Table S1. Reactions and parameters in simple models

No.	Mathematical representation	Reaction	Parameters	Unit	Reference	Description
v1	kf1*x1*x9	ErbB3 + ErbB2 -> ErbB3_ErbB2	1.81E-04	/molecules/min	estimated	HRG binding to ErbB3 monomer
v2	kr1*x2	ErbB3_ErbB2 -> ErbB3 + ErbB2	20	/min	estimated	
v3	kf2*hrg*x1	ErbB3(+[HRG]) -> HRG:ErbB3	0.125	/uM/min	estimated	HRG binding to ErbB3 monomer
v4	kr2*x3	HRG:ErbB3 -> ErbB3(+[HRG])	20	/min	estimated	Dissociation of HRG from ErbB3 monomer
v5	kf22*hrg*x2	ErbB3_ErbB2(+[HRG])-> HRG:ErbB3_ErbB2	6	/uM/min	estimated	HRG binding to ErbB3_ErbB2 dimer
v6	kr2*x4	HRG:ErbB3_ErbB2 -> ErbB3_ErbB2(+[HRG])	20	/min	estimated	Dissociation of HRG from ErbB3_ErbB2 dimer
v7	kf3*x3*x9	HRG:ErbB3 + ErbB2 -> HRG:ErbB3_ErbB2	8.68E-03	/molecules/min	estimated	Dimer formation of HRG-bound ErbB3 and ErbB2
v8	kr3*x4	HRG:ErbB3_ErbB2 -> HRG:ErbB3 + ErbB2	20	/min	estimated	Dissociation of ErbB2 from ErbB3_ErbB2 dimer
v9	V4*x4/(Km4+x4)	HRG:ErbB3_ErbB2 -> pErbB3_pErbB2	V4 = 2000 Km4 = 2000	/min molecules	estimated	Autophosphorylation of HRG-binding ErbB3_ErbB2 dimer
v10	k5*x5*x8/(Km5+x5)	pErbB3_pErbB2 -> HRG:ErbB3_ErbB2	k5 = 1.6 Km5 = 4000	/min molecules	estimated	Dephosphorylation of pErbB3_pErbB2 mediated by PTPN
v11	kf6*x5*x5	pErbB3_pErbB2 *2 -> (pErbB3_pErbB2)2	0.002	/molecules/min	estimated	Tetramer formation of pErbB3_pErbB3 dimers
v12	kr6*x6	(pErbB3_pErbB2)2 -> pErbB3_pErbB2 *2	4	/min	estimated	Dissociation of tetramer
v13	ks7	-> PHLDA1 synthesis	[initial x7]*kd7 †	molecules/min	estimated	Basal synthesis of PHLDA1
v14	ks7d*x5+ks7dd*x6	pErbB3_pErbB2 & (pErbB3_pErbB2)2 -> PHLDA1 syr	ks7d = 0.04 ks7dd = 0.04	/min /min	estimated	Synthesis of PHLDA1 promoted by pAkt and c-Fos
v15	kd7*x7	PHLDA1 -> degradation	0.02	/min	estimated	Degradation of PHLDA1
v16	ks8*x5+ks8d*x6	pErbB3_pErbB2 & (pErbB3_pErbB2)2 -> PTPN synth	ks8 = 0.04 ks8d = 0.4	/min /min	estimated	Synthesis of PTPN promoted by pErbB3_pErbB2 dimer and tetramer
v17	kd8*x8	PTPN -> degradation	0.02	/min	estimated	Degradation of PTPN
Feedback	1 - x7/(ki+x7)	Negative feedback from PHLDA1	6000	molecules	estimated	

† Synthetic constants of DUSP and PHLDA1 were calcurated from randomly selected initial values in the simulation with cell-to-cell variability.

Table S2. Initial concentration of molecules in simple models

Symbol	Species	Molecules/cell	Reference
HRG	Concentration of HRG	10 (uM)	Experimental condition in this study
y1	HRGR	10000	estimated based on [1]
y2	HRGR_ErbB2	0	
y3	HRG:HRGR	0	
y4	HRG:HRGR_ErbB2	0	
y5	pHRGR_pErbB2	0	
y6	(pHRGR_pErbB2)2	0	
y7	PHLDA1	6000	estimated
y8	Phosphatase	0	
y9	ErbB2	8000	estimated based on [1]

[1] Zhang, Q., Park, E., Kani, K., and Landgraf, R. (2012). Proc. Natl. Acad. Sci. USA 109, 13237–13242

Table S3. CV of molecules in simple models

Species	CV of protein concentrations (%)	Reference
HRGR	31	estimated by measuring CV of ErbB3 (Fig. 3D)
ErbB2	40	measured in this study (Fig. 3D)
PHLDA1	100	

Table S4. Rank correlations in simple models

Species	Rank correlation	Reference
HRGR-PHLDA1	0.13	estimated by measuring rank correlation between ErbB3 & PHLDA1 (Fig. 3F)
ErbB2-PHLDA1	0.25	measured in this study (Fig. 3F)

Table S5. Negative feedback from PHLDA1 in simple models

Name	Reactions inhibited by negative feedback from PHLDA1	Description
Model M0	-	No feedback from PHLDA1
Model M1	v1 and v7	Inhibition of dimer formation
Model M2	v9	Inhibition of phosphorylation
Model M3	v11	Inhibition of tetramer formation
Model M4	v1, v7 and v11	Inhibition of dimer and tetramer formation

Table S6. Reactions and parameters in the expanded model

No.	Reaction	Mathematical representation	Parameters	Value	Unit	Reference	Description
Complex formation of ErbB receptors							
			kb0_1	2.34.E-04	/molecules/s	fitted	
v1	HRGR + ErbB2 -> HRGR_ErbB2	kb0_1*y1*y59 + kb0_2*y1*y59*ki/(ki+y57)	kb0_2	2.71.E-05	/molecules/s	fitted	Dimer formation of HRGR and ErbB2 inhibited by PHLDA1 (this study)
			ki	4.19.E+07	molecules	fitted	
v2	HRGR_ErbB2 -> HRGR + ErbB2	kd0*y2	kd0	8.16.E-01	/s	fitted	Dissociation of ErbB2 from HRGR_ErbB2 dimer
v3	HRGR(+[HRG]) -> HRG:HRGR	kb1*y1*HRG	kb1	3.37.E+01	/uM/s	fitted	HRG binding to HRGR monomer
v4	HRG:HRGR -> HRGR(+[HRG])	kd1*y3	kd1	1.10.E+04	/s	fitted	Dissociation of HRG from HRGR monomer
v5	HRGR_ErbB2(+[HRG])-> HRG:HRGR_ErbB2	kb1*y2*HRG	kb1	3.37.E+01	/uM/s	fitted	HRG binding to HRGR_ErbB2 dimer
v6	HRG:HRGR_ErbB2 -> HRGR_ErbB2(+[HRG])	kd1*y4	kd1	1.10.E+04	/s	fitted	Dissociation of HRG from HRGR_ErbB2 dimer
			kdim1	4.18.E-02	/molecules/s	fitted	
v7	HRG:HRGR + ErbB2 -> HRG:HRGR_ErbB2	kdim1*y3 + kdim2*y3*ki/(ki+y57)	kdim2	4.85.E-03	/molecules/s	fitted	Dimer formation of HRG-bound HRGR and ErbB2 inhibited by PHLDA1 (this study)
			ki	4.19.E+07	molecules	fitted	
v8	HRG:HRGR_ErbB2 -> HRG:HRGR + ErbB2	kmono*y4	kmono	8.60.E+00	/s	fitted	Dissociation of ErbB2 from HRG-binding HRGR_ErbB2 dimer
v9	HRG:HRGR_ErbB2 -> pHRGR_pErbB2	k1*y4*y4	k1	1.00.E-02	/molecules/s	fitted	Autophosphorylation of HRG-binding HRGR_ErbB2 dimer
			k2	7.35.E+02	/s	fitted	
v10	pHRGR_pErbB2 -> HRG:HRGR_ErbB2	k2*y58*y5/(km2+y5)	km2	2.26.E+05	molecules	fitted	Dephosphorylation of pHRGR_pErbB2 mediated by PTPN
			ktet1	2.76.E+02	/molecules/s	fitted	
v11	pHRGR_pErbB2 *2 -> (pHRGR_pErbB2)2	ktet1*y5*y5 + ktet2*y5*y5*ki/(ki+y57)	ktet2	3.20.E+01	/molecules/s	fitted	Tetramer formation of pHRGR_pHRGR dimers inhibited by PHLDA1 (this study)
			ki	4.19.E+07	molecules	fitted	
v12	(pHRGR_pErbB2)2 -> pHRGR_pErbB2 *2	kdim*y3d	kdim	8.98.E+01	/s	fitted	Dissociation of tetramer
PI3K-Akt pathway							
v13	pHRGR_pErbB2+PI3K ->	kf3*y5*y7	kf3	1.08E-01	/molecules/s	fitted	Phosphorylation of PI3K by pHRGR_pErbB2 dimer
v14	pHRGR_pErbB2_PI3K -> pHRGR_pErbB2+PI3K	kr3*y8	kr3	1.62E+02	/s	fitted	
v15	pHRGR_pErbB2_PI3K -> pHRGR_pErbB2+PI3KA	kc3*y8	kc3	4.74E+00	/s	fitted	
v16	(pHRGR_pErbB2)2+PI3K ->	kf3*y6*y7	kf3	1.08E-01	/molecules/s	fitted	Phosphorylation of PI3K by pHRGR_pErbB2 tetramer
v17	(pHRGR_pErbB2)2_PI3K -> (pHRGR_pErbB2)2+PI3K	kr3*y9	kr3	1.62E+02	/s	fitted	
v18	(pHRGR_pErbB2)2_PI3K -> (pHRGR_pErbB2)2+PI3KA	kc3d*y9	kc3d	4.40E+00	/s	fitted	
v19	PIP2 + PI3KA ->	kf4*y10*y11	kf4	1.28E-03	/molecules/s	fitted	Phosphorylation of PIP2 by PI3K
v20	PI3KA_PIP2 -> PIP2 + PI3KA	kr4*y12	kr4	1.38E+02	/s	fitted	
v21	PI3KA_PIP2 -> PIP3 + PI3KA	kc4*y12	kc4	1.90E+01	/s	fitted	
v22	PIP3 + PDK1 ->	kf5*y13*y14	kf5	1.68E-04	/molecules/s	fitted	Phosphorylation of PDK1
v23	PIP3_PDK1 -> PIP3 + PDK1	kr5*y15	kr5	3.08E+02	/s	fitted	
v24	PIP3_PDK1 -> PIP3 + p-PDK1	kc5*y15	kc5	3.45E+02	/s	fitted	
v25	p-PDK1 + Akt ->	kf6*y16*y17	kf6	5.02E-03	/molecules/s	fitted	Phosphorylation of Akt by PDK1
v26	p-PDK1_Akt -> p-PDK1 + Akt	kr6*y18	kr6	3.81E+02	/s	fitted	
v27	p-PDK1_Akt -> p-PDK1 + p-Akt	kc6*y18	kc6	1.17E+03	/s	fitted	
v28	PI3KA + Pase1 ->	kf7*y10*y20	kf7	7.16E-04	/molecules/s	fitted	Dephosphorylation of PI3K
v29	Pase1_PI3KA -> PI3KA + Pase1	kr7*y21	kr7	3.64E+00	/s	fitted	
v30	Pase1_PI3KA -> PI3K + Pase1	kc7*y21	kc7	4.34E+00	/s	fitted	
v31	PIP3 + PTEN ->	kf8*y13*y22	kf8	6.56E-03	/molecules/s	fitted	Dephosphorylation of PIP3 by PTEN
v32	PTEN_PIP3 -> PIP3 + PTEN	kr8*y23	kr8	1.21E+02	/s	fitted	
v33	PTEN_PIP3 -> PIP2 + PTEN	kc8*y23	kc8	1.11E+02	/s	fitted	
v34	p-PDK1 + Pase2 ->	kf9*y16*y24	kf9	4.30E-03	/molecules/s	fitted	Dephosphorylation of PDK
v35	Pase2_p-PDK1 -> p-PDK1 + Pase2	kr9*y25	kr9	1.53E+01	/s	fitted	
v36	Pase2_p-PDK1 -> PDK1 + Pase2	kc9*y25	kc9	1.68E+02	/s	fitted	
v37	p-Akt + PP2A ->	kf10*y19*y26	kf10	1.12E-03	/molecules/s	fitted	Dephosphorylation of pAkt by PP2A
v38	PP2A_p-Akt -> p-Akt + PP2A	kr10*y27	kr10	1.16E+02	/s	fitted	
v39	PP2A_p-Akt -> Akt + PP2A	kc10*y27	kc10	5.22E+00	/s	fitted	
Ras-ERK pathway							
v40	pErbB2_pHRGR+RasGDP ->	kf11*y5*y28	kf11	2.41E-01	/molecules/s	fitted	Activation of RasGDP by pHRGR_pErbB2 dimer
v41	pHRGR_pErbB2_RasGDP -> p-ErbB + RasGDP	kr11*y29	kr11	2.32E+03	/s	fitted	
v42	pHRGR_pErbB2_RasGDP -> p-ErbB + RasGTP	kc11*y29	kc11	5.54E+02	/s	fitted	
v43	(pErbB2_pHRGR)2+RasGDP ->	kf11*y6*y28	kf11	2.41E-01	/molecules/s	fitted	Activation of RasGDP by pHRGR_pErbB2 tetramer
v44	(pHRGR_pErbB2)2_RasGDP -> (pErbB2_pHRGR)2+RasGDP	kr11*y30	kr11	2.32E+03	/s	fitted	
v45	(pHRGR_pErbB2)2_RasGDP -> (pErbB2_pHRGR)2+RasGTP	kc11d*y30	kc11d	1.39E+04	/s	fitted	
v46	RasGTP -> RasGDP	k12*y31	k12	3.76E+03	/s	fitted	Inactivation of RasGTP
v47	RasGTP + Raf ->	kf13*y31*y32	kf13	3.32E-02	/molecules/s	fitted	Activation of Raf by RasGTP
v48	RasGTP_Raf -> RasGTP + Raf	kr13*y33	kr13	8.04E+01	/s	fitted	
v49	RasGTP_Raf -> RasGDP + p-Raf	kc13*y33	kc13	6.17E+02	/s	fitted	
v50	p-Raf + MEK ->	kf14*y34*y35	kf14	5.36E-03	/molecules/s	fitted	Phosphorylation of MEK by Raf
v51	p-Raf_MEK -> p-Raf + MEK	kr14*y36	kr14	1.46E+03	/s	fitted	
v52	p-Raf_MEK -> p-Raf + p-MEK	kc14*y36	kc14	5.79E+04	/s	fitted	
v53	p-Raf + p-MEK ->	kf15*y34*y37	kf15	1.32E-01	/molecules/s	fitted	Phosphorylation of p-MEK by Raf
v54	p-Raf_p-MEK -> p-Raf + p-MEK	kr15*y38	kr15	4.45E+03	/s	fitted	
v55	p-Raf_p-MEK -> p-Raf + pp-MEK	kc15*y38	kc15	7.00E+03	/s	fitted	
v56	pp-MEK + ERK ->	kf16*y39*y40	kf16	1.81E-03	/molecules/s	fitted	Phosphorylation of ERK by MEK
v57	pp-MEK_ERK -> pp-MEK + ERK	kr16*y41	kr16	8.46E+02	/s	fitted	
v58	pp-MEK_ERK -> pp-MEK + p-ERK	kc16*y41	kc16	1.80E+02	/s	fitted	
v59	pp-MEK + p-ERK ->	kf17*y39*y42	kf17	1.78E-02	/molecules/s	fitted	Phosphorylation of p-ERK by MEK
v60	pp-MEK_p-ERK -> pp-MEK + p-ERK	kr17*y43	kr17	1.50E+01	/s	fitted	
v61	pp-MEK_p-ERK -> pp-MEK + pp-ERK	kc17*y43	kc17	1.01E+02	/s	fitted	
v62	RSK + pp-ERK ->	kf18*y44*y45	kf18	5.20E-05	/molecules/s	fitted	Phosphorylation of RSK by ERK
v63	RSK_pp-ERK -> RSK + pp-ERK	kr18*y46	kr18	3.25E+02	/s	fitted	
v64	RSK_pp-ERK -> p-RSK + pp-ERK	kc18*y46	kc18	4.08E+00	/s	fitted	
v65	p-RSK -> RSK	k19*y47	k19	1.12E+01	/s	fitted	Dephosphorylation of RSK
v66	p-Raf + Pase3 ->	kf20*y34*y48	kf20	2.15E-01	/molecules/s	fitted	Deactivation of p-Raf
v67	Pase3_p-Raf -> p-Raf + Pase3	kr20*y49	kr20	1.47E+02	/s	fitted	
v68	Pase3_p-Raf -> Raf + Pase3	kc20*y49	kc20	9.98E+01	/s	fitted	
v69	pp-MEK + Pase4 ->	kf21*y39*y50	kf21	3.70E-06	/molecules/s	fitted	Dephosphorylation of pp-MEK
v70	Pase4_pp-MEK -> pp-MEK + Pase4	kr21*y51	kr21	2.42E+03	/s	fitted	
v71	Pase4_pp-MEK -> p-MEK + Pase4	kc21*y51	kc21	2.42E+02	/s	fitted	
v72	p-MEK + Pase4 ->	kf22*y37*y50	kf22	6.89E-04	/molecules/s	fitted	Dephosphorylation of p-MEK
v73	Pase4_p-MEK -> p-MEK + Pase4	kr22*y52	kr22	2.96E+02	/s	fitted	
v74	Pase4_p-MEK -> MEK + Pase4	kc22*y52	kc22	5.98E+00	/s	fitted	
v75	pp-ERK + DUSP ->	kf23*y44*y53	kf23	2.43E-04	/molecules/s	fitted	Dephosphorylation of pp-ERK by DUSP
v76	DUSP_pp-ERK -> pp-ERK + DUSP	kr23*y54	kr23	3.94E+01	/s	fitted	
v77	DUSP_pp-ERK -> p-ERK + DUSP	kc23*y54	kc23	3.10E+02	/s	fitted	
v78	p-ERK + DUSP ->	kf24*y42*y53	kf24	5.51E-05	/molecules/s	fitted	Dephosphorylation of p-ERK by DUSP
v79	DUSP_p-ERK -> p-ERK + DUSP	kr24*y55	kr24	2.18E+00	/s	fitted	
v80	DUSP_p-ERK -> ERK + DUSP	kc24*y55	kc24	2.07E+01	/s	fitted	
Transcription regulatory network for PHLDA1 expression							
v81	pp-ERK & p-RSK -> c-Fos synthesis	Vcfos*(y44*y47)/(Kcfos+(y44*y47))	Vcfos	2.42E+04	molecules/s	fitted	Synthesis of c-Fos promoted by both pp-ERK- and p-RSK [1]
			Kcfos	2.59E+12	molecules^2	fitted	
v82	c-Fos -> degradation	d_cfos*y56	d_cfos	1.91E-04	/s	fitted	Degradation of c-Fos
v83	-> PHLDA1 synthesis	Vphlda1	Vphlda1	initial_y57*d_phl †	molecules/s	-	Basal synthesis of PHLDA1
v84	c-Fos & p-Akt -> PHLDA1 synthesis	s_phlda1*(y19*y56)	s_phlda1	1.14E-08	/molecules/s	fitted	Synthesis of PHLDA1 promoted by both pAkt and c-Fos
v85	PHLDA1 -> degradation	d_phlda1*y57	d_phlda1	1.00E-05	/s	fitted	Degradation of PHLDA1
			Vptpn	3.05E+02	molecules/s	fitted	
v86	pHRGR_pErbB2 & (pHRGR_pErbB2)2 -> PTPN synthesis	Vptpn*(y8+y9)/(Kptpn+(y8+y9))	Kptpn	8.11E+04	molecules	fitted	Synthesis of PTPN promoted by pHRGR_pErbB2 dimer and tetramer
v87	PTPN -> degradation	d_ptpn*y58	d_ptpn	1.76E-05	/s	fitted	Degradation of PTPN
v88	-> DUSP synthesis	Vdusp	Vdusp	initial_y53*d_dusp †	molecules/s	-	Basal synthesis of DUSP
v89	pp-ERK -> DUSP synthesis	s_dusp*y44	s_dusp	2.04E-02	/s	fitted	Synthesis of DUSP promoted by pp-ERK
v90	DUSP -> degradation	d_dusp*y53	d_dusp	7.55E-03	/s	fitted	Degradation of DUSP

† Synthetic constants of DUSP and PHLDA1 were calcurated from randomly selected initial values in the simulation with cell-to-cell variability.

[1] Nakakuki et al., Cell, 141, 884-896 (2010)

Table S7. Initial concentration of molecules in the expanded model

Symbol	Species	Molecules/cell	Reference
HRG	Concentration of HRG	10 (uM)	Experimental condition in this study
y1	HRGR	10000	We regard ErbB3 as HRG receptor (HRGR) in this study. Number of ErbB3 receptors is below 10,000 molecules per cell [1].
y2	HRGR_ErbB2	0	
y3	HRG:HRGR	0	
y4	HRG:HRGR_ErbB2	0	
y5	pHRGR_pErbB2	0	
y6	(pHRGR_pErbB2)2	0	
y7	PI3K	669000	fitted
y8	pE2pE3_PI3K	0	
y9	(pE2pE3)2_PI3K	0	
y10	active_PI3K	0	
y11	PIP2	7840000	fitted
y12	active_PI3K_PIP2	0	
y13	PIP3	0	
y14	PDK1	1520000	fitted
y15	PIP3_PDK1	0	
y16	p-PDK1	0	
y17	Akt	338000	fitted
y18	p-PDK1_Akt	0	
y19	pAkt	0	
y20	Pase1	213000	fitted
y21	Pase1_PI3KA	0	
y22	PTEN	68300	fitted
y23	PTEN_PIP3	0	
y24	Pase2	674000	fitted
y25	Pase2_p-PDK1	0	
y26	PP2A	2370	fitted
y27	PP2A_p-Akt	0	
y28	RasGDP	250000	400 nM per cell [2]
y29	pE2pE3_RasGDP	0	
y30	(pE2pE3)2_RasGDP	0	
y31	RasGTP	0	
y32	Raf	8000	13 nM per cell [2]
y33	RasGTP_Raf	0	
y34	p-Raf	0	
y35	MEK	850000	1400 nM per cell [2]
y36	p-Raf_MEK	0	
y37	p-MEK	0	
y38	p-Raf_MEKP	0	
y39	pp-MEK	0	
y40	ERK	600000	960 nM per cell [2]
y41	pp-MEK_ERK	0	
y42	p-ERK	0	
y43	pp-MEK_p-ERK	0	
y44	pp-ERK	0	
y45	RSK	498000	fitted
y46	RSK_pp-ERK	0	
y47	p-RSK	0	
y48	Pase3	5930	fitted
y49	Pase3_p-Raf	0	
y50	Pase4	1920000	fitted
y51	Pase4_pp-MEK	0	
y52	Pase4_p-MEK	0	
y53	DUSP	10000	fitted
y54	DUSP_pp-ERK	0	
y55	DUSP_p-ERK	0	
y56	c-Fos	0	
y57	PHLDA1	60000	fitted
y58	Phosphatase	0	
y59	ErbB2	8000	Number of ErbB2 receptors is below 10,000 molecules per cell [1].

[1] Zhang, Q. et al., Proc. Natl. Acad. Sci. USA 109, 13237–13242 (2012).

[2] Fujioka, A. et al., J. Biol. Chem., 281, 8917–8926 (2006).

Table S8. CV of molecules in the expanded model

Species	CV of protein concentrations (%)	Reference
HRGR (ErbB3)	31	Measured in this study (Fig. 3D)
ErbB2	42	
Akt	39	
ERK	35	
PHLDA1	60	
Others	35	Typical order [1, 2]

[1] Meyer, R. et al., Frontiers in physiology, 3, 451-451 (2011).

[2] Spencer, S. et al., Nature 459.7245, 428-432 (2009).

Table S9. Rank correlations in the expanded model

Combination	Rank correlation	Reference
HRGR(ErbB3)-PHLDA1	0.13	Measured in this study (Fig. 3F)
ErbB2_PHLDA1	0.27	
ERK-PHLDA1	0.15	
Akt-PHLDA1	0.14	