

Figure S1. Validation of Cell Motility Array. Immunoblotting for STAT3 and SH3PXD2A. GAPDH was used as a loading control. Ratios indicated below each band represent the relative band intensity of the protein of interest/GAPDH and were quantified with ImageJ v.1.440. STAT3, signal transducer and activator of transcription 3; SH3PXD2A, SH3 and PX domains 2A.

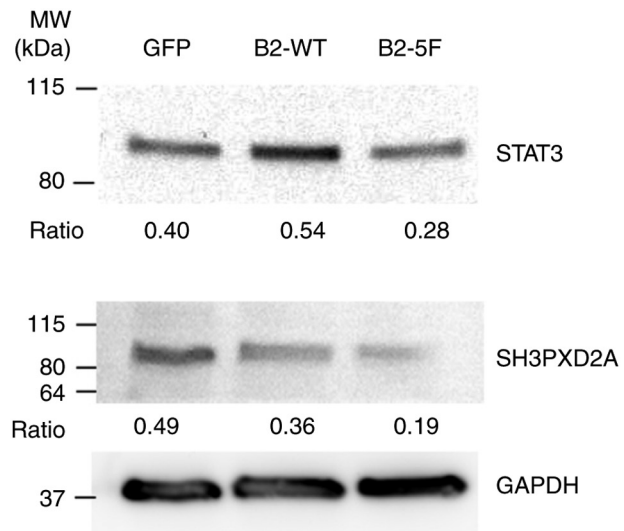


Figure S2. *EFNB* and *EPHB4* mRNA expression in the Van de Vijver public dataset. Distant recurrence-free survival differences related to the expression of (A) *EFNB2*, (B) *EPHB4* or (C) the combination variable *EFNB2/EPHB4*. Hazard ratios (HR) for univariate and multivariate analysis are inserted in the plots.

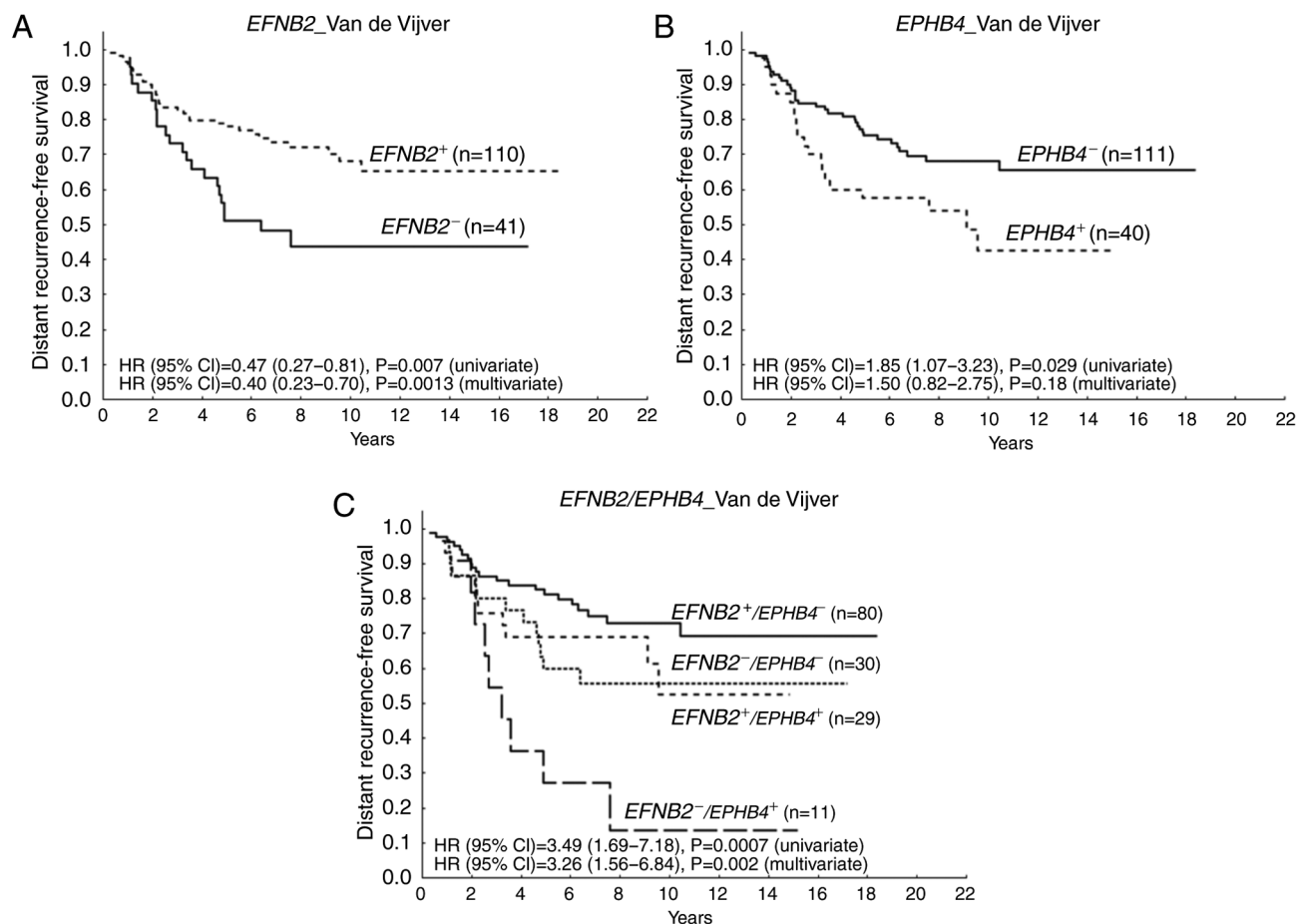


Figure S3. Kaplan-Meier survival plots. Gene expression levels of (A and B) *EFNB1*, (C and D) *EFNB2* and (E and F) *EFNB3* were analyzed in relation to the relapse-free survival in the Ki (A, C and E) or the Van de Vijver datasets (B, D and F). A high *EFNB2* expression, but not *EFNB1* nor *EFNB3* expression indicated a better prognosis compared with a lower expression in both datasets.

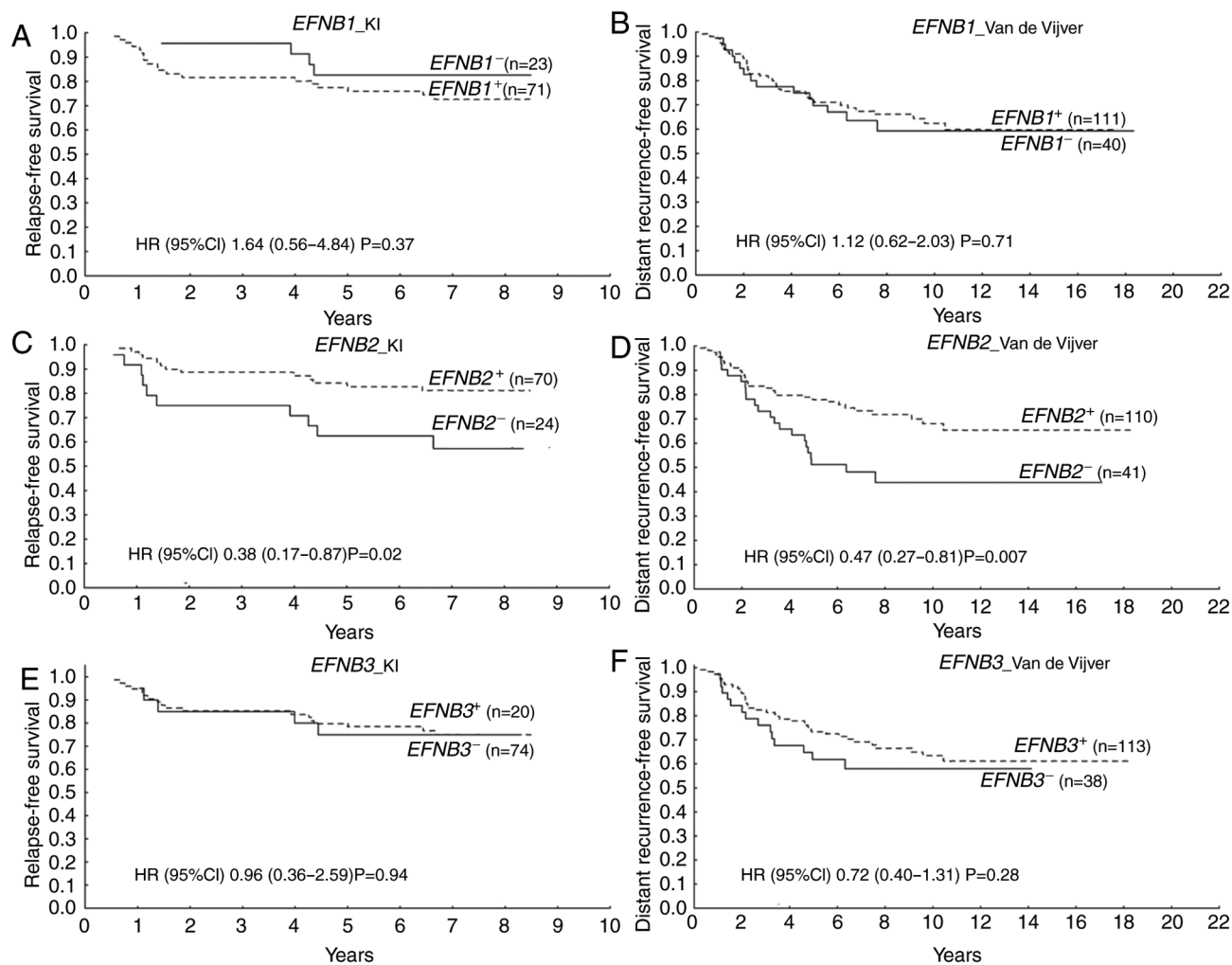


Table SI. Genes involved in cell motility with &gt;2 fold over- or underexpression.

B2-WT vs. GFP			B2-5F vs. GFP			B2-5F vs. B2-WT		
Genes overexpressed			Genes overexpressed			Genes overexpressed		
Gene symbol	Fold regulation	P-value	Gene symbol	Fold regulation	P-value	Gene symbol	Fold regulation	P-value
<i>CAPN1</i>	19.8688	0.031806						
<i>EGFR</i>	2.3314	0.000918						
<i>MET</i>	2.3437	0.000487						
<i>PLD1</i>	2.4879	0.000144						
<i>PTK2B</i>	2.8556	0.000511						
<b><i>SH3PXD2A</i></b>	4.0238	0.000022						
<b><i>STAT3</i></b>	2.4296	0.000215						
Genes underexpressed			Genes underexpressed			Genes underexpressed		
Gene symbol	Fold regulation	P-value	Gene symbol	Fold regulation	P-value	Gene symbol	Fold regulation	P-value
<i>CAV1</i>	-2.8521	0.002522	<i>CAV1</i>	-2.4593	0.016757	<i>ARF6</i>	-2.0973	0.022609
<i>DPP4</i>	-2.7914	0.013098	<i>CSF1</i>	-4.6255	0.000924	<i>ARHGDI1</i>	-2.0717	0.067095
<i>EGF</i>	-2.0786	0.001368	<i>EGF</i>	-2.2872	0.024226	<i>CSF1</i>	-2.4854	0.005364
<i>FGF2</i>	-2.0856	0.014434	<i>FGF2</i>	-2.4847	0.028613	<i>DIAPH1</i>	-2.0107	0.025514
<i>MMP9</i>	-2.0791	0.000169	<i>PLAUR</i>	-2.3749	0.002902	<i>ILK</i>	-2.3839	0.025836
<i>TGFb1</i>	-3.2158	0.001746	<i>RAC2</i>	-2.7364	0.040511	<i>MMP14</i>	-2.7125	0.017
<i>WASF1</i>	-2.6457	0.000046	<i>RDX</i>	-2.0914	0.003366	<i>PLAUR</i>	-2.5411	0.000201
			<i>TGFB1</i>	-3.2984	0.002634	<i>PRKCA</i>	-2.1633	0.020408
			<i>WIPF1</i>	-3.052	0.001231	<i>PTK2B</i>	-2.8333	0.003493
						<i>RND3</i>	-2.0062	0.003461
						<b><i>SH3PXD2A</i></b>	-6.3849	0.000952
						<b><i>STAT3</i></b>	-2.6849	0.019775
						<i>VASP</i>	-2.2249	0.004431

Genes validated by immunoblotting are shown in bold font.

Table SII. Spearman's rank order correlations.

Variables	Ephrin B	EphB4
Nodes	0,115	-0,013
NGH I	0,079	-0,034
NGH II	0,076	0,113
NGH III	<b>-0,162</b>	-0,097
Tumor size	-0,023	-0,025
ER	<b>0,181</b>	0,124
HER2	<b>-0,140</b>	0,033
Ephrin B		<b>0,162</b>
EphB4	<b>0,162</b>	

Significant correlations are marked in bold font and are significant at  $P < 0.05$ .

Table SIII. *EFNB2* mRNA expression has independent prognostic value in a multivariate Cox analysis (in bold font).

Variables	KI <sup>a</sup> n=94		Van de Vijver <sup>b</sup> n=151	
	H.R (95%CI)	P-value	H.R (95%CI)	P-value
<i>EFNB2</i>				
-	1.0		1.0	
+	0.32 (0.12-0.85)	<b>0.02</b>	0.40 (0.23-0.70)	<b>0.001</b>
<i>EPHB4</i>				
-	1.0		1.0	
+	0.92 (0.33-2.54)	0.87	1.46 (0.79-2.70)	0.23
ER				
-	1.0		1.0	
+	0.75 (0.29-1.99)	0.57	0.65 (0.34-1.22)	0.18
ERBB2				
-	1.0		1.0	
+	2.90 (1.03-7.87)	<b>0.04</b>	2.12 (1.08-4.17)	<b>0.03</b>
Systemic treatment				
-	1.0		1.0	
+	0.24 (0.09-0.62)	<b>0.003</b>	1.88 (0.74-4.80)	0.19

The endpoints analyzed were relapse-free survival (KI) and distant-recurrence-free survival (Van de Vijver). Only patients without lymph nodal infiltration were included. <sup>a</sup>Karolinska Institute dataset (22), <sup>b</sup>van de Vijver dataset (23).

Video S1. Migration assay showing the lateral movement of MCF7 cells infected with the GFP control vector. GFP-infected cells were able to fill the initial gap after 48 h by moving towards the opposite cell layer across the gap. Time-lapse of the cell's lateral migration was recorded every 5 min and the frames from the differential interference contrast (DIC) and the green fluorescence protein (GFP) recordings were merged using the software Image J. Scale bar included in the video corresponds to 100  $\mu\text{m}$ .

Video S2. Migration assay showing the lateral movement of MCF7 cells infected with the B2-WT vector. Infected cells were unable to fill the initial gap after 48 h. The time-lapse of the cell's lateral migration was recorded every 5 min and the frames from the differential interference contrast (DIC) and the green fluorescence protein (GFP) recordings were merged using the software Image J. Scale bar included in the video corresponds to 100  $\mu\text{m}$ .

Video S3. Migration assay showing the lateral movement of MCF7 cells infected with the B2-5F vector. B2-5F-infected cells were unable to fill the initial gap after 48 h and did not moved toward the opposite cell layer across the gap. Time-lapse of the cell's lateral migration was recorded every 5 min and the frames from the differential interference contrast (DIC) and the green fluorescence protein (GFP) recordings were merged using the software Image J. Scale bar included in the video corresponds to 100  $\mu\text{m}$ .