Robust and Easy-to-Use Lyophilized Magnetic Particle Beads

Abstract

Lyophilization, or freeze-drying, has gained prominence in the formulation of In Vitro Diagnostic (IVD) assays. Magnetic nanoparticles (MNPs) play a pivotal role in nucleic acid isolation and biomolecular interactions essential for IVD applications, specifically molecular diagnostics (MDx). The use of lyophilized MNPs offers several advantages, including enhanced stability for storage, improved handling, ease of transport, and expedited reconstitution, all while preserving their functional properties over time. Evik Diagnostics (Evik Dx) possesses extensive expertise in the development of MNPs in lyophilized bead format. This white paper presents a case study on the lyophilization of iron oxide MNPs, with a focus on assessing their physical and chemical stability before and after the lyophilization process.

Excipient Selection

The selection of excipients is a critical factor influencing the lyophilization process and the quality of the final product. The properties of the lyophilized MNPs can be significantly affected by the nature and concentration of excipients. Key attributes influenced include:

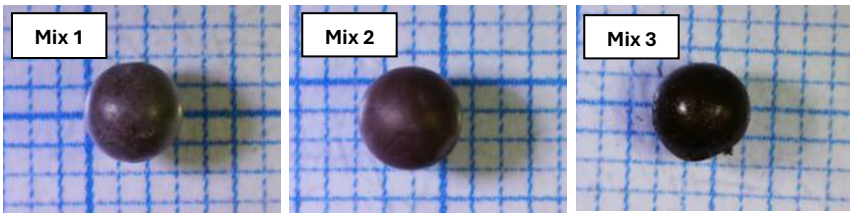
1. Robustness
2. Residual moisture content
3. Bead-to-bead variability
4. Dissolution/reconstitution kinetics
5. Particle size and polydispersity
6. Long-term stability

In this study, we evaluated several excipient mixtures for the lyophilization of iron oxide MNPs with -COOH functionality. Here, we present data for three excipient mixtures, encompassing both carbohydrate-based and non-carbohydrate-based excipients. MNPs were suspended in excipient solutions and dispensed into liquid nitrogen to achieve a concentration of 1 mg per lyophilized bead, followed by the lyophilization process. The dispensing was performed with continuous agitation to prevent sedimentation of the formulations.

Quality Assessment of Magnetic Particle Lyo-Beads

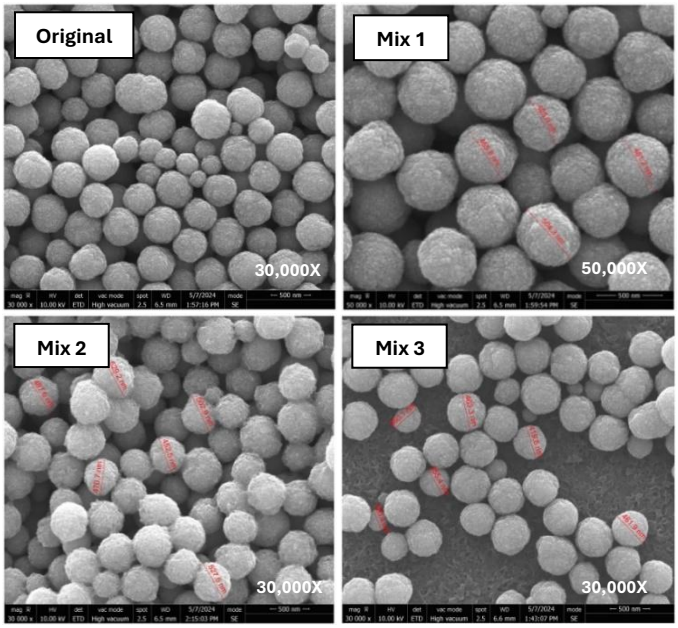
The overall morphology of the lyophilized beads was assessed using microscopy techniques. All beads exhibited uniform visual quality and homogeneity. Residual moisture content was

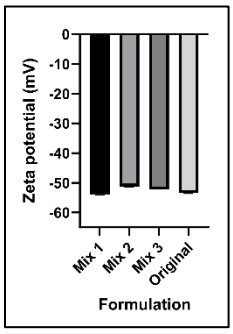
quantitatively measured, revealing that all three formulations maintained moisture levels within the acceptable range (<4%), with the lowest residual moisture observed in Mix 2 (1.5%). To evaluate robustness, crush force testing was conducted using a force gauge; all lyophilized beads demonstrated a crush force exceeding 0.4 N, indicative of suitable hardness for assembly in manufacturing of devices and practical handling.

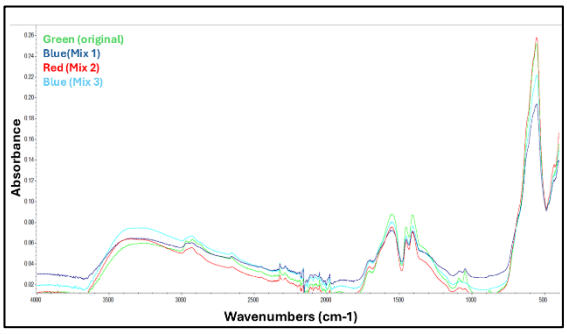


Physical and Chemical Properties of Magnetic Particles: Pre- vs. Post-Lyophilization

To comprehensively assess the physical and chemical properties of the lyophilized beads, we employed techniques such as Scanning Electron Microscopy (SEM), Dynamic Light Scattering (DLS), and Fourier-Transform Infrared Spectroscopy (FT-IR). Both SEM and DLS analyses indicated that freeze-drying did not significantly alter the size of the iron oxide particles, with all samples exhibiting a Z-average size of 500-600 nm, consistent with the original MNPs size prior to lyophilization. SEM images revealed similar morphologies and particle sizes (~500 nm) across the three lyophilized formulations and the original samples. The polydispersity index (PDI) was found to be <0.2 for both the original and resuspended lyophilized beads, confirming that freeze-drying under the studied conditions did not introduce significant heterogeneity in particle size distribution. Zeta potential measurements yielded values of approximately -50 mV for all samples, indicating a strong negative surface charge that remained unaffected by the freeze-drying process. FT-IR spectra of the three lyophilized formulations matched that of the original suspension, with Mix 1 showing the most similarity, suggesting that introduction of excipients and lyophilization did not induce any significant chemical







alterations in the functional groups of the MNPs. Our post-lyo thermal analysis suggested the lyophilized beads can be stored at room temperature with no impact on their integrity.

Conclusion

This case study demonstrates lyophilization as a successful technique to produce user-friendly and robust MNP beads with characteristics comparable to their original liquid suspensions. In contrast to traditional MNP suspensions, lyophilized beads formulated with appropriate excipients can be stored at room temperature and do not require complex concentration preparations, as each bead contains a defined mass of MNPs. Therefore, lyophilized MNP beads facilitate quicker preparation and assay setup, streamline workflows, and minimize the risk of contamination. All these make lyophilized MNP beads well-suited for high-throughput and automated assay applications, facilitating rapid, reproducible, and efficient techniques for applications such as Point of Care MDx.