

**Supplementary Figure 1.** (A) Detection of ligation products in a large excess of unligated probes. A 32bp synthetic ligation product (SLP), which was 5'-end biotinylated and 3'-end labeled with OG488, was used to mimic the ligation products. Sequence of SLP: Biotin – GTG CAC CTG ACT CCT GAG GAG AAG TCT GCC GT – OG488. Sequence of biotinylated discrimination probes (βDP1): Biotin – GTG CAC CTG ACT CCT G. Sequence of Oregon Green 488 labeled reporter probes (βRP): p AGG AGA AGT CTG CCG T – OG488. Ratio between probes and QD was kept at 30. The concentrations of the βDP1 and βRP probes were kept the same and R is the concentration ratio between the probes and SLPs. (B) Results of detecting synthetic ligation products in a varying excess of unligated probes. SLPs can be detected in a 200-fold excess of unligated probes (R=200). The detection time was 500 sec and error bars represent standard deviations from five measurements.