

Electronic Supplementary Information

Preparation of HEK cells for genomic DNA extraction and DNA amplification by asymmetric PCR

Human Embryonic Kidney (HEK) 293T cells (ATCC-LGC Nr: CRL-11268) were seeded and grown in a Tissue Culture Flask of 75 cm² (T-75 Flask) with 10 mL of Dulbecco's Modified Eagle Medium (DMEM), supplemented with glucose and 10 % fetal bovine serum (FBS) and collected for genomic DNA (gDNA) extraction. The extracted gDNA was used as the template for generation of ALU115 and ALU247 by asymmetric PCR. The reaction mixture consisted of 25 µL of NovaTaq PCR Master Mix plus 10 ng of gDNA template; 0.4mM MgCl₂; 0.02 µM forward primer and 0.5 µM reverse primer. The forward and reverse primers were as follows, respectively: For ALU115 – 5'-GTAGGTACCCTGAGGTCAGGAGTTC-3' and 5'-Biotin-CCCGAGTAGCTGGGATTACA-3'.and for ALU247 - 5'-GTGGCTCACGCCTGTAATC-3' and 5'-Biotin-CAGGCTGGAGTGCAGTGG-3'. The PCR amplification reactions was performed with a pre-cycling heat activation at 95 °C for 5 min, followed by 35 cycles of denaturation at 95 °C for 1 min, annealing at 54 °C for 50 s, and extension at 72 °C for 50 s. Obtained target fragments were analyzed by gel electrophoresis in 2 % agarose (Fig. S1).

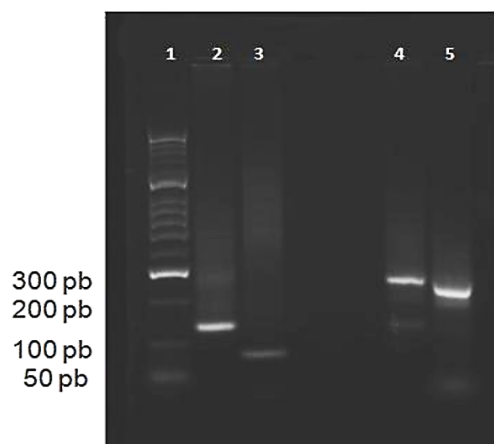


Fig S1. DNA product analysis for ALU115 and ALU247. Agarose gel with ALU115 and ALU247 generated by standard PCR and asymmetric PCR. HyperLadder IV (ranging from 10 kb to 200 bp) was used for fragment size assessment.

Biomolecule target conjugation with 250 nm and 100 nm MNPs

The generated DNA targets were purified from the PCR mixture contaminants using the DNA Clean & Concentrator™-25 from Zymo research and diluted in TE buffer (Tris 10 mM-EDTA 1mM, pH 7.4) for further use in labeling assays. Magnetic nanoparticles of 250 nm (Nanomag-D 250 nm), modified with a streptavidin outer layer, were used for the labeling of the generated targets through streptavidin/biotin interactions. At first, from the stock suspension (4.9×10^{11} nanoparticles/mL), 10 μ L of Nanomag-D 250 nm were collected in a clean sterile *ependorf* tube and placed on a magnetic concentrator (DynaMag™-2 supplied by Invitrogen) for 1-2 min to attract the magnetic nanoparticles at the magnet and separated from the supernatant. After, the supernatant was removed by aspiration with a pipette while the tube resided on the magnetic concentrator. The tube was then removed from the magnet and the magnetic nanoparticles were washed with the PB/Tween-20 buffer. This process was repeated for a total of 3 washes. Finally, the magnetic nanoparticles were diluted 10x to a total volume of 100 μ L of PB/Tween-20 buffer and to a final concentration of 4.9×10^7 nanoparticles/ μ L. Given the supplier's information, each nanoparticle is covered with about 500 streptavidin molecules. Thus, 2.45×10^{10} streptavidin molecules can be considered to be available per μ L of the prepared suspension. 10 μ L of this suspension was put into contact with 1 μ L of the solution containing the amplified DNA fragment ssALU115-biot at 1 μ M. Thereafter, the magnetic labeling reaction occurred for about 30 min at room temperature ($\sim 25^\circ\text{C}$). Finally, the magnetic labeling suspension was placed again on the magnetic concentrator to capture the magnetically labeled DNA targets that have bound to the nanoparticles by the streptavidin-biotin interaction. The supernatant was discharged and the magnetic labeled molecules suspended in 10 μ L of PB buffer.