Supporting Information

Synergistic targeting HER2 and EGFR with a bivalent aptamer-siRNA chimera efficiently inhibits HER2-positive tumor growth

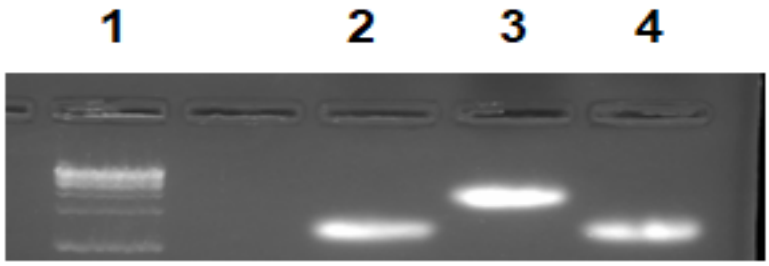
Lu Xue† § ⊥, Nita J. Maihle†⊥, Xiaolin Yu †, Shou-Ching Tang ‡, and Hong Yan Liu†*

†Georgia Cancer Center, Department of Biochemistry and Molecular Biology, Medical College of Georgia, Augusta University, Augusta, GA, 30912

‡ University of Mississippi Medical Center Cancer Institute, Jackson, MS 39216

§ Department of Pediatrics Hematology, the First Hospital of Jilin University, Changchun, 130021, China

*Email: holiu@augusta.edu



1. MW marker
2. HER2 apt-EGFR siRNA sense strand
3. Annealed HEH
4. HER2 apt-EGFR siRNA anti-sense strand

Figure S1. Detection of annealed HEH with 3 % agarose. To generate HEH, equal moles of HER2 aptamer-EGFR siRNA sense strand and HER2 aptamer-EGFR siRNA anti-sense strand were mixed together and heated to 95 °C for 3 min, followed by slowly cooling to room temperature. After annealing, in HEH lane, no free aptamer-EGFR siRNA sense strand or HER2 aptamer-EGFR siRNA anti-sense strand is detectable. Lane 1(L1): Molecular weight marker; L2: HER2 aptamer-EGFR siRNA sense strand; L3: annealed HEH; L4: HER2 aptamer-EGFRsiRNA anti-sense strand.

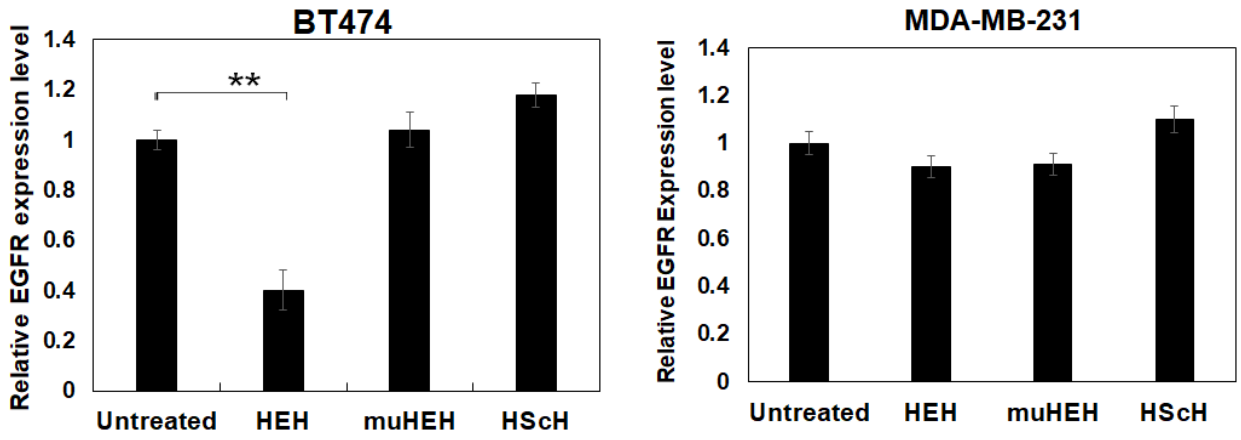


Figure S2. Detection of EGFR mRNA with qRT-PCR in BT474 and MDA-MB-231 cells. HER2 positive BT474 cells and HER2 negative MDA-MB-231 cells were treated with HEH, muHEH or HER2 aptamer-scrambled siRNA at 2μM for 48h. RNA was extracted and reverse transcribed as described in material and methods. Gene copy numbers were normalized against GAPDH. *P< 0.05. **P<0.001.

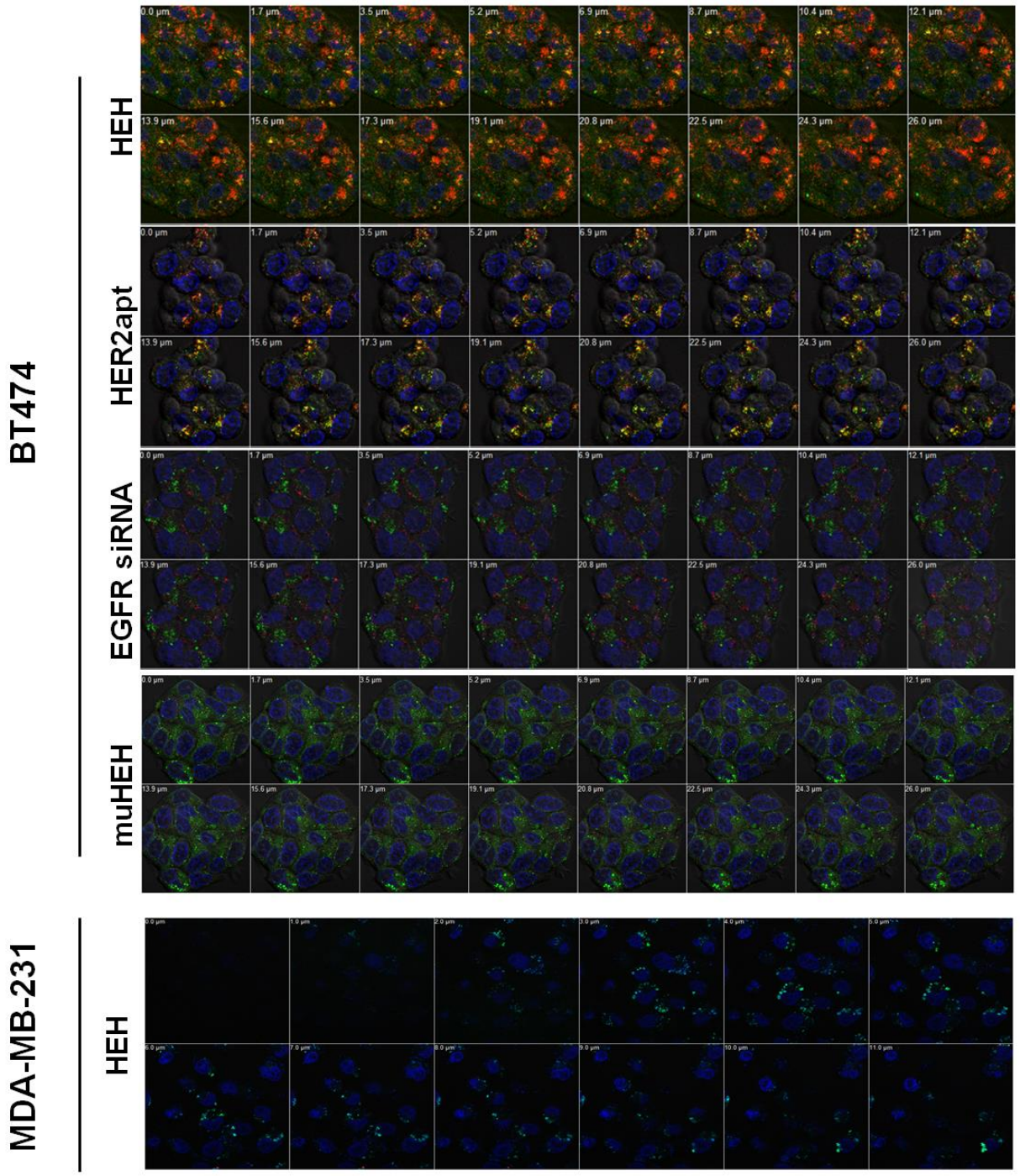

Red: Cy5 chimera; Green: lysotracker; blue: DAPI.
BT474
HEH
HER2apt
EGFR siRNA
muHEH
MDA-MB-231
HEH

Figure S3. Detection of HEH internalization by Z-Stack Confocal Microscopy. Cy5-labeled HEH, EGFRsiRNA, HER2 aptamer, or muH2EH3 was individually added into BT474 cells for 12 h at 37°C. LysoTracker Green was used to show lysosomes and endosomes. DAPI was used to display nucleus. Confocal laser scanning microscopy with z stack was performed to show cell binding and internalization. As a cell control, HER2 negative MDA-MB-231 cells were treated with Cy5-HEH for 12h at 37°C and the internalization was determined with Z-stack confocal microscopy.