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Artificial Intelligence-Aided Massively Parallel Spectroscopy for Bioaffinity Assays and Droplet Microfluidics

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Summary

Single-molecule heterogeneous immunochemical assays with photon-upconversion labels (UCNPs) appeared as an ultrasensitive advancement of immunochemical methods possessing femtomolar detection limits [1]. However, the method relies on separation on solid surfaces, which complicates its adaptation for microfluidic systems. Here, we discuss massively parallel spectroscopy (MPS) as a new tool for single-molecule bioaffinity assays in a free dispersion without a need for any separation [2]. Compared to cross-correlation spectroscopy – a method, that detects single-molecules by confocal detectors, MPS operates in 1000 times larger detection volumes. This provides opportunities for faster analysis and low detection limits. To investigate the capabilities of MPS in bioaffinity assays, bioconjugated UCNPs with excitation in the near-infrared region (976 nm) are prepared as a model. The UCNPs are doped with either Tm^{3+} or Er^{3+} providing virtually background-free emission at 450 and 802 nm or 554 and 660 nm, respectively. These UCNPs are conjugated to biotinylated bovine serum albumin (Tm^{3+} doped) or streptavidin (Er^{3+} doped). The MPS data are processed by a specialized convolutional neural network, and the limit of detection (1.6 fmol L^{-1}) and linearity range (4.8 fmol L^{-1} to 40 pmol L^{-1}) for the bioconjugated UCNPs are estimated [2,3]. MPS is then used to observe the bioaffinity clustering of bioconjugated UCNPs. This observation is correlated with native electrophoresis and bioaffinity assays on microtiter plates. A competitive MPS bioaffinity assay for biotin with a limit of detection of 6.6 nmol L^{-1} is developed. MPS in complex biological matrices (cell culture medium) is performed without apparently increasing background. Compatibility with polydimethylsiloxane microfluidics is demonstrated by recording MPS from microdroplets containing UCNPs, which pass through a detection area in the channel of the microfluidic chip.

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References

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