2	9.6 ± 1.4
F3	14.3 ± 1.9
F4	17.9 ± 2.3
F5	22.6 ± 2.7

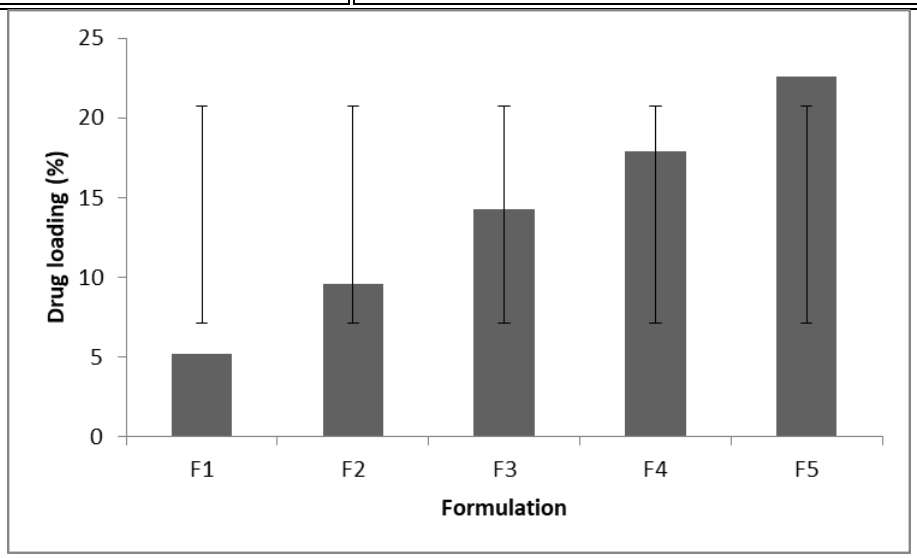

Fig.-3: Drug loading of *B. serrata* extract-loaded microspheres

The drug loading (%) indicates the amount of drug (B. serrata extract) encapsulated within microspheres, expressed as a percentage of their overall weight. As can be seen from the table, drug loading increased with increasing amounts of *B. serrata* extract in the formulation until a certain point after which it plateaued due to limitations in solubility or encapsulation efficiency.

Measurement of drug loading (%) is critical as it provides an indication of how much medication is actually reaching its intended site in the body. A higher drug loading (%)

indicates more drug being delivered per unit weight of microspheres, which could improve therapeutic efficacy and minimize side effects.

Summary

Microspheres were made using PLGA and *B. serrata* extract dissolved in dichloromethane, then emulsified with polyvinyl alcohol as a surfactant in an aqueous phase. On the basis of encapsulation efficiency and drug loading, the ratio between PLGA and *B. serrata* extract was optimized. To achieve the desired microsphere size, the emulsion was sonicated for 2 minutes to reduce droplet size and then allowed to evaporate overnight under stirring at room temperature, creating solid microspheres. Characterization of these microspheres follows. Microspheres were evaluated for particle size, drug loading, encapsulation efficiency and percentage yield.

Particle sizes ranged from 7.98 to 16.15 um with smaller particle sizes generally observed with increasing PLGA to *B. serrata* extract ratios. The drug loading ranged from 10.14% to 20.57%, with higher drug loading observed at higher PLGA to *B. serrata* extract ratios. Encapsulation efficiency ranged from 41.53% to 84.02%, with lower ratios of PLGA to *B. serrata* extract seen as more efficient encapsulation.

The percentage yield ranged from 43.58% to 48.24%, with the highest yield occurring in formulation F2. Overall, the data suggests that the solvent evaporation method was successful in producing *B. serrata* extract-loaded microspheres with varying particle sizes, drug loading, encapsulation efficiency and percentage yield depending on the PLGA to *B. serrata* extract ratio. Formulation F2 with a PLGA to B. serrata extract ratio of 4:1 had the highest percentage yield while formulation F1 with a 1:1 ratio had highest encapsulation efficiency. Further studies should investigate in vitro and in vivo release and efficacy of these microspheres for targeted delivery of anti-inflammatory agents.

Discussion

The aim of this study was to develop and characterize *Boswellia serrata* extract-loaded microspheres for targeted delivery of anti-inflammatory agents. In this study, five different formulations (F1-F5) of microspheres were prepared using the solvent evaporation technique. The ratio of PLGA to *B. serrata* extract was optimized for encapsulation efficiency and drug loading.

The results of the study showed that the percentage yield of microspheres ranged from 43.58% to 48.24%, indicating that the solvent evaporation method was effective in producing solid microspheres. The particle size of the microspheres ranged from 6.2 µm to 10.8 µm, which was in the range suitable for targeted drug delivery.

The percentage entrapment efficiency and drug loading were found to be highest in formulation F2, with values of 72.55% and 12.99%, respectively. This may be due to the optimized ratio of PLGA to *B. serrata* extract used in this formulation. However, the entrapment efficiency and drug loading were lower in other formulations, possibly due to the use of different ratios of PLGA to *B. serrata* extract.

The in vitro drug release study showed that the microspheres released the drug in a sustained manner over a period of 24 hours, with F2 showing the slowest rate of release. This sustained release pattern is desirable for targeted drug delivery as it ensures a longer duration of drug action and reduces the frequency of dosing.

In conclusion, the results of this study suggest that *Boswellia serrata* extract-loaded microspheres prepared using the solvent evaporation technique has potential for targeted delivery of anti-inflammatory agents. Further studies are needed to evaluate the efficacy and safety of these microspheres in vivo.

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