

# The Effects of Mutations on DNA as a Materials Assembly Tool.

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## Introduction

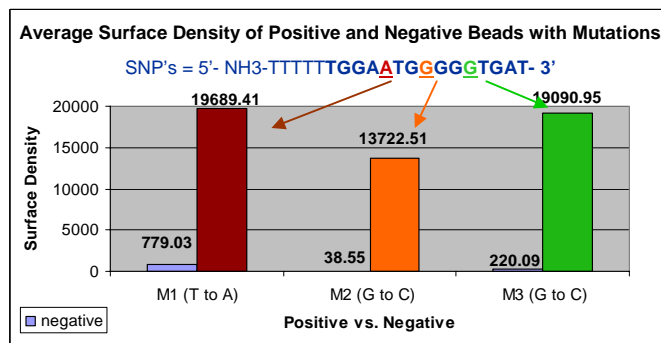
DNA is a useful materials assembly tool for the programmed assembly of colloidal particles. This technique was pioneered by Mirkin, et.al, using surface coupled DNA strands to assemble gold nanoparticles<sup>1</sup>. Recently, studies by Milam<sup>2</sup> and Kim<sup>3</sup> have investigated DNA assemblies using micron-sized polystyrene colloids. In each of these previous studies, perfectly matched sequences were used for hybridization and assembly purposes. My research expands upon these studies to investigate the effects that sequence mutations have on the hybridization activity of DNA coupled to one micron polystyrene microspheres. As part of this study, the effects of the location of the mutation on hybridization activity were also investigated.

## Experimental Procedure

Throughout the summer numerous experiments were carried out in Dr. Valeria Milam's bio-inspired colloidal assembly lab. Single stranded oligonucleotides carrying one base mutation (probe strand) were coupled to the surface of carboxylated beads. Multiple copies of the amine-terminated DNA (probe sequence) were covalently coupled to 1.04 micron carboxylated beads using EDAC. Fluorescently-labeled complementary oligonucleotides (target sequence) were then hybridized to the surface coupled probe strand. Data was collected on a flow cytometer using fluorescence standards to quantify the hybridization activity of fluorescently-labeled targets to the surface coupled probes with both perfect base pair matching and with a single nucleotide polymorphism in one of three locations.

## Results and Discussion

It was hypothesized, based on the variance in calculated melting temperatures and previous studies published by Guo et.al.<sup>4</sup>, that a mutation in the center of the probe strand would reduce hybridization activity with its target more than mutations at either end. After hybridizing fluorescent-labeled target strands to surface-coupled probe strands, flow cytometry was used to quantify the hybridization activity that occurred, and the surface densities for these target strands were found using calibrated standards. Flow cytometry revealed that hybridization did occur between the surface-coupled probe strands (carrying a mutation in one of three locations) and the fluorescently-labeled, soluble target strands of DNA.



**Figure 1.** Surface density of probe strands hybridized to mutated (positive) and noncomplementary (negative) targets are shown for each mutation site. The negative control was a target sequence identical and therefore noncomplementary to the probe sequence.

Preliminary data shown in Figure 1 suggests that a mutation in the center of a probe sequence results in a lower surface density than mutations near either end of a sequence, indicating that the location of a mutation on DNA does affect hybridization activity. Future work includes tuning the affinity of hybridization with mutated probe strands for colloidal particle assembly.

## References

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