

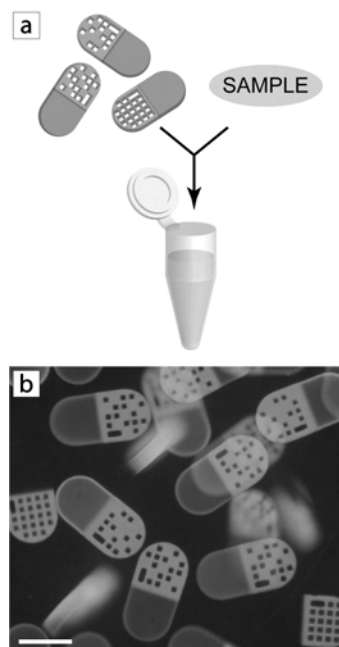
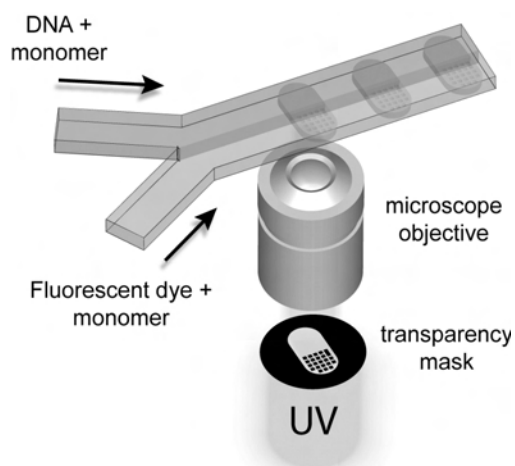
Barcoded Microparticles for Multiplexed Detection

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The detection of multiple targets in a single sample is important for many applications, including medical diagnostics, genotyping, and drug discovery. The current approaches to multiplexing, such as planar arrays (such as DNA microarrays) and suspension (particle-based) arrays, require expensive or cumbersome means of encoding, decoding, or functionalizing substrates. Currently, commercially available approaches for multiplexed analysis are cost-prohibitive for high sample throughput, low-cost applications such as bedside diagnostics.

We have developed a method [1], based on multifunctional barcoded particles, for the sensitive and accurate multiplexed detection of biomolecules. Our method is unique in that (1) we can fabricate, encode, and functionalize particles in a single step, (2) the particles are composed of poly(ethylene glycol) hydrogel to

increase both sensitivity and specificity, and (3) only a single fluorescent wavelength is required to decode the particles and quantify the corresponding targets. Using an efficient one-step method based on continuous-flow lithography, we synthesize microparticles with multiple functional regions (Figure 1). Each particle bears a fluorescent dot-pattern barcode (capable of providing over a million unique codes) to identify the target(s) it is looking for and one or more spatially separated regions containing a probe where those targets can bind and be detected via fluorescence. In this way, particles from a library can be mixed and incubated in a single sample to simultaneously detect many targets, such as DNA oligomers (Figure 2). The detection of targets is not only sensitive but also extremely specific due to the porous and bio-inert nature of the hydrogel structure that allows target molecules to diffuse and bind deep into the transparent particle surfaces.



▲ Figure 1: Schematic of one-step particle synthesis in a microfluidic device. Two monomer streams (one with a DNA probe and the other with a fluorescent dye) are flowed adjacently along a channel where they are repeatedly hit with burst of UV light shone through a microscope objective.

▲ Figure 2: (a) Schematic of multiplexed sample analysis. Particles (with unique barcodes corresponding to their target) are mixed and incubated in a single sample. (b) Fluorescence image of particles after incubation with DNA oligomers targets. Positive detection is indicated by fluorescence in the probe-region of the particles. Scalebar = 100 μm .

REFERENCES

- [1] D.C. Pregibon, M. Toner, and P.S. Doyle, "Multifunctional encoded particles for high-throughput biomolecule analysis," *Science*, vol. 315, no. 5817, pp. 1393-1396, Mar. 2007.