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Effect of agitation in magneto-assay performance

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Highlights

- MPO binding at magnetic beads is faster and more sensitive than ELISA.
- MP mixing conditions have significant effect on magneto-immunobinding efficiency.
- Higher sample volumes provide higher target recovery and signal under rotation.
- Vortexing generates the most efficient target recovery rate for low volumes.
- Efficient automation of magneto-assays may need a combination of mixing types.

ABSTRACT

Magnetic particles (MP) are extensively used to improve immunocapture and immunodetection. Compared to two-dimensional surfaces, MP provide wider active areas, efficient mixing with the sample, and target separation from other sample components. Nevertheless, MP handling is more tedious than classical techniques, such as ELISA, in which tens/hundreds of samples are simultaneously processed. Accordingly, MP handling automation seems essential for the exploitation of magneto-detection in real diagnostics. This entails implementation of appropriate agitation strategies to prevent MP sedimentation and guarantee maximal target binding efficiency, minimal non-specific adsorption, and acceptable result reproducibility. Attempts to automate magnetic-based agitation have been reported. However, we could not find studies comparing the effect of different agitation procedures in magneto-immunobinding efficiency, which could be useful to define the most appropriate strategy for integration of MP handling in automated detection platforms.

Here, we optimized a classical ELISA for detection of myeloperoxidase (MPO) that took 3 h and generated a limit of detection (LOD) of 4,87 ng.mL⁻¹. This assay was then formatted into a 30-min magneto-immunoassay that had an LOD of 1,65 ng.mL⁻¹. MPO magneto-immunodetection was next performed under different agitation conditions. The results showed that sample volume had a high effect in the incubations performed under rotation, with signals 50% higher in 0,5-1 mL than in 100 µL samples. On the contrary, vortex agitation generated similar signals for the different volumes tested (100 µL, 500 µL and 1 mL), which entailed a 50% enhancement in target recovery in 100 µL samples compared to rotation. This demonstrated that sample volume had different effect in magneto-immunobinding efficiency under different agitation conditions.
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