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Supplemental information

Origin of diverse phosphorylation patterns in the ERBB system

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Supporting Material

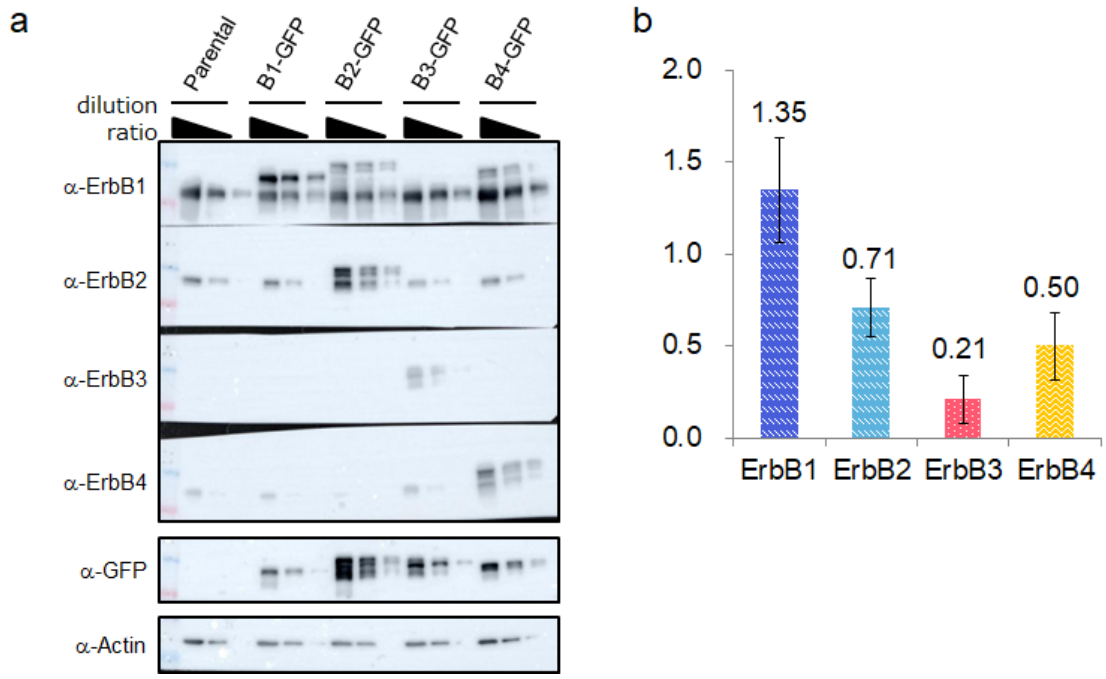

Fig. S1. Correction factors for the compensation of differences in the antibody titers. **a.** Western blots of ERBBs fused with GFP (ERBB-GFP). **b.** The correction factors were calculated by dividing the band intensities of anti-ERBBs by those of anti-GFP in order to reflect the amount of fused proteins. Error bars, SD.

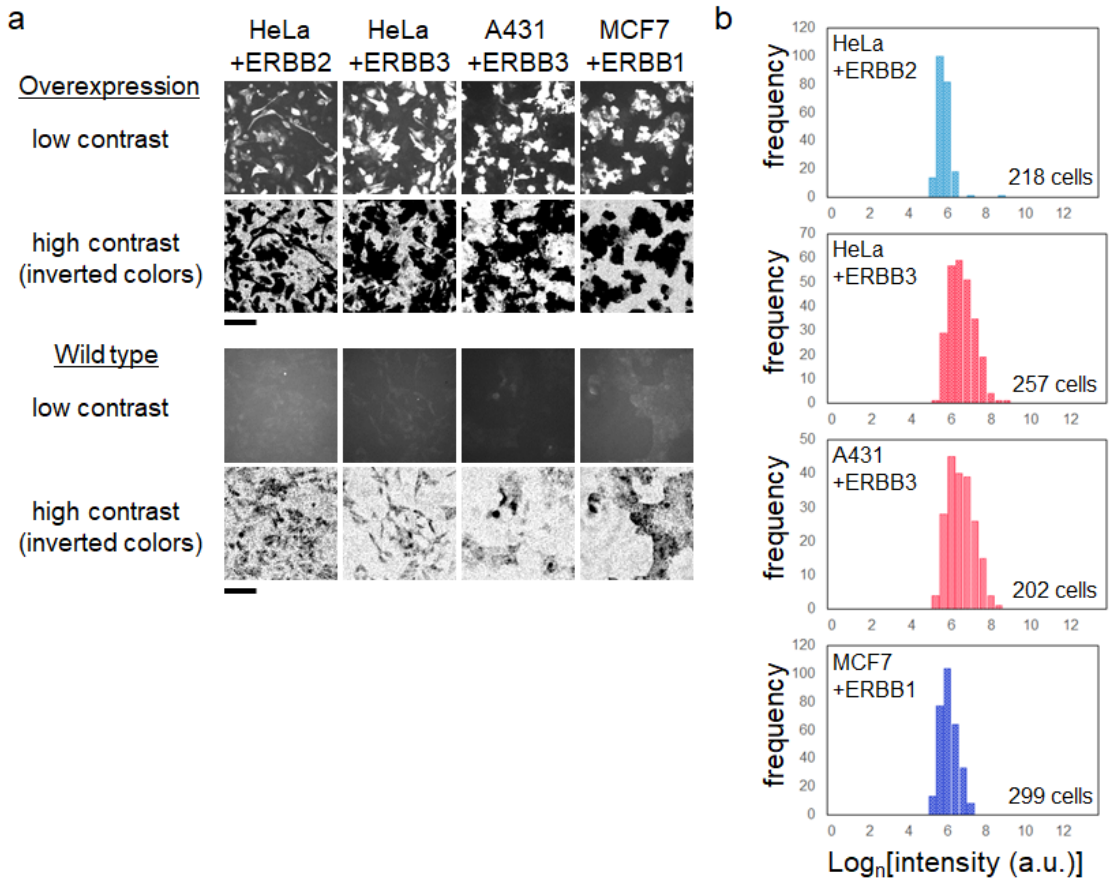

Fig. S2. Fluorescence images of ERBB-overexpressing and wild-type cells. **a.** Images shown in Fig. 4a (upper rows) are inverted in color, and the contrast was changed to recognize the cell locations (bottom rows). Scale bars, 50 µm. **b.** Distributions of the fluorescence intensities of ERBB-overexpressing cells indicate the populations can be considered single populations with regards to the protein expression.

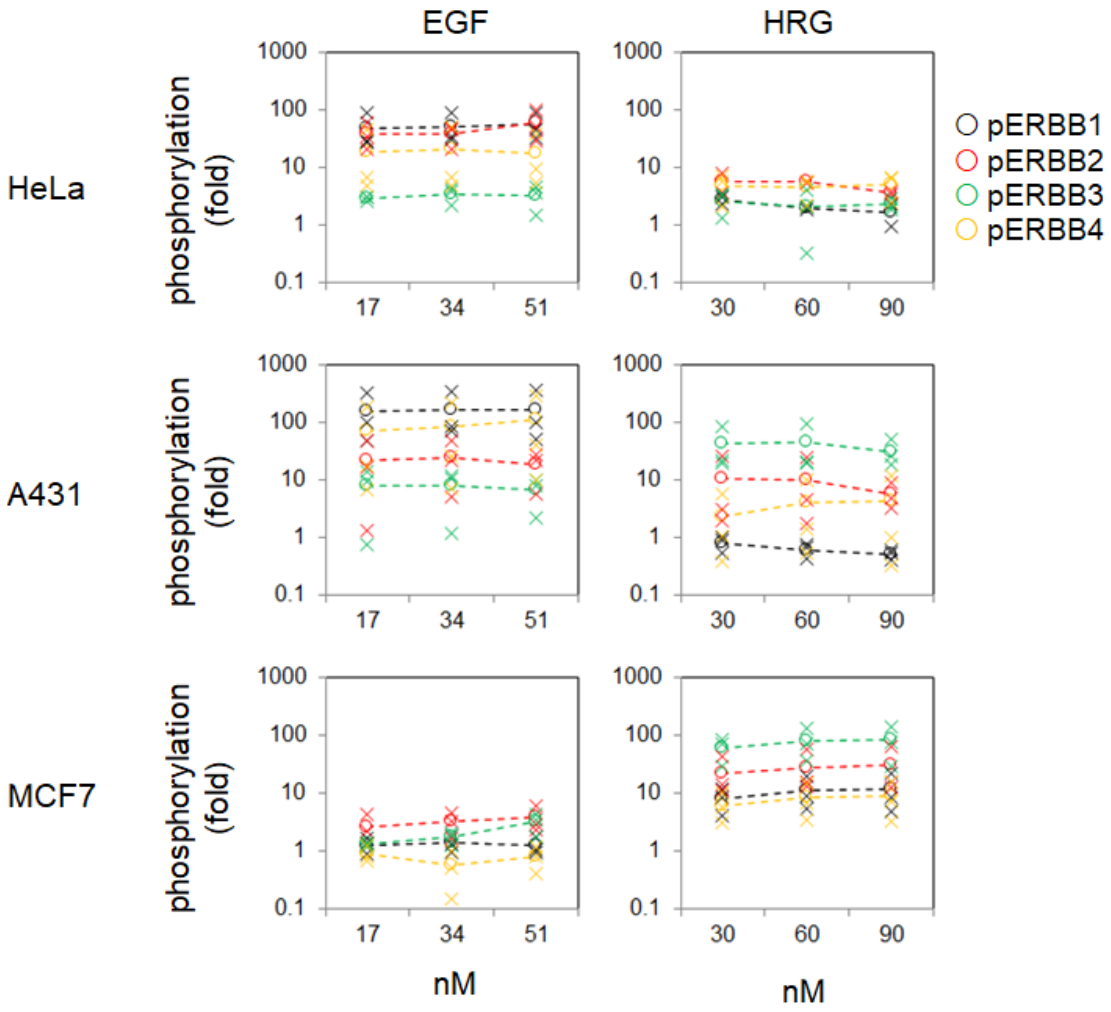

Fig. S3. Increasing the ligand concentration did not increase the phosphorylation levels of ERBBs, indicating saturation in the ERBB response. Crosses and circles indicate the data of individual trials and their averages, respectively, for each ERBB and ligand concentration.

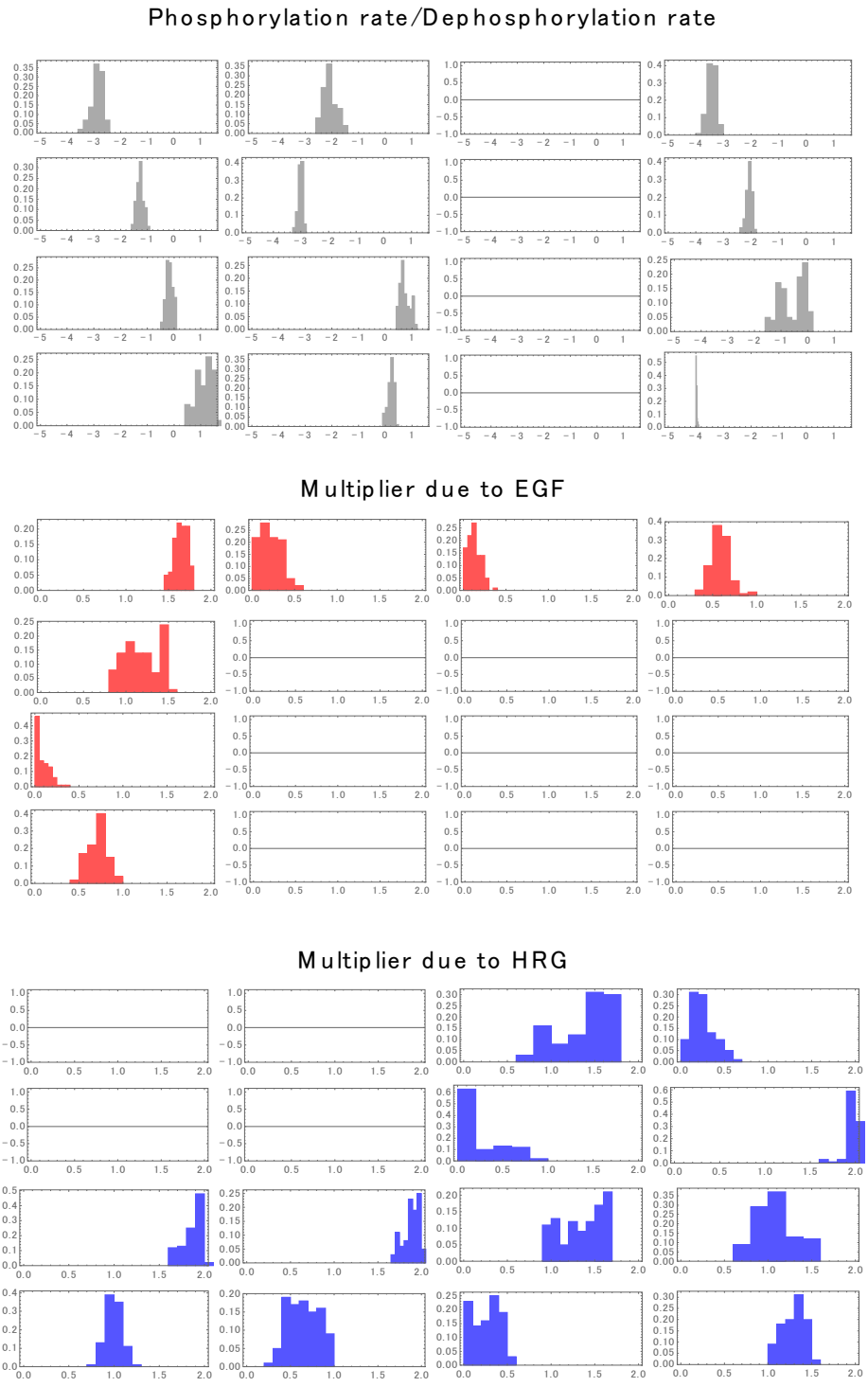

Fig. S4 a. The distributions of inferred values for the ratios of the phosphorylation rates to the dephosphorylation rates and the multipliers due to EGF or HRG stimulation shown in the phosphorylation networks in Fig. 6a. For each of the three panels, the four rows and columns are ERBB1, 2, 3, and 4, respectively. The horizontal axes show $\log_{10}$ values.

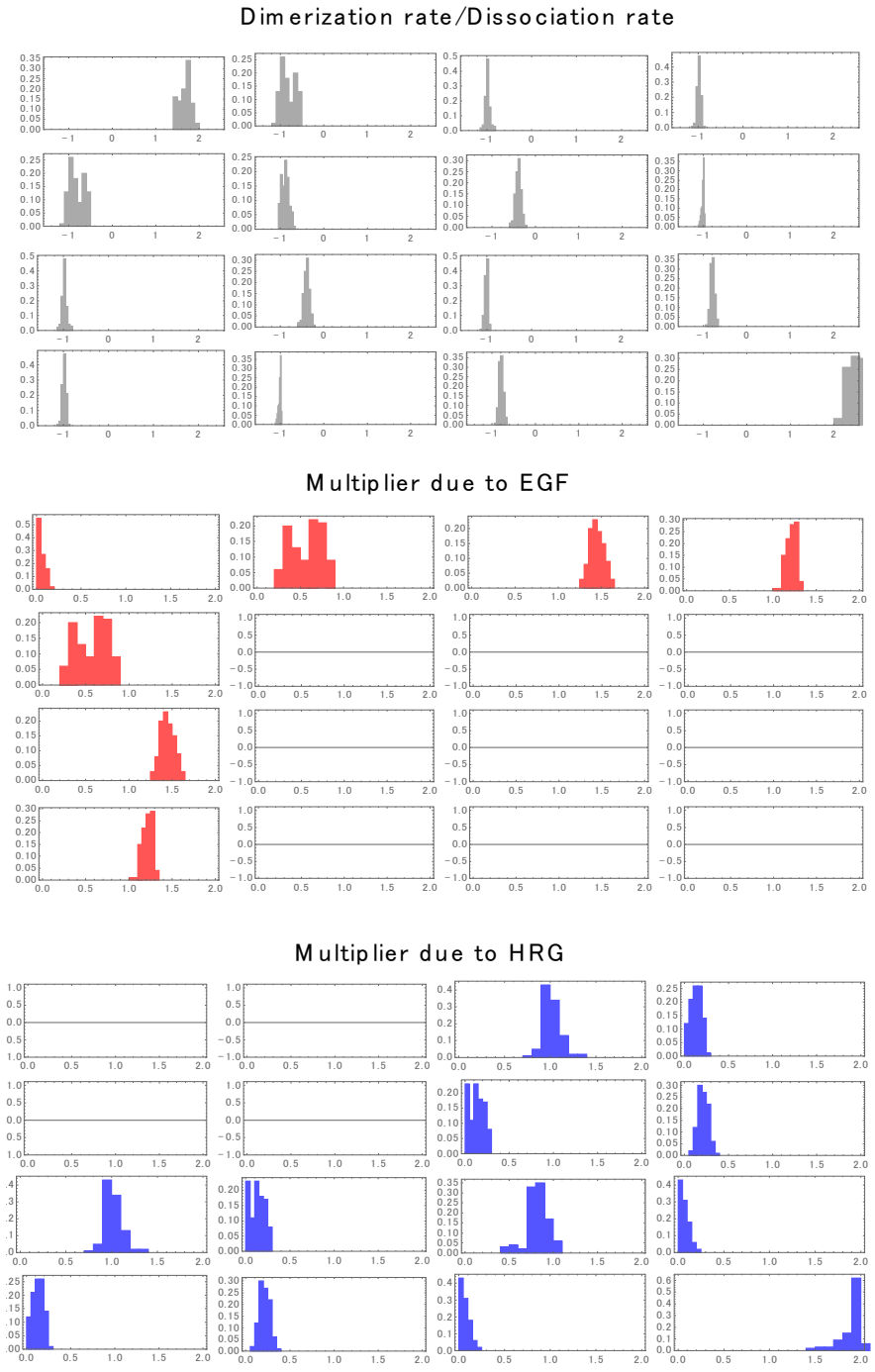

Fig. S4 b. The distributions of inferred values for the ratios of the dimerization rates to the dissociation rates and the multipliers due to EGF or HRG stimulation shown in the dimerization networks in Fig. 6b. For each of the three panels, the four rows and columns are ERBB1, 2, 3, and 4, respectively. The distribution in the i-th row and the j-th column is identical with that in the j-th row and the i-th column ($i, j = 1,2,3,4$). The horizontal axes show $\log_{10}$ values.

Monomer phosphorylation $k_{i,0!\ 1}^{phos}$	0.01	0.0001	0	0.0001
Monomer dephosphorylation $k_{i,1!\ 0}^{phos}$	0.002	0.002	0.0005	0.0006
Dimerization [1/nM sec] $k_{i,j}^{dim}$	0.09 for all pairs			
Dimer dissociation $r_{i,j}^{dim}$	0.002	0.6	0.9	0.9
	0.6	0.7	0.2	1.
	0.9	0.2	1.	0.6
	0.9	1.	0.6	0.0003
Dimer phosphorylation $k_{ij,0a\to 1a}^{phos}$	0.0005	0.005	0	0.0002
	0.0008	0.0009	0	0.008
	0.0001	0.009	0	0.0002
	0.005	0.0002	0	0.0001
Dimer dephosphorylation $k_{ij,1a\to 0a}^{phos}$	0.4	0.5	0.0002	0.4
	0.01	0.9	0.0002	0.9
	0.0002	0.002	0.09	0.0006
	0.0003	0.0001	0.0001	1.

Multiplier for dimer dissociation rates by EGF	0.9	0.1	0.9	0.5
	0.1	1.	1.	1.
	0.9	1.	1.	1.
	0.5	1.	1.	1.
Multiplier for dimer dissociation rates by HRG	1.	1.	0.2	0.9
	1.	1.	0.3	0.4
	0.2	0.3	0.02	0.9
	0.9	0.4	0.9	0.01
Multiplier for dimer phosphorylation rates by EGF	40.	2.	-	4.
	20.	1.	-	1.
	1.	1.	-	1.
	5.	1.	-	1.
Multiplier for dimer phosphorylation rates by HRG	1.	1.	-	2.
	1.	1.	-	90.
	70.	80.	-	10.
	10.	4.	-	20.

Table S1 Average parameter values estimated from the 7-cell fitting. In each row, the four columns represent the index of the ERBB subtypes ***i***=1,2,3,4. For a matrix of rate constants, the four rows represent the subtype index ***j***=1,2,3,4. For example, in "Dimer phosphorylation", the value 0.005 in the 4th row and 1st column denotes the phosphorylation rate of ERBB1 within the ERBB1/ERBB4 dimer.