# Single-Molecule DNA Visualization Using AT-specific Red and Non-specific Green DNA-Binding Fluorescent Protein Combination to Visualize DNA Sequences

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# **Supporting Information (SI)**

# PCR Primers Used in This Study

Name	Sequence
P1-HNS	5'-ACT TCA CAT ATG ATG AGC GAA GCA CTT AAA ATT CTG-3'
P2-HNS	5'-GCC ACC AGA ACC ACC TTG CTT GAT CAG GAA ATC GTC G-3'
P3-mCherry	5'-CAA GCA AGG TGG TTC TGG TGG CAT GGT GAG CAA GGG CGA GGA G-3'
P4-mCherry	5'-ATT TCA GGA TCC CTA CTT GTA CAG CTC GTC CAT GCC-3'
P5-BRCA	5'-TAT GCA CAT ATG GTA GAG AGT AAT ATT GAA GAC AAA ATA TTT GGG-3'
P6-BRCA	5'-GCT CAC CAT ACC GCC GCT GCC ACC TTT TGG CCC TCT GTT TCT ACC TAG-3'
P7-eGFP	5'-GGT GGC AGC GGC GGT ATG GTG AGC AAG GGC GAG GAG-3'
P8-eGFP	5'-TAT GCA GGA TCC TTA CGC CTT GTA CAG CTC GTC CAT G-3'
P9-HMG- mCherry	5'-ATA TTG CAT ATG AAA TGG AAA TGG AAA AAA GCG ATG GTG AGC AAG GGC GAG GAG-3'
P10-HMG- mCherry	5'-ATA TTG GGA TCC TTA TTT CCA TTT CCA TTT TTT CGC CTT GTA CAG CTC GTC CAT GCC-3'

# Full Sequences for Fluorescent Protein-DNA Binding Protein

#### H-NS-mCherry

MGSSHHHHHHSSGLVPRGSHMMSEALKILNNIRTLRAQARECTLETLEEMLEKLEVVV

NERREEESAAAAEVEERTRKLQQYREMLIADGIDPNELLNSLAAVKSGTKAKRAQRPAKYSYVDENGE TKTWTGQGRTPAVIKKAMDEQGKSLDDFLIKQ-GGSGG-mCherry.

#### BRCA1-eGFP

MGSSHHHHHHSSGLVPRGSHMVESNIEDKIFGKTYRKKASLPNLSHVTENLIIGAFVTEP QIIQERPLTNKLKRKRRPTSGLHPEDFIKKADLAVQKTPEMINQGTNQTEQNGQVMNITNSGHENKTKG DSIQNEKNPNPIESLEKESAFKTKAEPISSSISNMELELNIHNSKAPKKNRLRRKSSTRHIHALELVVSRNL SPPNCTELQIDSCSSSEEIKKKKYNQMPVRHSRNLQLMEGKEPATGAKKSNKPNEQTSKRHDSDTFPEL KLTNAPGSFTKCSNTSELKEFVNPSLPREEKEEKLETVKVSNNAEDPKDLMLSGERVLQTERSVESSSIS LVPGTDYGTQESISLLEVSTLGKAKTEPNKCVSQCAAFENPKGLIHGCSKDNRNDTEGFKYPLGHEVNH SRETSIEMEESELDAQYLQNTFKVSKRQSFAPFSNPGNAEEECATFSAHSGSLKKQSPKVTFECEQKEEN QGKNESNIKPVQTVNITAGFPVVGQKDKPVDNAKCSIKGGSRFCLSSQFRGNETGLITPNKHGLLQNPY RIPPLFPIKSFVKTKCKKNLLEENFEEHSMSPEREMGNENIPSTVSTISRNNIRENVFKEASSSNINEVGSS TNEVGSSINEIGSSDENIQAELGRNRGPK-GGSGG-eGFP

### 2HMG-mCherry<sup>1</sup>

MGSSHHHHHHSSGLVPRGSHM-TPKRPRGRPKK-GGSGG-mCherry-GGSGG-KKPRGRPRKPT

# $2(KW)_2$ -eGFP<sup>2</sup>

MGSSHHHHHHSSGLVPRGSHM-KWKWKKA-eGFP-AKKWKWK



Figure S1. H-NS-mCherry and BRCA1-eGFP stained T4 DNA (166 kb) with λ DNA (48.5 kb).



**Figure S2.** Length distribution of  $\lambda$  DNA molecules stained in H-NS-mCherry and BRCA1-eGFP. The average length is 13.4 µm (53 pixels).

- 1. S. Lee, C. Wang, J. Song, D. G. Kim, Y. Oh, W. Ko, J. Lee, J. Park, H. S. Lee and K. Jo, *Rsc Adv*, 2016, **6**, 46291-46298.
- 2. S. Lee, Y. Oh, J. Lee, S. Choe, S. Lim, H. S. Lee, K. Jo and D. C. Schwartz, *Nucleic Acids Res*, 2016, 44, e6.