



Figure S5. Digital amplification strategies prior to probes validation. A) Emulsion based PCR; adapters are optional since CIPers already contain universal segments flanking the target of interest. The technology involves the inclusion of DNA and a primed magnetic bead in mineral oil (an emulsion), which allows for single molecule amplifications. Suitable upcoming methods for downstream sequence validation include the 454 Life Sciences platform (<http://www.454.com>), the Solexa platform (<http://www.illumina.com>), polony sequencing (<http://www.agentcourt.com>), Helicos Biosciences (<http://www.helicosbio.com>), or use of a resequencing array (GeneChip CustomSeq, www.affymetrix.com). B) Rolling circle amplification (RCA) with single-molecule detection (SMD). Digital quantification combines RCA and SMD in form of fluorescent-labeled target specific oligonucleotides. The amplified CIPers can be quantified using microfluidic analysis and visualized with a microscope for ultimate levels of quantification. The number of available fluorescent labels limits the degree of multiplexing.