Supporting Information

Using an RNA aptamer probe for super-resolution imaging of native EGFR

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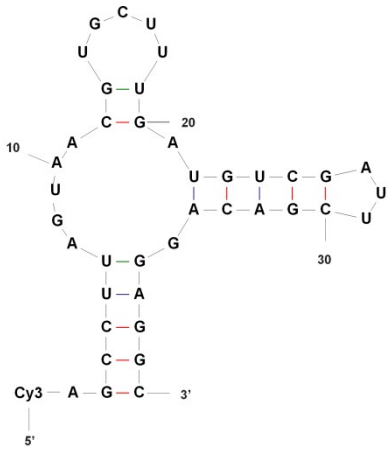

Fig. S1 The secondary structure of the EGFR aptamer predicted by the mfold web server. At the 5' end, an adenosine residue and a Cy3 were inserted for super-resolution imaging. The dissociation constant (Kd) is 10 nM[1].

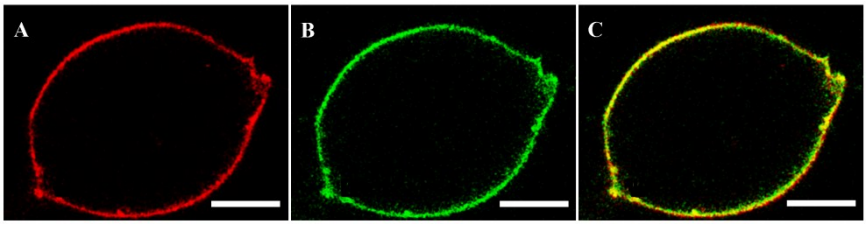

Fig. S2 Confocal images of live COS-7 cells. Cells were stained with membrane fluorescent probe DiO (A) and Cy3-conjugated EGFR aptamer (B). The merged image of DiO and aptamer labeling was shown in (C). Scale bars are 10 μm.

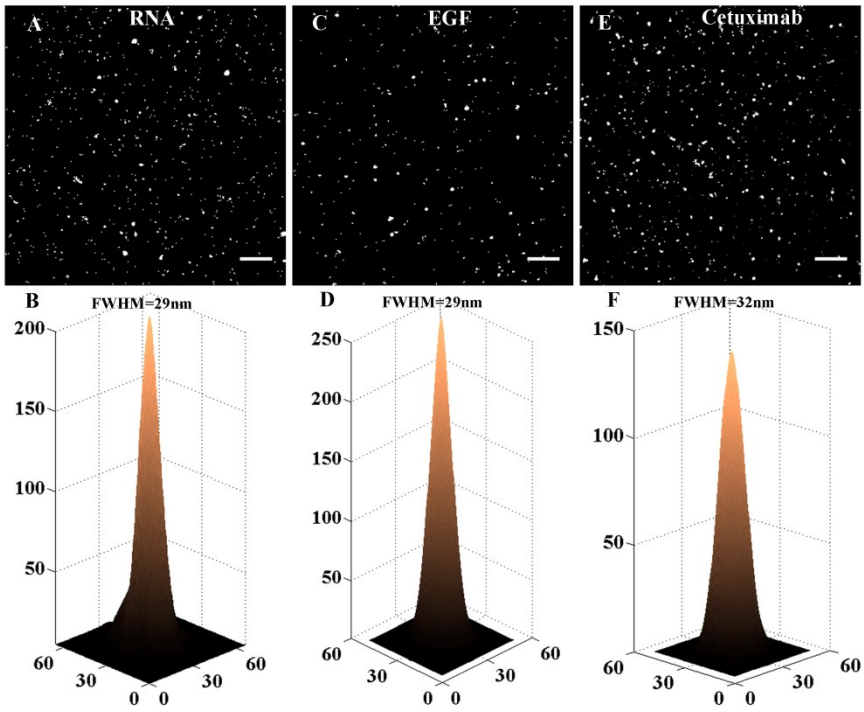

Fig. S3 The localization precision of single Cy3-conjugated RNA, EGF or Cetuximab on the membranes of fixed COS-7 cells. (A) A representative dSTORM image of Cy3-aptamer labeled EGFR on COS-7 cell membrane at a low labeling concentration of ~5 nM. (B) Three-dimensional Gaussian profile generated by aligning two-dimensional distribution of localizations from 60 fluorescent molecules with the FWHM of ~29 nm that represented the mean localization precision of aptamer probes. The average number of localizations in single Cy3-aptamer was 33. (C-F) The same measurement of the localization precision of Cy3-EGF and Cy3-Cetuximab. The values of FWHM were 29 nm and 32 nm, respectively. The average number of localizations were 33 and 35, respectively. Scale bars are 2 μm.

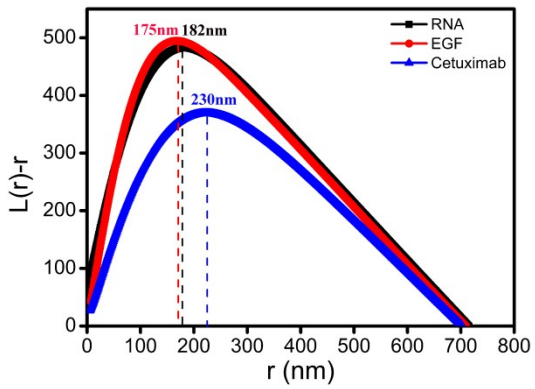

Fig. S4 The representative Ripley's K function plots under the different labeling conditions. The r_{max} values of Cy3-aptamer, Cy3-EGF and Cy3-Cetuximab are 182 nm, 175 nm and 230 nm, respectively.

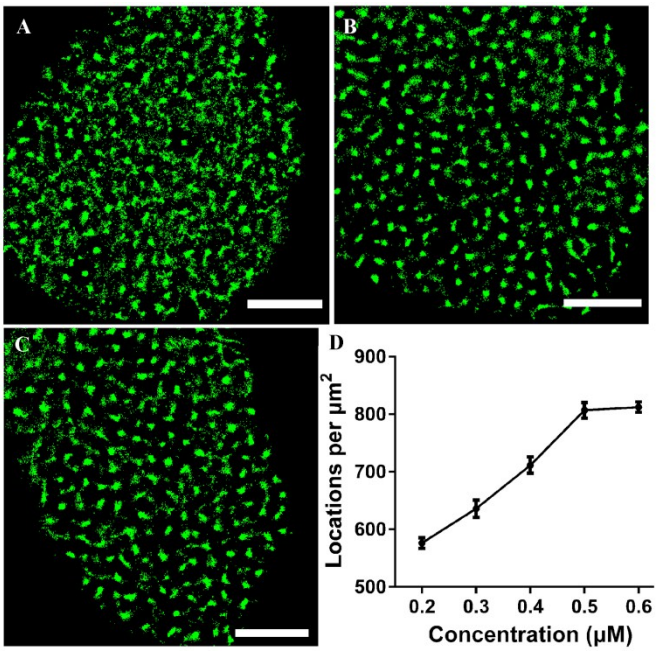

Fig. S5 Determination of the labeling concentration of EGFR aptamer. (A-C) The reconstructed dSTORM images of EGFR labeled with Cy3-conjugated aptamer under different concentrations of 0.4 μM (A), 0.5 μM (B) and 0.6 μM (C). Scale bars are 5 μm. (D) The plot of localization number per μm^2 at five labeling concentrations (0.2 μM, 0.3 μM, 0.4 μM, 0.5 μM, and 0.6 μM).

Notes and References

1. C. L. Esposito, D. Passaro, I. Longobardo, G. Condorelli, P. Marotta, A. Affuso, V. de Franciscis and L. Cerchia, *PLoS One*, 2011, **6**, e24071.