Ind. J. Pharm. Edu. Res., 2025; 59(4):1285-1292.
https://www.ijper.org

Original Article

Screening and Optimization of Various Formulation and Processing Parameters for the Development of Aquasomes

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ABSTRACT

Objectives: The research aims to enhance understanding of aquasome preparation, offering insights into optimizing formulations for potential applications in drug delivery systems. **Materials and Methods:** Aquasomes composed of lactose and trehalose as oligosaccharides encapsulate calcium phosphate cores. The cores synthesized by sonication and co-precipitation methods were screened based on % yield for calcium phosphate synthesis. Various coating methods were employed for the screening based on carbohydrate loading capacity. The study evaluates these formulations to optimize carbohydrate loading and particle size for potential applications in bioactive drug delivery to enhance drug loading capacity and minimize use of bioactive drugs for initial screening and optimization. **Results and Discussion:** Ceramic core preparation via sonication yielded higher percentages (69.6%) compared to co-precipitation (54.06%). Trehalose demonstrated superior carbohydrate loading (67.74%) compared to lactose (55.81%) using the lyophilisation method. The Plackett-Burman design identified four significant factors affecting particle size, which are sonication time, amplitude, temperature and carbohydrate concentration. The Central Composite Design (CCD) optimization revealed that increased sonication time and carbohydrate amount led to higher carbohydrate loading. Lower sonication temperatures resulted in greater carbohydrate loading. Particle size decreased with increased sonication time but increased with higher carbohydrate amounts and sonication temperatures. The optimized formulation achieved a carbohydrate loading of 104.17±0.021 mg and a particle size of 407.2±0.34 nm, closely matching predicted values. **Conclusion:** The study shows sonication time, temperature and carbohydrate concentration influence aquasome development. Essential for optimizing their performance in various biomedical applications, particularly in drug delivery. Further exploration and validation of these optimized formulations in practical settings are warranted to fully exploit their potential benefits.

Keywords: Aquasomes, Calcium Phosphate, Carbohydrate, Sonication, Plackett-Burman design.

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Received: 22-07-2024;
Revised: 16-01-2025;
Accepted: 05-06-2025.

INTRODUCTION

Nanotechnology has revolutionized the field of medicine, offering innovative solutions for diagnosing and treating various diseases. Among the numerous nano carrier systems developed, aquasomes have emerged as a promising platform with significant potential in biomedical research.[1] Aquasomes are three-layered, core-shell nanoparticles composed of a solid core enveloped by an inner polymer shell and an outer polyhydrated oligomeric layer.[2]

Aquasomes possess a unique structure that confers several desirable properties. The solid core is typically composed of materials like tin oxide, calcium phosphate, ceramic carbon (diamond), or hydroxyapatite, which serves as a reservoir for the encapsulation of hydrophilic or hydrophobic molecules.[3] The oligomeric layer is made up with pyridoxal-5-phosphate, cellobiose, trehalose, sucrose, or lactose, which provides structural stability and protection to the encapsulated cargo. The outer polyhydrated oligomeric layer enhances the bioavailability and biocompatibility of the nanoparticles, facilitating their interaction with biological systems.[4]

One of the most promising applications of aquasomes lies in their capability to encapsulate and deliver therapeutic compounds efficiently. These nano carriers can accommodate a wide range of small-molecule drugs, vitamins, enzymes, peptides, proteins and genetic materials and protect them from degradation and premature clearance.[5] Aquasomes have demonstrated enhanced solubility, improved bioavailability by targeted delivery of encapsulated drugs and increased stability, leading to increased therapeutic efficacy and decreased side effects.[6]

The screening of aquasome formulation is a crucial step in the development and optimization of these unique nanocarrier systems. To enhance solubility, improve bioavailability, target

DOI: 10.5530/ijper.20250371

drug delivery and control release of encapsulated drugs or to increase stability of bioactive molecules, aquasome development is required. However, the formulation process and the selection of suitable components play a vital role in the overall performance and stability of aquasomes. Screening is necessary to evaluate various parameters.[7]

Plackett-Burman design is a widely used statistical design technique for screening of formulations, including aquasomes. The Plackett-Burman design is particularly useful when there are numerous variables that need to be looked into, and it helps to know the significant factors that influence the responses. The design involves generating a series of experimental runs, where each factor is varied over two levels (high and low) in a specific pattern.[8]

In the context of aquasome formulations, the factors that can be screened using the Plackett-Burman design may include type and concentration of core materials; type and concentration of carbohydrates for coating on core material; preparation method; processing parameters (sonication time, sonication temperature, etc.,).[9]

The Central Composite Design (CCD) involves a factorial design with center points and axial points, allowing for the estimation of quadratic effects and interactions among the factors. By conducting experiments according to the CCD design matrix, researchers can develop mathematical models (e.g., quadratic polynomial equations) that describe the relationship between the input factors and the responses.[10]

Once the experimental data is collected, statistical analysis techniques, such as Analysis of Variance (ANOVA) and regression analysis, are used to analyze the data and identify the significant factors and their interactions. The resulting mathematical models can then be used to generate response surface plots, which provide a visual representation of the effects of the factors on the responses.[11]

The central composite design, combined with the screening results from the Plackett-Burman design, provides a systematic and efficient approach for optimizing aquasome formulations by considering the significant factors and their interactions, ultimately leading to the development of optimized nanocarrier systems with desired characteristics and performance. Additionally, screening helps identify potential challenges or limitations associated with specific formulations, enabling researchers to make informed decisions and further refine the aquasome system for enhanced therapeutic efficacy and safety.[12]

MATERIALS AND METHODS

Materials

Disodium Hydrogen Orthophosphate, Calcium Chloride, Diammonium Hydrogen Phosphate were purchased from Sisco Research Laboratories Pvt. Ltd., Mumbai, Maharashtra, India. Calcium nitrate, Lactose, Trehalose were purchased from HiMedia Laboratories Pvt. Ltd., Mumbai, Maharashtra, India.

Methods

Preparation of Ceramic core

By Sonication method

Weighed 8.9 g disodium hydrogen phosphate and 7.35 g calcium chloride were dissolved in 60 mL distilled water in a separate beaker. Calcium chloride solution was dropwise added in disodium hydrogen phosphate in an ultrasonic bath for 2 hr at 4°C. The prepared ceramic core was separated by centrifugation for 15 minutes at 4°C at 15,000 rpm. The prepared core material was washed with double distilled water and suspended in 50 mL distilled water. Then filtered through a 0.2 µM Millipore filter. The prepared core material was collected and dried.[13,14]

By co precipitation under reflux conditions

The 100 mL of diammonium hydrogen phosphate solution and 100 mL of calcium nitrate solution were prepared. The solution of diammonium hydrogen phosphate was dropwise added into the calcium nitrate solution. The flask was kept with the charge funnel and reflux condenser with carbon dioxide trap installed at 75°C. The calcium nitrite pH was kept at 8-10 by employing a 25% conc. aqueous ammonia solution. For 4-6 days, the above combination was stirred on a magnetic stirrer while the pH and temperature were maintained. Core material was cleaned twice in deionized water and then reconstituted in 50 mL of deionized water. Thereafter, they passed through a Millipore 0.2 µM filter. Core material was collected and dried in a hot air oven for an entire night at 100°C.[14]

Carbohydrate coating on the core

For the preparation of aquasomes, two carbohydrates (lactose and trehalose) were selected for the screening study. Carbohydrate coating on the core material was carried out by three methods: direct incubation method, non-solvent method and lyophilization method.[15] Solutions of lactose and trehalose were prepared. Core material was added to carbohydrate solution, then sonicated by using a probe sonicator (18 W, 30% amplitude).[16] These dispersions were incubated in an orbital shaker at 100 rpm and 25°C for 1 hr. The prepared aquasome molecules were centrifuged at 2000 rpm for 15 min, then collected and dried for the direct incubation method. In the non-solvent method, when dispersion was put for sonicate, at that time 1 mL of acetone was also added, followed by centrifugation, collection and drying. For the lyophilization method, this dispersion was put for the lyophilization process following sonication to facilitate the nearly irreversible adsorption of carbohydrates onto the ceramic surfaces.[17]

Screening of Process and Formulation variables for optimization of Aquasomes Formulation

The Plackett and Burman design was chosen to screen the formulation and process factors for the development of aquasomes. There were 7 factors (X1-X7) selected for at 2 levels -1 and +1. The 1 dependent variable (Y1) was selected to see the response of different factors, as given in Table 1.[18,19]

Optimization study of Aquasomes Formulation

Based on the screening study by Plackett-Burman design was carried out successfully to check process and formulation variables. From this study, there are three variables whose more significant effects are given in Table 2. They are optimized by using central composite design.[20]

Characterizations

% Yield of Calcium Phosphate

The % yield of each synthesis was calculated based on the molecular weight of the starting material and ceramic core material. % Yield was calculated using the following equation:[21]

$$\%Yield = \frac{Practical\ Yield}{Theoratical\ Yield} \times 100$$

FT-IR study

The FTIR of ceramic core and carbohydrate coated core materials were measured using Fourier Transform Infrared Spectrophotometer (Bruker). Excipient mixture and KBr was used for making the pellets, which were then scanned over a 400-4000 cm^{-1} range.[22,23]

Particle size analysis

Zetasizer Nanoseries ZS (Malvern Instruments, Malvern and Worcestershire, UK), Photon Correlation Spectroscopy (PCS) was used to evaluate the size and size distribution of the ceramic core. Filtered distilled water was used to dilute each sample to an appropriate concentration. The analysis was done at 25°C and 90° angles.[24]

Zeta-potential measurement

Zeta potential of suitably diluted ceramic core dispersion was determined using Zetasizer Nanoseries ZS (Malvern Instruments, UK).[25]

% Carbohydrate loading

It was carried out by using the anthrone reagent method. Anthrone reagent was added to the sample for this method, and it was heated in a boiling water bath before it rapidly cooled. Carbohydrate acidic instances caused the hydrolysis of carbohydrates into hydroxymethyl furfural, which was then combined with anthrone reagent to produce a complex with a blue-green color. Absorbance was recorded by using a UV-visible spectrophotometer at 625 nm.[26]

RESULTS AND DISCUSSION

Screening of methods for preparation of ceramic core material for aquasomes was done based on % Yield of Calcium Phosphate. Then prepared ceramic core material characterized by FTIR spectroscopy, particle size and zeta potential.

% Yield

The % Yield of ceramic core by sonication method was 69.3% and by co-precipitation method was 54.06%.

The sonication process was chosen based on yield percentage for further analysis and preparation of calcium phosphate core material for aquasome preparation.

FT-IR study of ceramic core

The FTIR spectra of the ceramic core showed characteristic peaks at 3554.10, 3166.61 cm^{-1} due to hydroxyl stretch (-OH), at 1129.81, 873.47, 527.55 and 577.55 cm^{-1} for different vibrational b and s of Phosphate (PO_4^{-3}).

Particle size analysis

The particle size for ceramic core was performed in triplicate by using a zetasizer. The particle size of the prepared ceramic core was found to be 250.3±1.1 nm.

Zeta-potential measurement

Charge on the ceramic core was directly obtained from the instrument. For calcium phosphate ceramic core, it was found to be 4.81±0.156 mV.

Selection of Carbohydrate

For screening of lactose and trehalose, carbohydrate loading was estimated by the anthrone reaction method. It was found that 47.56%, 42.28% and 55.81% for lactose and 59.27%, 50.58% and 67.74% for trehalose by the direct incubation, by non-solvent and by lyophillization method respectively. From the above data by the lyophilization method, carbohydrates were bound in higher percentages. As compared to lactose, trehalose was a higher percentage bound to ceramic core. So, trehalose also forms more hydrogen bonds, which increased the aquasomes stability. Trehalose is selected for further aquasome preparation. Trehalose coated ceramic core particles were further characterized by FTIR spectroscopy, particle size and zeta potential.

FT-IR study Carbohydrate coated core

The FTIR spectra of carbohydrate-coated core particles showed peaks at 3288.17 cm^{-1} due to lactose alcohol group (-OH stretching vibration), at 2387.72 cm^{-1} due to C-H bond of glucose and galactose unit (C-H stretching vibration), at 1388.84 cm^{-1} C-H bending vibrations and at 989.93 cm^{-1} C-O-C ether unit bond (glucose and galactose).

Table 1: Different variables for Plackett and Burman design.

Process and Formulation variables		Coded value	
Variables		-1	+1
X1	Concentration of Core (mg)	50 mg	100 mg
X2	Concentration of Carbohydrate (mg)	100 mg	300 mg
X3	Sonication temperature (°C)	4	25
X4	Sonication time (min)	5	10
X5	Sonication amplitude (Watt)	18	30
X6	Incubation Period (min)	30	60
X7	Concentration of non-solvent (mL)	0	1
Y1	Particle Size (nm)		

The total 12 formulations were prepared by following Plackett-Burman design given in Table 3.

Table 2: Variables for Central Composite Design.

Variables		-1	+1
X1	Sonication time (min)	5	10
X2	Sonication temperature (°C)	4	24
X3	Amount of Carbohydrate (mg)	100 mg	300 mg
Y1	Carbohydrate loading (mg)		
Y2	Particle Size (nm)		

Screening of Process and Formulation variables for Optimization of Aquasomes Formulation

Plackett and Burman design was used for screening process and formulation variables. The composition and conditions for F1-F12 formulations are given in Table 3 with their response variable.

Mathematical model for Plackett-Burmann Design

The result of each experiment is linear, as is the effect of each variable X1, X2, X3, …. Xk. The response measured for each experiment is y. An additive model for 7 variables is proposed to:

$$Y=\beta0+\beta1x1+\beta2x2+\beta3x3+\beta4x4+\beta5x5+\beta6x6+\beta7\times7$$

From the above equation, the effects of each variable found were shown in the form of a parato chart in Figure 1.

Calculation of the effect

To check the repeatability of variables centre point was tested four times (Batch F9-F12). It showed also similar result which is given in Table 3.

The mean value was found to be 325.2 nm and the standard deviation is estimated as 5.8. The degree of freedom is 3. So, the standard error for each coefficient was found to be $5.8/\sqrt{8}=2.04\%$. The value of t for probability was 95% and 3 degrees of freedom was 3.18. Thus, the parameters that exceed the value of 2.04x3.18=6.48 are significant. As we concluded that 4 factors are statistically significant at a confidence level better than 95%. The factors sonication time, sonication amplitude, sonication temperature and concentration of carbohydrate affect the particle size. So, these are used for further optimization study.

Optimization study of Aquasome Formulation

From the screening study, there were three variables we have taken for optimization which showing more significant effects. The optimization study was carried out by Central Composite Design. Three independent variables were sonication time (X1), sonication temperature (X2) and amount of carbohydrate (X3) and dependent variables were carbohydrate loading (Y1) and particle size (Y2) were selected. The responses are given in Table 4.

ANOVA on carbohydrate loading for different formulations

As sonication time increased and carbohydrate amount increased, the carbohydrate loading was increased. At a lower sonication temperature, it gave more carbohydrate loading. ANOVA was applied on carbohydrate loading to study the effect and fitting of the model.

The effect of various variables is represented mathematically by multiple linear regressions as reduce polynomial equation:

Carbohydrate Loading=+98.18+2.56*A-2.41*B+45.9
8*C-2.40*A2-0.34*B2-2.23*C2

ANOVA model for response carbohydrate loading, which indicates that sonication time, sonication temperature and

amount of carbohydrate show significant effect. The model was further evaluated by creating the three-dimensional response surface graph of the carbohydrate loading as shown in Figure 2.

ANOVA on Particle size for different formulations

As sonication time increased, particle size decreased and the amount of carbohydrate and sonication temperature increased, particle size increased. ANOVA was applied on particle size to study the effect and fitting of the model.

The effect of various variables represented mathematically by multiple linear regression as reduce polynomial equation:

Particle size=+408.87-9.18*A-3.83*B+9.51*C+2.37*A2+6.18*B 2+1.55*C2

The ANOVA model for response particle size indicated that sonication time, sonication temperature and concentration of carbohydrate showed significant effects. The model was further estimated by plotting a three-dimensional response surface plot, as shown in Figure 3.

Overlay plot of Carbohydrate loading and Particle size

The best dependent variable effect was found by superimposing the response surfaces or contour plots over one another. The area that was colored yellow was when both responses were satisfactory and appropriate. An optimal region was found here, balancing both responses.

In the presented overlay plot (Figure 4), the check point method was employed to compare the results obtained from the performed check point batch (C1 batch) with those of the actual check point batch (A1 batch). The comparison focuses on two

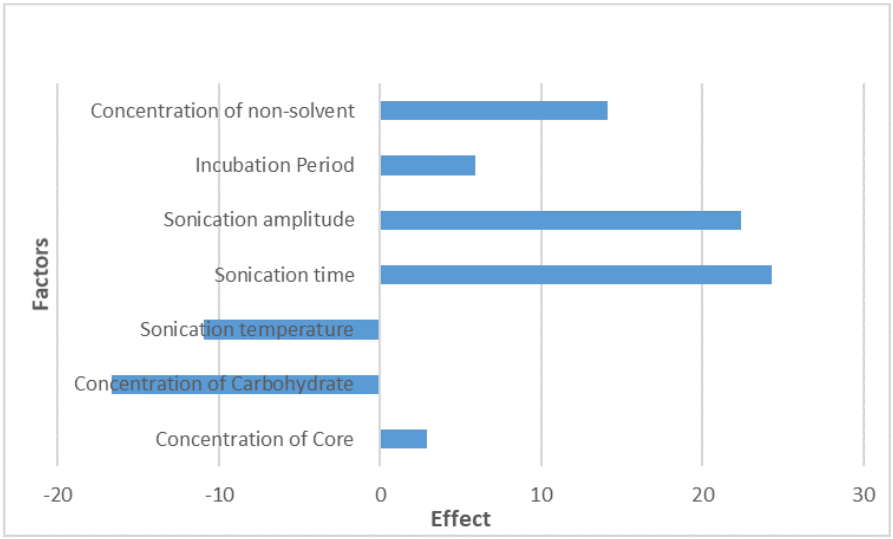


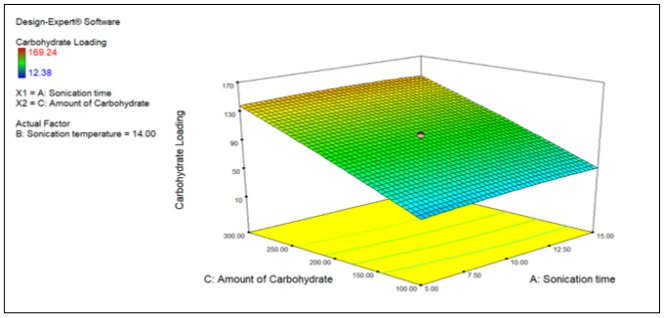
Figure 1: Pareto chart for effect of different factors.

Table 3: Formulations variables for Plackett and Burman design.

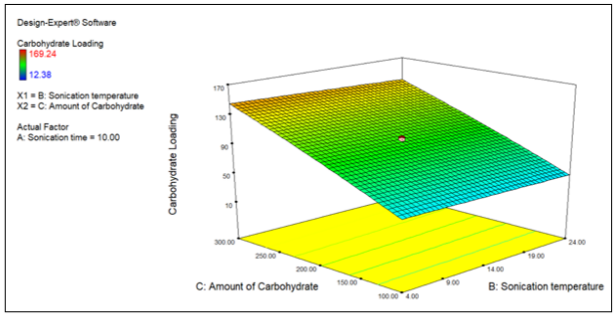
Batch	Variables level in actual form							Response variables
	X1	X2	X3	X4	X5	X6	X7	Y1
F1	50	300	4	10	18	60	1	431.3
F2	100	100	4	5	30	30	1	421.5
F3	100	300	4	5	18	60	0	326.8
F4	50	300	25	5	18	30	1	315.6
F5	100	100	25	10	18	30	0	375.1
F6	50	300	4	10	30	30	0	402.7
F7	50	100	25	5	30	60	0	377.3
F8	100	300	25	10	30	60	1	426.7
F9	75	200	14.5	7.5	24	45	0.5	322.4
F10	75	200	14.5	7.5	24	45	0.5	328.7
F11	75	200	14.5	7.5	24	45	0.5	318.5
F12	75	200	14.5	7.5	24	45	0.5	331.2

Table 4: Variables for Central Composite Design.

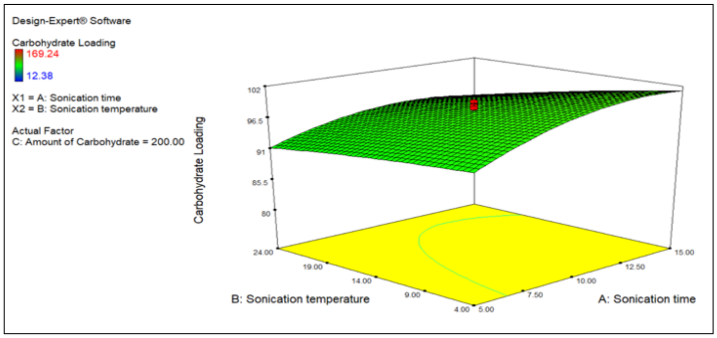
Std.	Variables level in Actual form			Response variables	
	X1	X2	X3	Y1	Y2
1	5	4	100	48.21	417.83
2	15	4	100	51.12	409.32
3	5	24	100	45.26	425.31
4	15	24	100	49.23	398.65
5	5	4	300	142.53	448.45
6	15	4	300	143.28	420.55
7	5	24	300	139.59	426.31
8	15	24	300	132.54	411.59
9	1.59	14	200	80.13	428.62
10	18.41	14	200	100.56	400.34
11	10	-2.82	200	100.42	430.63
12	10	30.82	200	91.89	419.89
13	10	14	31.82	12.38	390.15
14	10	14	368.18	169.24	434.18
15	10	14	200	98.21	414.12
16	10	14	200	98.69	410.56
17	10	14	200	99.35	410.94
18	10	14	200	97.84	410.92
19	10	14	200	98.26	405.81
20	10	14	200	97.12	401.23



(a) Interaction between Sonication time and Amount of Carbohydrate

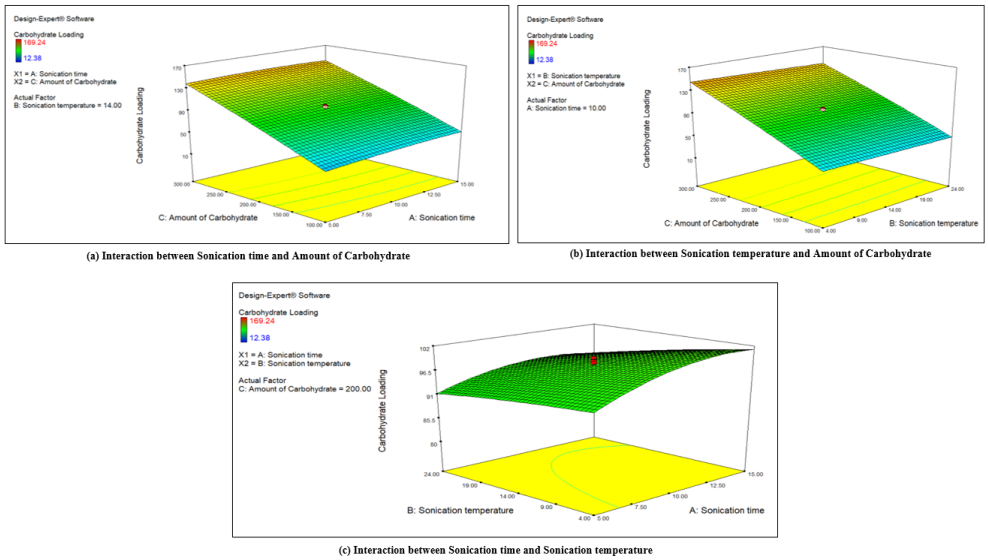


(b) Interaction between Sonication temperature and Amount of Carbohydrate



(c) Interaction between Sonication time and Sonication temperature

Figure 2: Response Surface Plot of Carbohydrate Loading.



(a) Interaction between Sonication time and Amount of Carbohydrate

(b) Interaction between Sonication temperature and Amount of Carbohydrate

(c) Interaction between Sonication time and Sonication temperature

Figure 3: Response Surface Plot of Particle size.

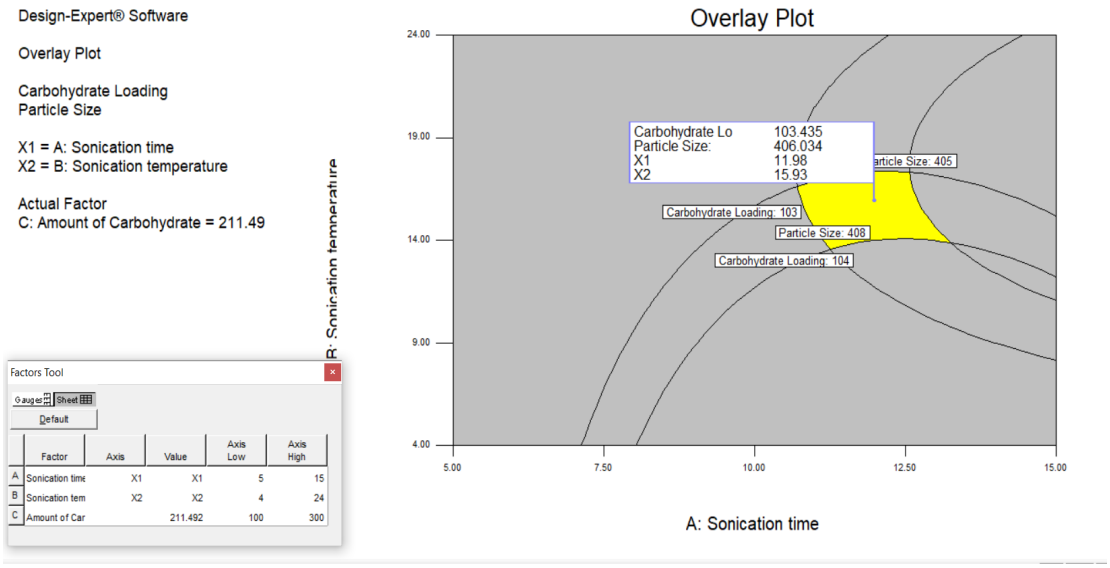


Figure 4: Overlay plot.

crucial parameters: carbohydrate loading (Y1) and particle size (Y2).

For carbohydrate loading (Y1), the Performed Check Point batch exhibits a value of 104.17 mg with a minimal standard error of 0.021. In contrast, the Actual Check Point batch shows a slightly lower value of 103.43 mg. This minor discrepancy suggests a negligible deviation between the performed and actual batches in terms of carbohydrate loading.

Similarly, for particle size (Y2), the performed check point batch records a size of 407.2 nm with a standard error of 0.34 nm, while the actual check point batch shows a slightly smaller size of 406.03 nm. This difference, though minimal, indicates a consistent trend with the carbohydrate loading comparison, suggesting congruence between the performed and actual batches.

The overlay plot visually illustrates these comparisons, showing the proximity of the data points representing the performed check point batch to those of the actual check point batch. The tight clustering of the data points reinforces the notion of minimal deviation between the two batches, further validating the robustness and reliability of the Check Point method in ensuring consistency and accuracy in batch production.

Overall, the discussion highlights the effectiveness of the Check Point method in verifying the performance of batches, with the presented results indicating close agreement between the performed and actual batches for both carbohydrate loading and particle size parameters.

CONCLUSION

In conclusion, our study was specially conducted to identify the optimum value of formulation and processing parameters because aquasomes are widely used for biological products that are costly and difficult to synthesize. One of the studies was carried out to conduct and optimize formulation before drug loading. This study demonstrates the critical influence of sonication time, sonication temperature and carbohydrate concentration on aquasome development. Significant effects on carbohydrate loading and particle size were observed, as confirmed by ANOVA models. The utilization of three-dimensional response surfaces and contour plots facilitated the identification of an optimal region, depicted in yellow, where both responses met the desired criteria. This integrated approach enables the fine-tuning of aquasome formulations, balancing between carbohydrate loading and particle size, essential for optimizing their performance in various biomedical applications, particularly in drug delivery. Further exploration and validation of these optimized formulations in practical settings are performed to fully exploit their potential benefits. So, this work is beneficial in the formulation of bioactive compounds or drugs to formulate aquasomes.

ACKNOWLEDGEMENT

We extend our sincere appreciation to the School of Pharmacy, Parul University, Vadodara, Gujarat, for their support in equipping our laboratories with a full of chemicals. This contribution has undoubtedly strengthened our research infrastructure and will continue to play a crucial role in fostering innovation and excellence in pharmaceutical education and research. This work is a part of Ph.D. research.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

mg: Microgram; **g:** Gram; **mL:** Millilitre; **nm:** Nanometer; **Rpm:** Rotations per minute; **SEM:** Scanning Electron Microscope; **ºC:** Degree Centigrade; **ANOVA:** Analysis of Variance; **CCD:** Central Composite Design.

REFERENCES

1. Sucharitha P. Somes: A review on composition, formulation methods and evaluations of different types of "somes" drug delivery system. Int J App Pharm. 2020;12(6):7-18.
2. Peeriga R, Sridevi AR. Unlocking the Potential of Aquasomes: A Comprehensive Review on Innovative Nanocarriers in Drug Delivery and Beyond. Int J Chem Biochem Sci. 2023;24(6):405-10.
3. Ayom GE, Malima NM, Owonubi SJ, Revaprasadu N. Aquasomes: a novel nanocarrier system for drug delivery. In Adv Nano. 2023 Jan. Academic Press. 281-301.
4. Banerjee S, Sen KK. Aquasomes: A novel nanoparticulate drug carrier. J Drug Deliv Sci Technol. 2018;43:446-52.
5. Umashankar MS, Sachdeva RK, Gulati M. Aquasomes: a promising carrier for peptides and protein delivery. Nanome: Nanotechnol Biol Med. 2010;6(3):419-26.
6. Kulkarni S, Prabhakar B, Shende P. Aquasomes: Advanced vesicular-based nanocarrier systems. Current Pharma Design. 2022;28(29):2404-14.
7. Pradhan M, Parihar AK, Singh D, Singh MR. Quality by design and formulation optimization using statistical tools for safe and efficient bioactive loading. InAdvances and Avenues in the Development of Novel Carriers for Bioactives and Biological Agents. 2020;555-94. Academic Press.
8. Damera DP, Kaja S, Janardhanam LS, Alim S, Venuganti VV, Nag A. Synthesis, detailed characterization and dual drug delivery application of BSA loaded aquasomes. ACS App Bio Materials. 2019;2(10):4471-84.
9. Sahu AK, Jain V. Screening of process variables using Plackett-Burman design in the fabrication of gedunin-loaded liposomes. Artif Cells Nanomed Biotechnol. 2017;45(5):1011-22.
10. Bei YY, Zhou XF, You BG, Yuan ZQ, Chen WL, Xia P, *et al.* Application of the central composite design to optimize the preparation of novel micelles of harmine. Int J Nanomed. 2013:1795-808.
11. Bhattacharya S. Central composite design for response surface methodology and its application in pharmacy. InResponse surface methodology in engineering science 2021 Jan 28. IntechOpen.
12. Luiz MT, Viegas JS, Abriata JP, Viegas F, de Carvalho Vicentini FT, Bentley MV, *et al.* Design of experiments (DoE) to develop and to optimize nanoparticles as drug delivery systems. European Journal of Pharmaceutics and Biopharmaceutics. 2021;165:127-48.
13. Vishwas C. Bhagat*, Pravin B. Awate, Dipak P. Kardile, Nilesh A. Narote, Rajkumar V. Shete and Adinath C. Bhusari. Aquasomes: A Novel Approach in Drug Delivery System for Poorly Water-Soluble Drug. Biol Forum. 2023. 15(4):144-9.
14. Gupta AK, Gupta D, Gupta V. Aquasomes: a self-assembled nano-particulate carrier system. Int J Cur Res Rev. 2021;13(4):44-52.
15. Dhar AD, Sarkar SS, Das SD. An overview on aquasomes: a novel approach for delivering macromolecules. World Wide J. 2022;11(12).
16. Jagdale S, Karekar S. Bird's eye view on aquasome: Formulation and application. J Drug Deliv Sci Technol. 2020;58:101776.
17. Rajak P, Islam R, Karmakar A, Bhuyan B. Aquasomal drug delivery system: A special emphasis on the formulation techniques and applications. Ars Pharmaceutica (Internet). 2023;64(4):359-75.
18. Ramani VD, Jani GK, Sailor GU. Application of Plackett-Burman design for screening of factors affecting pitavastatin nanoparticle formulation development. Folia medica. 2021;63(5):775-85.
19. Banerjee SR, Sen KK. Preparation and evaluation of surface modified nanoparticles of calcium phosphate as extract carrier. Int J Appl Pharm. 2020;12(4):248-57.
20. Hassan H, Adam SK, Alias E, Meor Mohd Affandi MM, Shamsuddin AF, Basir R. Central composite design for formulation and optimization of solid lipid nanoparticles to enhance oral bioavailability of acyclovir. Molecules. 2021;26(18):5432.
21. Vengala PA, Subrahmanyam CV, Gangaraju M. *In vitro* and *in vivo* evaluation of piroxicam loaded ceramic nanoparticles. Int J Pharmaceut Sci Res. 2016;7:303-9.
22. Patel JK, Patel KN, Patel HK, Patel BA, Patel PA. Aquasomes: a self-assembling nanobiopharmaceutical carrier system for bio-active molecules: a review. Int J Pharm Res. 2012;1(1):11-21.
23. Kallingal A, Suresh AM. Aquasomes Nanocarriers for Delivery of Small Molecules. Int J Med Phar Sci. 2021;11(7):1-7.
24. Nikhil Shrivastava and Sandeep Jain. Formulation and Evaluation of Aquasomes of Dapsone for Improved Solubility. Eur J Biomed Pharm Sci. 2023;10(7):243-8.
25. Shirole PL, Sanap DP, Jadhav KR. Aquasomes: A Nanoparticulate approach for the Delivery of Drug. Res J Pharm Technol. 2023;16(12):6081-6.
26. Padmavathi S, Nandini V, Manasa B, Manasa J, Dharani P, Roshanara S. A comprehensive knowledge on the Aquasome novel drug delivery system. J Cardiovasc Dis Res. 2021;12(5):1335-9.

Cite this article: Katharotiya K, Katharotiya D, Jha LL. Screening and Optimization of Various Formulation and Processing Parameters for the Development of Aquasomes. Indian J of Pharmaceutical Education and Research. 2025;59(4):1285-92.