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"FORMULATION AND IN-VITRO CHARACTERIZATION OF CHLORPHENIRAMINE MALEATE-LOADED MUCOADHESIVE MICROSPHERES INCORPORATED IN IN-SITU NASAL GEL"

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ABSTRACT

Background: Nasal drug delivery offers rapid onset of action and avoids first-pass metabolism; however, conventional formulations suffer from rapid mucociliary clearance. Mucoadhesive microspheres incorporated into in-situ nasal gels can enhance nasal residence time and sustain drug release. Chlorpheniramine maleate, a widely used antihistamine, can benefit from such delivery to reduce systemic side effects and improve local efficacy. **Methods:** Chlorpheniramine maleate-loaded mucoadhesive microspheres were prepared using chitosan as the mucoadhesive polymer and sodium tripolyphosphate (TPP) as the cross-linking agent via the emulsification cross-linking method. Formulations F1–F9 were evaluated for % yield, drug entrapment efficiency, particle size, swelling index, mucoadhesion, and in-vitro drug release. The optimized microspheres were incorporated into an in-situ nasal gel and characterized for pH, gel strength, viscosity, and mucoadhesive force. **Results:** The microspheres exhibited % yield of 58.9–77.0%, drug entrapment efficiency of 48.6–72.3%, particle sizes of 16.2–35.0 µm, swelling indices of 22.5–56.2%, and mucoadhesion of 45.2–75.6%. In-vitro release studies showed sustained drug release over 8 hours, with the optimized gel displaying suitable pH (6.0–6.3), gel strength, viscosity, and mucoadhesive force, ensuring compatibility with nasal mucosa. **Conclusion:** The study demonstrates that Chlorpheniramine maleate-loaded mucoadhesive microspheres incorporated into in-situ nasal gels are a promising approach for sustained intranasal drug delivery, offering enhanced residence time, improved bioavailability, and reduced systemic side effects.

Keywords
Chlorpheniramine maleate, Mucoadhesive microspheres, In-situ nasal gel, Chitosan, Sodium tripolyphosphate, Intranasal drug delivery

I. INTRODUCTION

Nasal drug delivery has emerged as a promising route for systemic and local therapy due to the high vascularization, large surface area, and avoidance of first-pass metabolism. However, conventional nasal formulations often suffer from rapid mucociliary clearance, resulting in limited drug absorption and reduced therapeutic efficacy. To overcome these challenges, mucoadhesive drug delivery systems have been extensively investigated to enhance residence

time on the nasal mucosa and achieve sustained drug release. [1,2]

Chlorpheniramine maleate, a first-generation antihistamine, is widely used for the treatment of allergic rhinitis and nasal congestion. Oral administration of Chlorpheniramine maleate is associated with systemic side effects such as sedation and gastrointestinal disturbances. Nasal delivery of Chlorpheniramine maleate can provide rapid onset of action, localize therapeutic effect, and reduce systemic exposure, making it an ideal candidate for intranasal formulations. [3,4]

Mucoadhesive microspheres are polymer-based particulate systems capable of adhering to the nasal mucosa, providing controlled and targeted drug release. Incorporation of these microspheres into in-situ gelling systems further enhances nasal retention by forming a gel upon contact with nasal fluids, preventing rapid clearance. Chitosan, a natural cationic polymer, is commonly employed due to its biocompatibility, biodegradability, and mucoadhesive properties. Sodium tripolyphosphate (TPP) is often used as a cross-linking agent to stabilize chitosan microspheres. [5,6]

The present study focuses on the formulation and in-vitro characterization of Chlorpheniramine maleate-loaded mucoadhesive microspheres incorporated in an in-situ nasal gel, aiming to achieve prolonged residence time, improved drug bioavailability, and sustained therapeutic effect. The work involves systematic optimization of polymer concentration, cross-linking, and formulation parameters, followed by evaluation of particle size, drug entrapment efficiency, muco-adhesion, swelling, gelation, and in-vitro drug release. [6,7]

II. MATERIALS AND METHOD

Preliminary Evaluation of Chlorpheniramine Maleate

The preliminary evaluation of Chlorpheniramine maleate was carried out to confirm its physical characteristics and purity. The drug was observed as a white to off-white crystalline powder, which is typical for its pure form, and odorless, indicating the absence of volatile impurities. Solubility studies were performed in various solvents, showing that the drug is freely soluble in water and dilute hydrochloric acid, and soluble in alcohols such as ethanol and methanol, confirming compatibility with aqueous and hydroalcoholic formulations. The melting point was determined to be 130–135°C, consistent with pharmacopeial standards, thus confirming the identity and purity of Chlorpheniramine maleate and its suitability for intranasal delivery systems like mucoadhesive microspheres and in-situ gels.[8]

Solubility Profile of Chlorpheniramine Maleate

The quantitative solubility of Chlorpheniramine maleate was determined in different solvents to understand its solubility behavior for formulation development. The drug was freely soluble in water (26 mg/mL) and 0.1 N HCl (32 mg/mL), moderately soluble in ethanol (13 mg/mL) and methanol (19 mg/mL), and practically insoluble in non-polar solvents like ether and benzene. Slight solubility in 0.1 N NaOH (3.5 mg/mL) suggested reduced dissolution in basic environments. These findings confirm that the drug is predominantly polar, making it suitable for incorporation into aqueous-based mucoadhesive microspheres and in-situ gels.[9]

Calibration Curve of Chlorpheniramine Maleate

The **spectrophotometric method** was used to construct the calibration curve over a concentration range of **2–12 µg/mL**. Absorbance was measured at the **λmax specific for Chlorpheniramine maleate**, showing a linear increase with concentration. The low standard deviations (±0.001–0.004) indicate **excellent reproducibility**. The linearity confirms that **Beer's law was obeyed**, validating the method

for **quantitative determination of drug content, entrapment efficiency, and in-vitro release** in microsphere formulations.[10]

Preparation of Mucoadhesive Intranasal Microspheres:

The mucoadhesive intranasal microspheres of Chlorpheniramine Maleate (F1–F9) were prepared by the emulsification cross-linking method using chitosan as the mucoadhesive polymer and sodium tripolyphosphate (TPP) as the cross-linking agent. Accurately weighed quantities of chitosan were dissolved in 1% v/v acetic acid to obtain polymer solutions of different concentrations (1.0%, 1.5%, and 2.0% w/w) corresponding to formulations F1–F9. Chlorpheniramine Maleate (10% w/w) was dispersed uniformly in the chitosan solution under magnetic stirring. The dispersion was then added dropwise into light liquid paraffin (50 mL) containing 0.5% v/v Span 80 as an emulsifying agent, under continuous stirring to form a stable emulsion. Aqueous TPP solution (0.5–1.5% w/v) was added slowly to induce cross-linking and microsphere formation. Stirring was continued for a specific duration to complete the cross-linking process, after which the microspheres were separated by filtration, washed repeatedly with n-hexane to remove oil residues, and dried at room temperature. The dried microspheres were finally blended with 2% w/v mannitol as a cryoprotectant and stored in a desiccator until further evaluation.[10,11]

Table 1: Composition of Mucoadhesive Intranasal Microspheres (F1–F9)

Ingredients / Parameters	F1	F2	F3	F4	F5	F6	F7	F8	F9
Chlorpheniramine Maleate (% w/w)	10	10	10	10	10	10	10	10	10
Chitosan (% w/w)	1.0	1.0	1.0	1.5	1.5	1.5	2.0	2.0	2.0
TPP (% w/v)	0.5	1.0	1.5	0.5	1.0	1.5	0.5	1.0	1.5
Span 80 (% v/v)	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Liquid Paraffin (mL)	50	50	50	50	50	50	50	50	50

Evaluation of Mucoadhesive Microspheres

Mucoadhesive microspheres of Chlorpheniramine maleate (F1–F9) were prepared by emulsification cross-linking using chitosan as the polymer and TPP as the cross-linker. The formulations were evaluated for % yield, drug entrapment efficiency, particle size, swelling index, mucoadhesion, and in-vitro drug release. The % yield and drug entrapment efficiency increased progressively with higher polymer and cross-linker concentrations, indicating efficient microsphere formation and drug loading. Particle size increased from 16.2 ± 1.1 µm (F1) to 35.0 ± 1.9 µm (F9), showing the effect of polymer and cross-linker on particle growth. The swelling index increased proportionally, enhancing mucoadhesion and sustaining drug release. Correspondingly, % mucoadhesion after 1 h increased from 45.2 ± 2.4% to 75.6 ± 1.6%, indicating stronger adhesion and prolonged residence time. The in-vitro cumulative drug release exhibited controlled release over 8 h, decreasing from 78.0 ± 3.0% (F1) to 40.8 ± 3.0% (F9), confirming that formulation parameters effectively modulate sustained release. These results suggest that microsphere properties can be optimized to balance mucoadhesion, particle size, and release kinetics for efficient intranasal delivery.[12-18]

III. RESULTS AND DISCUSSION

Preliminary Evaluation of Chlorpheniramine Maleate

The preliminary evaluation of Chlorpheniramine maleate revealed that the drug is a white to off-white crystalline powder, which is typical for its pure form, and odorless, indicating the absence

of volatile impurities. The solubility study demonstrated that the drug is freely soluble in water and soluble in alcohol, suggesting good compatibility with aqueous and hydroalcoholic formulations. Its melting point of 130–135°C corresponds with the standard range reported in pharmacopeial references, confirming the identity and purity of the compound. These characteristics indicate that Chlorpheniramine maleate possesses suitable physicochemical properties for formulation into intranasal delivery systems such as mucoadhesive microspheres and in-situ gels.

Table 2: Preliminary Evaluation of Chlorpheniramine Maleate

Sr. No.	Parameter	Observation
1	Appearance	White to off-white crystalline powder
2	Odor	Odorless
3	Melting Point	130–135°C

Solubility Profile of Chlorpheniramine Maleate

The solubility profile of Chlorpheniramine maleate indicates that the drug is freely soluble in water (26 mg/mL) and dilute hydrochloric acid (32 mg/mL), demonstrating its good solubility in polar aqueous media, which is advantageous for nasal formulations. It is moderately soluble in ethanol (13 mg/mL) and methanol (19 mg/mL), suggesting compatibility with hydroalcoholic systems. The drug shows limited solubility in chloroform (2 mg/mL) and acetone (2 mg/mL) and is practically insoluble in ether (0.1 mg/mL) and benzene (0.1 mg/mL), indicating low solubility in non-polar solvents. Slight solubility in 0.1 N NaOH (3.5 mg/mL) suggests reduced dissolution in basic environments. These results confirm that Chlorpheniramine maleate is predominantly polar and ionizable, which supports its suitability for incorporation into aqueous-based mucoadhesive microspheres and in-situ nasal gels.

Table 3: Solubility Profile of Chlorpheniramine Maleate

Sr. No.	Solvent	Quantitative Solubility (mg/mL)	Descriptive Solubility
1	Water	26 mg/mL	Freely soluble
2	Ethanol (95%)	13 mg/mL	Soluble
3	Methanol	19 mg/mL	Soluble
4	Chloroform	2 mg/mL	Slightly soluble
5	Ether	0.1 mg/mL	Practically insoluble
6	Acetone	2 mg/mL	Slightly soluble
7	Benzene	0.1 mg/mL	Insoluble
8	(0.1 N HCl)	32 mg/mL	Freely soluble
9	(0.1 N NaOH)	3.5 mg/mL	Slightly soluble

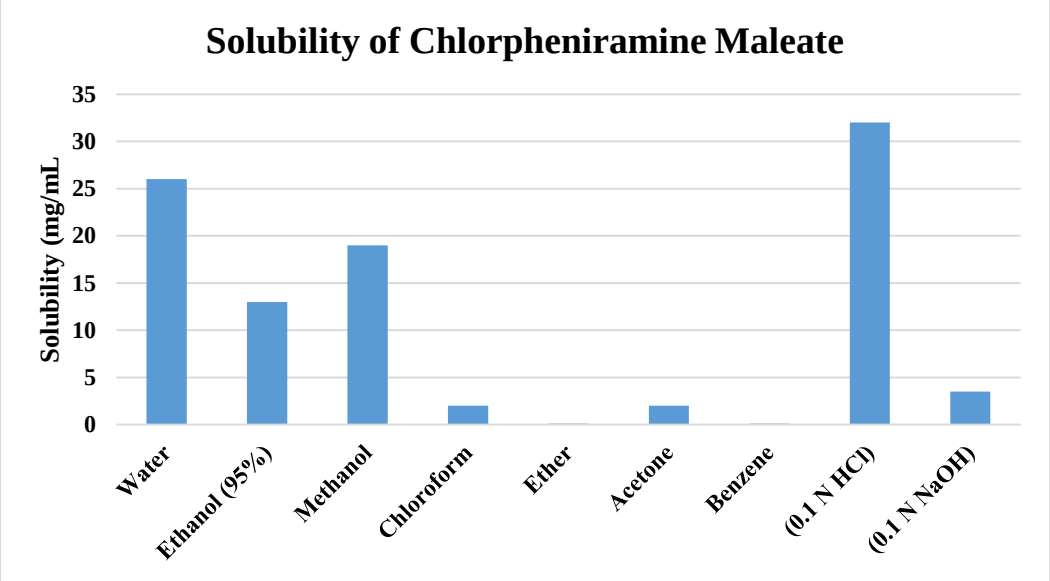

Fig 1: Solubility of Chlorpheniramine Maleate

Calibration curve of Chlorpheniramine maleate

The calibration curve of Chlorpheniramine maleate was constructed over a concentration range of 2–12 µg/mL, showing a linear increase in absorbance with concentration. The measured absorbance values (0.102–0.613 nm) demonstrated excellent reproducibility, as reflected by the low standard deviations (±0.001–0.004), indicating the reliability of the spectrophotometric method. This linear relationship confirms that Beer's law was obeyed within the selected range, validating the method for quantitative determination of Chlorpheniramine maleate in microsphere formulations and in-vitro release studies. The calibration curve provides a critical tool for assessing drug content, entrapment efficiency, and release kinetics in subsequent formulation evaluations.

Table 4: Calibration Curve of Chlorpheniramine Maleate

S. No.	Concentration (µg/mL)	Absorbance (nm)	Mean ± SD
1	2	0.102	0.102 ± 0.001
2	4	0.205	0.205 ± 0.002
3	6	0.306	0.306 ± 0.003
4	8	0.407	0.407 ± 0.002
5	10	0.510	0.510 ± 0.004
6	12	0.613	0.613 ± 0.003

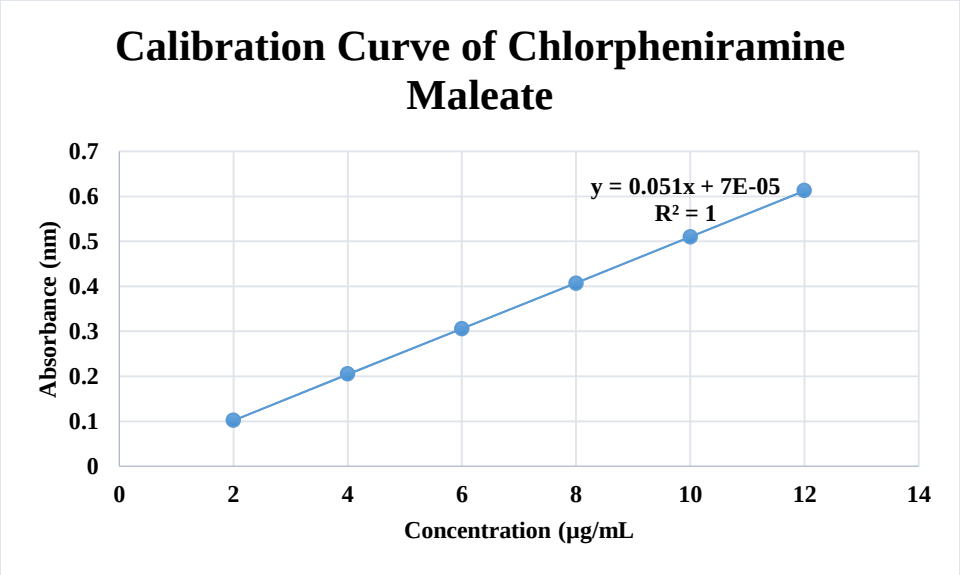

Fig 2: Calibration Curve of Chlorpheniramine Maleate

Evaluation of Mucoadhesive Microspheres

The evaluation of Chlorpheniramine maleate-loaded mucoadhesive microspheres (F1–F9) demonstrated a progressive increase in % yield and drug entrapment efficiency with higher polymer and cross-linker concentrations, indicating efficient microsphere formation and drug incorporation. The average particle size increased from 16.2 ± 1.1 µm (F1) to 35.0 ± 1.9 µm (F9), reflecting the influence of chitosan and TPP levels on particle growth. The swelling index showed a proportional increase, suggesting enhanced water uptake with larger and more cross-linked microspheres, which is beneficial for mucoadhesion and sustained release. Correspondingly, % mucoadhesion after 1 h increased from 45.2 ± 2.4% to 75.6 ± 1.6%, indicating stronger adhesion to the nasal mucosa and prolonged residence time. The in-vitro cumulative drug release displayed a controlled release pattern, decreasing from 78.0 ± 3.0% in F1 to 40.8 ± 3.0% in F9 over 8 h, confirming that higher polymer and cross-linker concentrations effectively sustain drug release. Overall, these results suggest that formulation parameters can be optimized to balance mucoadhesion, particle size, and release kinetics for effective nasal delivery.

Table 5: Evaluation of Mucoadhesive Microspheres

Parameter	F1	F2	F3	F4	F5	F6	F7	F8	F9
% Yield	58.9 ± 1.6	60.7 ± 1.5	62.4 ± 1.7	66.2 ± 1.6	68.0 ± 1.5	69.7 ± 1.8	73.0 ± 1.7	75.4 ± 1.6	77.0 ± 1.8
Drug Entrapment Efficiency (EE %)	48.6 ± 2.2	52.0 ± 2.0	55.1 ± 2.3	58.0 ± 2.1	61.4 ± 2.0	64.7 ± 2.2	67.5 ± 2.1	70.0 ± 2.0	72.3 ± 2.3
Average particle size (µm)	16.2 ± 1.1	17.8 ± 1.2	19.6 ± 1.3	22.0 ± 1.4	24.1 ± 1.5	26.7 ± 1.6	29.9 ± 1.7	32.1 ± 1.8	35.0 ± 1.9

% Swelling index (in pH 6.4, 1 h)	22.5 ± 1.0	25.0 ± 1.1	28.8 ± 1.2	33.0 ± 1.3	37.4 ± 1.4	41.8 ± 1.5	46.7 ± 1.6	51.0 ± 1.7	56.2 ± 1.8
% Mucoadhesion after 1 hr	45.2 ± 2.4	49.0 ± 2.3	53.1 ± 2.2	57.6 ± 2.1	61.8 ± 2.0	65.0 ± 1.9	68.9 ± 1.8	72.2 ± 1.7	75.6 ± 1.6
In-vitro cumulative drug release (%) at 8 h	78.0 ± 3.0	74.2 ± 2.8	70.5 ± 2.9	66.0 ± 2.7	61.2 ± 2.6	56.8 ± 2.5	51.5 ± 2.6	46.0 ± 2.8	40.8 ± 3.0

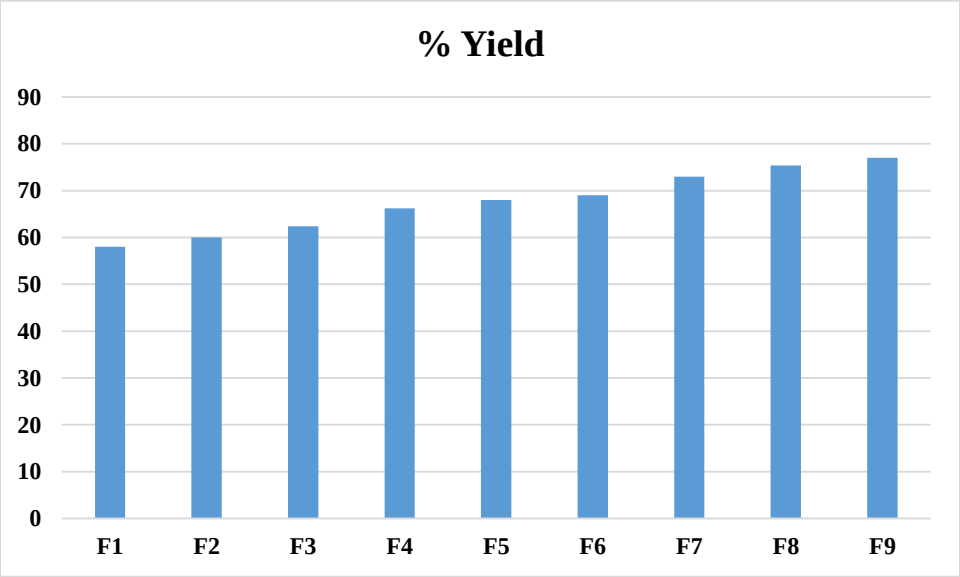

Fig 3: % Yield

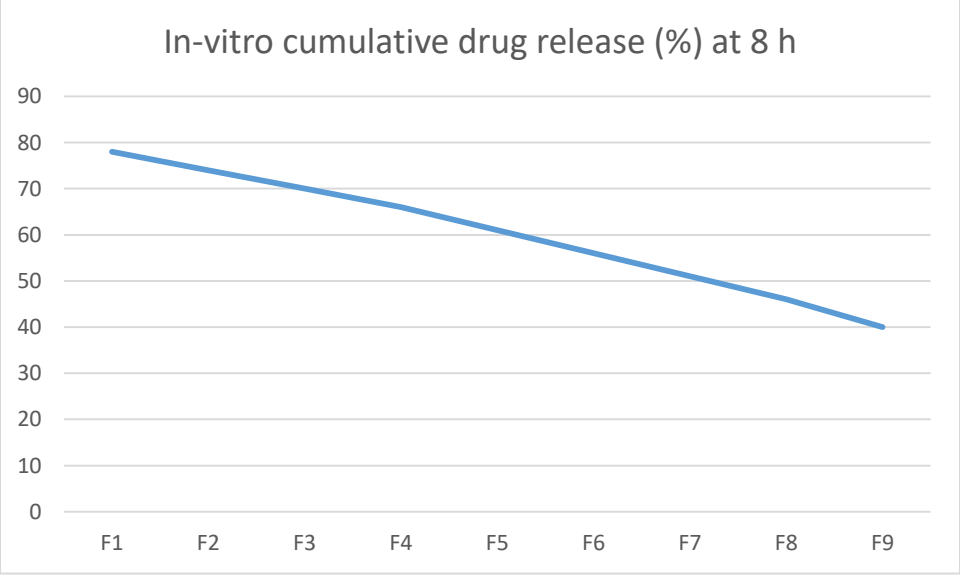

Fig 4: In-vitro cumulative drug release (%) at 8 h

Evaluation Parameters of Microsphere-Loaded In-situ Gel

The evaluation of Chlorpheniramine maleate-loaded microsphere in-situ gels (F1–F3) demonstrated that all formulations had solution pH values of 5.7–5.8 and gel pH values of 6.1–6.2, indicating compatibility with the nasal mucosa and minimal risk of irritation. Gel strength increased progressively from 38.5 ± 1.5 sec (F1) to 43.2 ± 1.5 sec (F3), suggesting the formation of robust gels capable of resisting mucociliary clearance. The viscosity of the formulations also increased upon gelation, from 110–130 cps in solution to 780–860 cps in gel form, ensuring adequate consistency for nasal retention. Correspondingly, the mucoadhesive force increased from 28.5 ± 2.0 to 31.5 ± 2.0 dyne/cm² across the formulations, reflecting stronger adhesion to the nasal mucosa and prolonged residence time. These findings indicate that the microsphere-loaded in-situ gels possess suitable physicochemical and mechanical properties for intranasal delivery, providing sustained drug release while maintaining nasal compatibility.

Table 6: Evaluation Parameters of Microsphere-Loaded In-situ Gel

Parameter	F1	F2	F3
pH (Solution)	5.7 ± 0.1	5.8 ± 0.1	5.8 ± 0.1
pH (Gel)	6.1 ± 0.2	6.2 ± 0.2	6.2 ± 0.2
Gel strength (sec)	38.5 ± 1.5	41.0 ± 1.6	43.2 ± 1.5
Viscosity (cps, Solution)	110 ± 5	120 ± 6	130 ± 5
Viscosity (cps, Gel)	780 ± 15	820 ± 14	860 ± 16
Mucoadhesive force (dyne/cm²)	28.5 ± 2.0	30.0 ± 2.1	31.5 ± 2.0

IV. CONCLUSION

The study successfully formulated Chlorpheniramine maleate-loaded mucoadhesive microspheres incorporated into an in-situ nasal gel with optimized drug entrapment, particle size, swelling, mucoadhesion, and sustained drug release. The optimized formulation demonstrated good physical stability, nasal pH compatibility, and enhanced in-vitro release, indicating its potential as an effective intranasal delivery system for prolonged therapeutic action and improved patient compliance.

REFERENCES

1. Achmad, N. A., Tuna, R. W., Kurniawan, I., Khairiyah, Asaf, M. B., Rahman, L., Manggau, M. A., Aliyah, Dominguez-Robles, J., Aswad, M., & Permana, A. D. (2025). Development of Thermosensitive Mucoadhesive Gel Based Encapsulated Lipid Microspheres as Nose-to-Brain Rivastigmine Delivery System. *Langmuir : the ACS journal of surfaces and colloids*, *41*(1), 314–328. https://doi.org/10.1021/acs.langmuir.4c03530
2. Tuna, R. W., Achmad, N. A., Kurniawan, I., Khairiyah, Asaf, M. B., Sapiun, Z., Himawan, A., Dominguez-Robles, J., Aswad, M., & Permana, A. D. (2025). Development of thermosensitive mucoadhesive gel incorporated lipid microspheres of donepezil for enhanced nose-to-brain delivery. *Journal of biomaterials science. Polymer edition*, 1–28. Advance online publication.

https://doi.org/10.1080/09205063.2025.2492455

3. Szekalska, M., Amelian, A., & Winnicka, K. (2015). Alginate microspheres obtained by the spray drying technique as mucoadhesive carriers of ranitidine. *Acta pharmaceutica (Zagreb, Croatia)*, *65*(1), 15–27. https://doi.org/10.1515/acph-2015-0008
4. Zhang, T., Zhang, C., Agrahari, V., Murowchick, J. B., Oyler, N. A., & Youan, B. B. (2013). Spray drying tenofovir loaded mucoadhesive and pH-sensitive microspheres intended for HIV prevention. *Antiviral research*, *97*(3), 334–346. https://doi.org/10.1016/j.antiviral.2012.12.019
5. Khalid, S., Abbas, G., Hanif, M., Shah, S., Shah, S. N. H., Jalil, A., Yaqoob, M., Ameer, N., & Anum, A. (2020). Thiolated sodium alginate conjugates for mucoadhesive and controlled release behavior of metformin microspheres. *International journal of biological macromolecules*, *164*, 2691–2700. https://doi.org/10.1016/j.ijbiomac.2020.08.116
6. Karavana, S. Y., Şenyiğit, Z. A., Çalışkan, Ç., Sevin, G., Özdemir, D. İ., Erzurumlu, Y., Şen, S., & Baloğlu, E. (2018). Gemcitabine hydrochloride microspheres used for intravesical treatment of superficial bladder cancer: a comprehensive in vitro/ex vivo/in vivo evaluation. *Drug design, development and therapy*, *12*, 1959–1975. https://doi.org/10.2147/DDDT.S164704
7. Jose, S., Ansa, C. R., Cinu, T. A., Chacko, A. J., Aleykutty, N. A., Ferreira, S. V., & Souto, E. B. (2013). Thermo-sensitive gels containing lorazepam microspheres for intranasal brain targeting. *International journal of pharmaceutics*, *441*(1-2), 516–526. https://doi.org/10.1016/j.ijpharm.2012.10.049
8. Meskelis, L., F Agondi, R., Duarte, L. G. R., de Carvalho, M. D., Sato, A. C. K., & Picone, C. S. F. (2024). New approaches for modulation of alginate-chitosan delivery properties. *Food research international (Ottawa, Ont.)*, *175*, 113737. https://doi.org/10.1016/j.foodres.2023.113737
9. Yadav, S. K., Khan, G., Bansal, M., Thokala, S., Bonde, G. V., Upadhyay, M., & Mishra, B. (2018). Multiparticulate based thermosensitive intra-pocket forming implants for better treatment of bacterial infections in periodontitis. *International journal of biological macromolecules*, *116*, 394–408. https://doi.org/10.1016/j.ijbiomac.2018.04.179
10. Díaz, A. G., Quinteros, D. A., Paolicchi, F. A., Rivero, M. A., Palma, S. D., Pardo, R. P., Clausse, M., Zylberman, V., Goldbaum, F. A., & Estein, S. M. (2019). Mucosal immunization with polymeric antigen BLSOmp31 using alternative delivery systems against Brucella ovis in rams. *Veterinary immunology and immunopathology*, *209*, 70–77. https://doi.org/10.1016/j.vetimm.2019.02.005
11. Cruz-Neves, S., Shirosaki, Y., Miyazaki, T., & Hayakawa, S. (2017). Characterization and degradation study of chitosan-siloxane hybrid microspheres synthesized using a microfluidic approach. *Materials science & engineering. C, Materials for*

biological applications, *81*, 571–579. https://doi.org/10.1016/j.msec.2017.08.035

12. Denora, N., Lopedota, A., Perrone, M., Laquintana, V., Iacobazzi, R. M., Milella, A., Fanizza, E., Depalo, N., Cutrignelli, A., Lopalco, A., & Franco, M. (2016). Spray-dried mucoadhesives for intravesical drug delivery using N-acetylcysteine- and glutathione-glycol chitosan conjugates. *Acta biomaterialia*, *43*, 170–184. https://doi.org/10.1016/j.actbio.2016.07.025
13. Bera, H., Kandukuri, S. G., Nayak, A. K., & Boddupalli, S. (2015). Alginate-sterculia gum gel-coated oil-entrapped alginate beads for gastroretentive risperidone delivery. *Carbohydrate polymers*, *120*, 74–84. https://doi.org/10.1016/j.carbpol.2014.12.009
14. Wiltsey, C., Christiani, T., Williams, J., Scaramazza, J., Van Sciver, C., Toomer, K., Sheehan, J., Branda, A., Nitzl, A., England, E., Kadlowec, J., Iftode, C., & Vernengo, J. (2015). Thermogelling bioadhesive scaffolds for intervertebral disk tissue engineering: preliminary in vitro comparison of aldehyde-based versus alginate microparticle-mediated adhesion. *Acta biomaterialia*, *16*, 71–80. https://doi.org/10.1016/j.actbio.2015.01.025
15. Tahtat, D., Mahlous, M., Benamer, S., Khodja, A. N., Oussedik-Oumehdi, H., & Laraba-Djebari, F. (2013). Oral delivery of insulin from alginate/chitosan crosslinked by glutaraldehyde. *International journal of biological macromolecules*, *58*, 160–168. https://doi.org/10.1016/j.ijbiomac.2013.03.064
16. Maestrelli, F., Cirri, M., Mennini, N., Bragagni, M., Zerrouk, N., & Mura, P. (2012). Influence of cross-linking agent type and chitosan content on the performance of pectinate-chitosan beads aimed for colon-specific drug delivery. *Drug development and industrial pharmacy*, *38*(9), 1142–1151. https://doi.org/10.3109/03639045.2011.641566
17. He, Q., Liao, Y., Zhang, H., Sun, W., Zhou, W., Lin, J., Zhang, T., Xie, S., Wu, H., Han, J., Zhang, Y., Wei, W., Li, C., Hong, Y., Shen, W., & Ouyang, H. (2024). Gel microspheres enhance the stemness of ADSCs by regulating cell-ECM interaction. *Biomaterials*, *309*, 122616. https://doi.org/10.1016/j.biomaterials.2024.122616
18. Jiang, W., Gao, X., Wang, Q., Chen, Y., Li, D., Zhang, X., & Yang, X. (2023). The Modified Exenatide Microspheres: PLGA-PEG-PLGA Gel and Zinc-Exenatide Complex Synergistically Reduce Burst Release and Shorten Platform Stage. *AAPS PharmSciTech*, *24*(8), 251. https://doi.org/10.1208/s12249-023-02705-6